

DOUGH CHARACTERISTICS AND GLUTEN PROFILE OF VARIOUS WHEAT
BLENDS DERIVED FROM A DROUGHT AND A NON-DROUGHT RESISTANT
VARIETY

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BY

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DECLARATION

This thesis is my original work and has not been presented for award of another degree in any other University.

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ABSTRACT

Differences in drought resistance in bread wheat varieties and blending wheat with non-wheat flours are important in bread making because they affect dough characteristics and the baking qualities by influencing wheat gluten. Establishing whether differences exist in blending ratios in the production of acceptable bread between a drought and a non-drought resistant wheat variety was one aim of this study. Composite flours were prepared from blend of wheat with maize, cowpea, soy, sorghum, arrowroots, cassava, banana and sweetpotato. The protein contents, (9.36-15.52 %), the gluten contents, (7.9-10.08 %), and the loaf volumes (480-685 c.c) in Chozi and it's blends were significantly higher than those of Kwale and it's blends at 6.4-12.08 % (protein content), 3.86-6.3%, (gluten content), and 315-550 c.c, (loaf volume). Differences in the glutenin subunits were observed with Chozi revealing four high molecular weight glutenin subunits, (HMW-GS) and Kwale having five HMW-GS. Kwale had no differences between the unblended flour and flour blends with regard to the HMW-GS. The low molecular weight glutenin subunits (LMW-GS) profile showed differences between Chozi and Kwale, differences were also observed between unblended flours and some blends. HMW-GS from Chozi had the more enhanceive influence on the bread quality and the dough properties. The dough characteristics did not show major differences with most flours range being acceptable at 3-20 minutes (stability), 3-10 minute, dough development time, (DDT), 55-65%, (absorption) and tolerance (0-120 Brabender units, B.Us), for bread wheat varieties.

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ABBREVIATIONS AND ACRONYMS

AACC	American Association of Cereal Chemists
BU	Brabender Units
Carr	Chozi/arrowroots blend
Cbn	Chozi/banana blend
Ccp	Chozi/cowpeas blend
Cmz	Chozi/maize blend
Cpl	Chozi (unblended)
Csor	Chozi/sorghums blend
Csoy	Chozi/soy beans blend
Csp	Chozi/sweet potato blend
FAO	Food and Agriculture Organization
HMW-GS	High molecular weight glutenin subunit(s)
KARI	Kenya Agricultural Research Institute
Karr	Kwale/ arrowroot blend
Kbn	Kwale/banana blend
Kcp	Kwale/cowpeas blend
Kmz	Kwale /maize blend
Kpl	Kwale (unblended)
Ksor	Kwale/sorghums blend
Ksoy	Kwale/soy beans blend
Ksp	Kwale/sweet potato blend
Ksoy	Kwale/ soybean
LWM-GS	Low molecular weight glutenin subunit(s)
NPBRC	National Plant Breeding Research Centre
PAGE	Poly acrylamide gel electrophoresis
SDS	Sodium dodecyl sulphate
TEMED	N, N, N', N- Tetra methyl ethylene diamine

DEFINITIONS OF TERMS

- F₂** A term used in breeding describing the second generation of progeny from genetically different parents.
- Gliadins** Proteins present in the endosperms of wheat characterised structurally by intra-molecular disulphide bonds and which in flour when mixed with water confer extensibility to dough.
- Glutelins** Proteins present in the endosperms of many cereals and other grains as storage proteins and which are structurally similar to the glutenins in wheat though not functionally similar.
- Glutenins** Proteins present in the endosperms of wheat characterised structurally by intermolecular disulphide bonds and which in flour when mixed with water confer resistance to the dough during mixing. They are considered the most important in bread making because of their influence on the quality and acceptability in the attributes.
- Prolamins** Proteins present in the endosperms of many cereals and other grains as storage proteins and which are structurally similar to the gliadins in wheat. Also used to refer to both gliadin-like proteins stored in the endosperm of seed of the plants in the Graminae family.

CHAPTER ONE

1.0 INTRODUCTION

1.1 Background

The Eastern and Southern Africa regions are prone to drought. Steps to fight its effects through yield improvement and yield stability under drought conditions are in progress. Chozi was introduced from Mexico for trials, further selection and breeding in countries with environmental conditions which were different from Mexico using F₂ generation. Yield trials in breeding from 1996 to 1999 on this variety were performed in the dry areas of Kenya leading to its release for commercial production as a bread wheat variety (Kinyua *et al*, 2000). Drought resistant (DR) wheat varieties are desirable in the country because they can be grown on a larger acreage. This would increase the wheat production and offset wheat shortages. Blending of wheat flours with non-wheat flours can also offset shortages. Blending with other cereals flours will save costs since these are cheaper. Blending with non-cereal flours has an added advantage since, root tuber flours for example, can enhance the nutritional quality. Sweet potatoes have high levels of β -carotenes as well as lysine (Carpio 1984). These advantages can be achieved if wheat flours are blended commercially with other non-wheat flours such as oilseed flours, legumes and fruits based flours.

Two important factors in baking bread include the protein content and the protein quality. There are two aspects to protein quality in wheat flour namely the gliadin / glutenin proportion and the type of high molecular weight glutenin subunits (HMW-GS) present in flour. The protein quality of wheat flour is heritable; a high quality variety will produce good bread over a range of protein percentages whereas a low quality variety will produce relatively poor quality bread even when the protein content is high. The relative proportions of HMW (glutenins) and low molecular weight glutenin subunits (LMW) and gliadins are a major factor in bread baking.

1.2 Statement of the problem

Kenya produces only 50 % of the wheat consumed annually; the rest is met through imports. The low level of production is partly because the country is over 75 % semi-arid. To meet the difference the country is developing DR wheat varieties and investigating the use of non-wheat flours in blending with wheat flours. In the development of DR wheat varieties and preparation of non-wheat-based composites, properties may be altered, hence the need to analyse the dough characteristics, sensory scores and the gluten profile of

composite flours derived from a DR wheat variety (Chozi) and a non-drought resistant (NDR) wheat variety (Kwale). Differences in drought resistance between bread wheat varieties and blending wheat with non-wheat flours are important in bread making. They affect dough characteristics and the baking qualities by influencing wheat gluten. Researching these factors so as to allow for the growing of wheat on a larger acreage (drought resistant varieties) and the blending (in industry) have been identified as ways to help meet rising wheat consumption in Kenya. Establishing whether differences exists in blending ratios in the production of acceptable bread between a drought and a non-drought resistant wheat variety is important in the promotion of use of non-wheat flours to the different ecological areas of the country. Differences in acceptability of loaves blended at the same level but with flours from the two wheat varieties indicated relationships between drought resistance and gluten quality.

1.3 Justification

There is an increasing demand of wheat due to the rising popularity of wheat-based foods. The country is currently producing only 50 % of the annual demand of 600 000 metric tons. The rest is being met through imports, which drains the foreign exchange earnings. Use of non-wheat flours for blending wheat flours to produce composite wheat flours may meet this shortfall. It would also, depending on the source of the blending flour, improve the nutritive value of the many wheat-based foods. Drought tolerant wheat varieties may also offset the shortfall since they can be grown on a larger acreage. Due to this it was important to investigate the effects of non-wheat flours in bread baking quality within and between a NDR and a DR wheat variety.

1.4 Objectives

- 1) Comparison of the dough characteristics and baking quality of the wheat flour composites from the two varieties, Chozi (DR) and Kwale (NDR).
- 2) Determining the protein (gluten) profiles of the flours.
- 3) Analysing the contribution of the non-wheat flour to the dough characteristics.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Introduction

Chozi is a new drought resistant (DR) wheat variety just released for commercial production in the year 2000. Kenya Agricultural Research Institute (KARI) classifies Kwale wheat variety in group 3 with wheat flours which give strong distensible dough and have good bread baking qualities, (Arama *et al*, 1997). Previous studies have led to the identification of other drought resistant wheat varieties (Kinyua *et al*, 2000).

Breeding of crops that are drought resistant is being emphasised as the more customary food crops continue to fail due to poor adaptability. Many DR varieties have a lower net yield per hectare compared to the non-drought resistant (NDR) varieties, for example Chozi, a DR variety produces 0.5-2.5t/ha, (Kinyua *et al*, 2000), compared to the Mbuni variety a NDR which produces on average 3.9t/ha. DR varieties are still attractive as they produce yields in areas where the NDR varieties cannot grow for example, Katumani, Elementaita, Lanet and Mogotio, areas which generally do not support NDR varieties (Kinyua *et al*, 2000).

Composite flour in food science is a major research area. These studies in most countries are aimed at reducing imports of wheat and improving the nutritive value of the wheat-based products. Some countries have embarked on courses designed to maximise the use of locally available resources to replace partially or wholly the use of wheat in products that traditionally have used wheat flours (Olugbemi, 1991). International bodies such as the Food and Agriculture Organization (FAO) are also promoting the production of wheat-free flours and blended wheat flours for baking of breads and other traditional wheat products to replace large proportions of wheat in wheat-based foods (FAO, 1979). The Kenyan strategy is to improve the wheat production capacity and to encourage the use of wheat composites rather than "all wheat flours" in wheat based foods consumed in the country.

Methods used in grouping wheat categories include the dough characteristics and the Zeleny test (Axford *et al*, 1978), both of which analyses the baking quality of wheat varieties. The SDS-sedimentation data has been found to be generally consistent with the volumes of baked loaves except in a few cases (Blackman and Payne, 1987).

Gliadins and glutenins (and other prolamines in the grass family) have a high level of heterogeneity determined strictly by the genotype. Gluten genes are tightly associated and inherited as blocks with several allelic forms at each locus. Genetic linkage has been

identified to certain genes though as yet no linkage has been identified between the osmoregulation gene and the storage proteins genes. The type of HMW-GS in wheat influences the baking quality. The protein quality is primarily genetically determined but may also be affected by growth conditions after anthesis (Huebner *et al*, 1990). Water stress during grain filling reduces starch storage more than that of protein (Lipsett, 1963), but baking quality is also affected even when the protein content is not because different protein fractions do not develop simultaneously (Huebner *et al*, 1990).

2.2 Wheat gluten proteins and drought stress in a DR and a NDR wheat variety.

Plants with good drought resistance often also have higher heat resistance (Levitt, 1972). Wheat is classified as a moderately drought tolerant species. Considerable varietal differences exist within different wheat varieties. Heat stress occurs accompanied with drought in many countries in the tropics and impairs many physiological processes. The genetic resistance to the key abiotic stress involved namely heat and drought in non-traditional wheat growing areas is not clearly defined. Drought resistance is osmoregulation irrespective of the source of the stress (like lack of water, high temperatures, alkalinity or salinity Edmeades *et al*, 1992). In Kenya, this stress is experienced as either lack of water or high temperatures though it is usually both because the country lies within the ME5B (mega-environment 5B) region, classified as hot and dry with temperatures of more than 22.5 °C, which are higher than, expected for wheat growing.

Wheat plant respond to heat stress by producing the heat shock proteins, which includes gliadins where multiple heat shock elements are present upstream of coding regions. Studies have shown changes in the normal balance of gluten polypeptides immediately after heat shock as well as in the mature grain (Blumenthal *et al*, 1994). The relative proportions of glutenins and gliadins are a major factor in the bread making character of flour. Gluten proteins develop 10-15 days after anthesis and are produced at different times. Water stress affects the period of gliadin production rather than the rate of production, while the glutenins ratio is decreased in water stress (Agenbag and DeVillers, 1995). High levels of gliadins in dough lead to bread of lower loaf volumes. Water stress during the first three weeks of grain filling affect the protein content, the protein composition and the bread volume in wheat by decreasing the production of reserve proteins. Drought resistant wheat varieties more efficiently metabolize proteins while the non-drought resistant varieties stop or reduce the rate of synthesis of glutenins affecting

their gliadin/glutenin ratio (Blumenthal *et al*, 1994). A decrease in glutenin was noted for plants experiencing heat (temperature $>30^{\circ}\text{C}$) during the first 14 days after anthesis. This effect on the dough properties was noted as an increasing in the dough development time and a more rapid dough breakdown as well as an increasing in resistance to mixing, R_{max} . The rate of increase of R_{max} with the increase in temperature was greater for cultivars with the *Glu-1Da* allele than those with the *Glu-1Dd* suggesting that the relative performance of cultivars with different alleles at this locus depend on the environment and the specific gene (Panozzo and Eagles, 2000).

Good and poor wheat of bread making quality is associated with two allelic pairs at the *Glu-1D* complex locus, designated as *1Dx5-1Dy 10* and *1Dx2-1Dy12*, respectively (Ahmad, 2000). Chromosomal location of genes influence baking qualities and resistance to stress in bread wheat are being studied. Studies have shown that the short arms of chromosome *6A* and *4D* might carry the gene heat suppressor *Delda*. Chromosome *1D* might carry genes that decrease the rate of water loss (Ehdaie and Waines, 1997). In a study of *Triticeae spp* dehydrins, the chromosomal arm *6AL* of Chinese spring wheat that also has certain gluten genes was one of the loci to which dehydrins were assigned. Putative regulatory factors of dehydrins genes, *dhn* have been located within the vicinity of 5B in wheat, another *dhn* genes loci. The relationship between the *dhn* genes, their regulatory factors and their linkage to proteins affecting the baking quality and dehydration is being studied (Werner and Close, 1998).

2.3 Flours

2.3.1 Flours: Blending

In cereal flours substituting at a level of 80:20 for wheat/maize and 90:10 for wheat/sorghum produces satisfactory bread (Keregero and Mtebe, 1994). Iwuoha *et al*, (1997), found cowpea flour (*Vigna unguiculata*) a grain legume crop is acceptable when substituted at 10 % replacement level with wheat. Ten- percent soyflour has been used to produce an acceptable white bread. Up to twelve- percent soyflour can replace wheat flour if emulsifiers such as sodium stearyl-2-lactylate are incorporated in the dough (Wolf and Cowan, 1971). Composite breads fortified with legume flours in different proportions were found to improve the physical, chemical and sensory qualities. The two legume flours affect the chemical and physical quality of composite flour breads differently. The volumes of cowpea / wheat flour composite bread samples (CBSs) were higher than those using soy/wheat CBSs as well as all wheat loaves (Iwuoha *et al*, 1997).

The poor acceptability of soy containing CBSs relative to those containing cowpea flour was attributed to the strong beany flavour and its poor cooking quality. Treatment with hydrogen peroxide or calcium chloride modifies the flavour (FAO, 1979). Soy flour however is popular in many countries as a rich protein source. It's especially rich in lysine, an amino acid usually deficient in wheat and other cereal flours except in certain lysine-rich maize genotypes (Klein *et al*, 1980). It facilitates greater water incorporation, improves dough handling and machineability when mixing, prolongs the freshness and storage stability of breads and improves the crust colour development (especially raw soy flour), moisture retention and the bleaching of bread (Raidl and Klein, 1983).

In a study to establish an acceptable level of wheat flour replacement with cassava flour, a root tuber crop, twenty percent replacement level gave acceptable bread (Omune *et al*, 2001).

In wheat / sweet potato blends, the ratios of 70:30, gave acceptable bread if the sweet potato variety has a low trypsin inhibitors concentration. Dough improvers increase the loaf volume in sweet potato / wheat bread flour if prepared at a ratio of 20:80 (Woolfe, 1992). Most studies have found a ten to fifteen percent wheat substitution level gives the most acceptable bread based products.

A 90:10 ratio of wheat / plantain has given acceptable bread (Bamidele *et al*, 1990). Bread with fifteen-percent tamarind seed powder in wheat flour was acceptable (Bhattacharya *et al*, 1994).

2.3.2 Flour: Components

2.3.2.1 Flour proteins

Flour composition and functionality vary greatly depending upon the milled wheats' heredity and the environmental conditions of its culture and harvest (Inglett, 1974). The gluten proteins are the most important in bread making. Bread wheat varieties have 12-14% protein content. There are two main proteins namely the gliadins (prolamins) and glutenins (glutenins) where glutelins is the general type of describing glutelin like proteins in wheat and other cereals whereas prolamins describes gliadin like proteins in wheat and other cereals. The prolamins, rich in glutamine and proline, are monomeric polypeptides stabilized by intramolecular disulphide bridges while disulphide bridges intermolecularly join glutelins. Bread wheat varieties may have up to 40 gliadin subunits that range in size from 30-40 kilodaltons. The glutenins vary in size from 40-20000 kilodaltons. Even though both gliadins and glutenins have a third glutamine and a tenth each of proline and

glycine they differ not just in size but also in solubility with gliadins being soluble in alcohols while glutenins are only soluble in weak acids and bases. Gliadins are viscous liquids with extensible properties while glutenins are cohesive elastic solids. Structurally, the presence of numerous repetitive β -turns structures as the central domain with α -helices at the nitrogen (N-terminals) and carboxyl (C-terminals) are responsible for elasticity. The glutenins and gliadins in the presence of an appropriate amount of water and with agitation form tough elastic complex, gluten that retains gases making a leavened product possible because the protein hydrates and associates with specific carbohydrates and lipids to form this gluten product (Bushuk, 1985).

Some common wheat varieties have up to 17 glutenin sub-units of differing molecular weights. In total, 22 glutenin subunits have been characterised from different wheat varieties. These have broadly been classified as either of the low molecular weights (LMW), (Jackson *et al*, 1983) or of the high molecular weights glutenin subunits (HMW-GS), (Payne *et al*, 1981). Bread wheat cultivars each have 3 to 5 subunits of HMW-GS. The genes of these glutenin subunits are allelic. The HMW-GS are vital to the bread making quality of wheat which has been correlated strongly with the proportion and type of glutenin proteins present in HMW-GS. Genetic studies have strongly correlated specific HMW-GS to good and poor bread making quality. Some subunits are more effective than others in conferring the bread making quality (Manley *et al*, 1992).

Analysis of storage proteins by electrophoretic techniques has been shown to be independent of site, year, and generation of seed production (Marchylo and LaBerge, 1980). PAGE of storage proteins has been used around the World to identify wheat cultivars (Shewry *et al*, 1988). The proportion of glutenin in flour protein is highly dependent on cultivar whereas although cultivar is still important, environmental variation is greater than cultivar variation for gliadin.

Across environments, the proportion of gliadin increases with the increase in flour proteins whereas the proportion of glutenin decreases (Panozzo and Eagles, 2000).

Sorghum as most cereals is low in lysine, threonine and tryptophan but has high levels of prolamine and leucine. Prolamins influence the dough characteristics of blended flour. Prolamins in the grass family of plants include zein in maize and hordein in barley, which are gliadin-like in properties. The prolamins in rice, sorghum and oats are glutenins-like (Pomeranz and Shellenberger, 1975). The gliadin / glutelin ratio in wheat and sorghum is equal while the ratio in maize and barley is higher for prolamins. Close evolutionary

relationship between prolamin of wheat, rye, barley has been noted, as is the relationship of maize, sorghum and millet to each other.

Grain legumes have high levels of protein, up to forty percent of which globulins are the highest followed by glutelins and albumins. In cowpea the abundance of proteins in the grain is of the order globulins > albumins > glutelins >prolamins. The glutelin and prolamin fractions of cowpea grain have high levels of essential amino acids.

2.3.2.2 Flour carbohydrates

Starch the predominant carbohydrate in wheat is vital to water absorption furnishing a suitable surface for the strong adhesion of gluten thus improving the dough rheology. During baking the starch held in granules imbibes water, swells and gelatinizes. This water previously held by gluten in a film is dehydrated from the protein aggregate causing the gluten to set. The non-starch carbohydrate including cellulose, hemi-cellulose, pectin and lignin form the fibrous content of wheat flour, their increase in flour has been correlated to an increase in the tenderness and the moistness of a baked product (Jeltema *et al*, 1983). High diastatic activities in cassava flours affect baking since levels above 145 mg of maltose are deleterious (Eggleston *et al*, 1993). Starch is the carbohydrate in most pulse legume seeds but is low in oil seeds. Flatulence factors including raffinose, present in sweet potato and stachyose and verboscose in soy are carbohydrateic (Garcia and Palmer, 1980).

2.3.2.3 Other compounds present in flours

Wheat contains carotenoids especially the xanthophylls, which have no vitamin A activity, in contrast the carotenoids of sweet potatoes and plantains are converted by the body to vitamin A. Germs of wheat have vitamin E and other tocopherols. Thiamin, riboflavin, niacin and pyridoxal are present in large quantities while pantothenate and folate are in low amounts (Pomeranz and Shellenberger, 1975).

Lipids make up to 2 % of the flour weight in wheat, mainly from the residue quantities of germ left during the preparation and milling. Ponte and Destefanis, (1969) found that free polar lipids have a favourable effect on the baking while non-polar lipids are detrimental.

Sorghum contains tannins that have an antinutritional effect, this effect is eliminated during the various steps of the preparative process. Raw soy flours have been used in colour control as bleaching agents in white breads. The lipooxygenases in soyflours oxidise

polyunsaturated fats and these fats presumably bleach the wheat flour carotenoids to a colourless form giving breads a whiter crumb, crust colour is also enhanced (Wolf and Cowan, 1971).

2.4 Physiochemical properties

Physical dough testing is carried out to evaluate bread making potentialities (strength) and performance characteristics of flours under mechanised conditions (Pomeranz and Shellenberger, 1975). Flour used in bread making is milled to standard specifications. Grain has to be tempered to 15 % moisture content. In white flour milling for strong wheat varieties used in bread making the flour yield is 70-75% of total grain weight. The flour physical dough testing involves the use of recording dough mixers mainly the farinographs and the mixographs. These record the power needed to mix dough at a constant speed. The record consists of an initial rising part that shows an increase in resistance with mixing time. In farinography, the plasticity and mobility of unyeasted dough subject to a prolonged relatively gentle mixing action at constant temperature is measured. 50 grams of flour and water are placed in mixing bowl with mixing blades running at 63 revolutions per minute for a period of time, usually 15 minutes. Resistance offered by the dough to mixing blades is transmitted through a dynamometer to a pen that traces a curve (graph) on a moving paper of special design (Chamberlain, 1995). This curve is referred to as a farinogram and is traced along the 500 Brabender units line (B.U). The general practice is to determine the standard absorption by titration curve with water added from a burette to the flour as it's mixed. The point of maximum resistance (minimum mobility) is the optimum dough development time that is followed by a second part of more or less rapid decrease in consistency and resistance to mixing, characterised by the production of sticky, more extensible, less elastic dough on further mixing, termed dough breakdown.

Generally, the standard absorption is the amount of water needed for an optimum consistency of 500BU. Absorption increases with increase in the protein content and as the gluten content quality improves, in wheat (Blackman and Payne, 1987). From the farinogram various parameters that are used to determine dough strength and therefore the flour quality are read. The reading of these parameters is described below.

- a) Dough development time (DDT) is the time to the nearest 0.5 minutes from the time of the first addition of water to the point of maximum consistency range immediately before the first indication of weakening, also termed "peak" or "peak time" (PT).

- a) Tolerance is the difference between the BUs from the top of the curve at the peak to the top of the curve measured 5 minutes after the peak is reduced, read in Brabender units.
- b) Stability is the time difference (at the nearest 0.5 minutes) between the top of the curve where the curve first intersects the 500 BU-line (arrival) and the point where the top of the curve first leaves the 500 BU-line (departure line).

Baking quality is assessed in loaves after the baking. This mostly involves the determining of the loaf volume and the sensory scoring. In a standardized baking test the ingredients, time-temperature regime are controlled so the only varying quality is the flour quality. In rapeseed displacement method in the determination of the loaf volume, loaf volumes below the standard wood volume are considered unacceptable. Other methods used to determine acceptable loaf volume include the determination of the specific gravity of bread. The weight of the bread is determined 10 minutes after removal from the oven. Specific gravity values below 3.5 grams per millilitre are considered as producing unacceptable bread. The values in Table 2.1 shows the characteristics of wheat required for bread from fermented dough.

Table 2.1 Dough characteristics in fermented dough preparation

Characteristic	Description of parameter
Preferred endosperm texture	Hard
Flour strength	Very strong to medium strong
Protein content	11-12 %
Damaged starch	25-30 Farrands
Total α -amylase	7-12 Farrands
Farinography stability	4-7 minutes
Dough development time	3-6 minutes
Water absorption	57-60%

CHAPTER THREE

3.0 MATERIALS AND METHODOLOGY

3.1 Materials, Preparation and blending ratios of flours

Table 3.1 Raw material, preparation and blending ratios of flours

Material	Preparation of flours	Blending, wheat: Non-wheat	
		Kwale	Chozi
Wheat (<i>Triticum aestivum</i>)	Milling 100 grams in one-minute grains tempered to fifteen percent moisture content, American Association of Cereal Chemists, (AACC, 1993).	100:0	100:0
Maize (<i>Zea mays</i>)	Maize grains were finely ground using a micro Wiley mill and sieved using a 60-mesh sieve (Othira, 2000).	80:20	80:20
		(Keregero and Mtebe, 1994)	
Sorghum (<i>Sorghum bicolor</i>)	..	85:15	85:15
		(Keregero and Mtebe, 1994)	
Cassava (<i>Manihot esculenta</i>)	Cassava was washed, peeled and chopped to small pieces followed by anaerobic fermentation for 6 days in plastic containers (Onjoro <i>et al</i> , 1996). Cassava was then dried for 7 days on concrete floor and ground to 60-mesh flour.	80:20	80:20
		(Omune <i>et al</i> , 2001)	
Sweet potato (<i>Ipomea batatas</i>)	The tubers were washed, peeled, sliced and soaked (20 % w/v) in water. Sodium metabisulphite solution was added for 2 minutes. These were then sun dried till <10% m.c and finally ground to a 60-mesh fine flour, (Sakamoto and Bouwkamp, 1985).	85:15	85:15
		(Woolfe, 1992)	
Arrowroots (<i>Maranta arudinacea</i>)	..	85:15	85:15
		(Othira, 2000)	
Soybean (<i>Glycine max</i>)	Soybeans in a perforated basket 2/3 rd s full were soaked for 6 hours in fresh water at 25 ^o C containing baking soda at a rate of 1-kg baking soda for 100-kg beans. These were cooked for 10 minutes followed by air drying for 30-36 hours then cracking to remove hulls and grinding using a micro-Wiley. (United State Department of Agriculture (USDA, 1967)	95:5	95:5
		(Iwuoha <i>et al</i> , 1997)	
Cow pea (<i>Vigna sinensis</i>)	The cowpeas were first soaked in distilled water (the ratio of water is 1:5 w/v) pea to water (21 ^o C). The coat then manually removed followed by freeze dried along with soak water and finally ground to a 60 mesh sieve flour in a micro Wiley mill (Despande <i>et al</i> , 1982).	90:10	90:10
		(Iwuoha <i>et al</i> , 1997)	
Banana (<i>Musa spp</i>)	Green plantains were blanched at 82-93 ^o C/ 5 minutes. The fruit was then peeled, sliced and sun dried or till the moisture is <10% and then ground to fine 60-mesh flour (Bamidele <i>et al</i> , 1990).	90:10	90:10
		(Bamidele <i>et al</i> , 1990)	

Table 3.1 shows the blends, the preparation and the blending ratios at maximum recommended replacement levels for acceptable bread in wheat for Kwale and Chozi. The blending ratios were used without any variation (as shown in the Table 3.1) for the protein profile analysis. The protein quality analysis was conducted at the department of Biochemistry and Molecular biology laboratories. The dough characteristics, the baking quality and the sensory evaluation were carried out at the National Plant Breeding Research Centre (NPBRC), Njoro.

3.2 Experimental procedures

3.2.1 Protein properties analysis

3.2.1.1 Gluten content

25 grams of each sample from Table 3.1 was manually washed using tap water according to the AACC, 1993 method. Clean tap water (15-20 mls) was added to the flour sample in a mortar and worked by pestle making sure no material adhered to mortar. The dough was then left to stand in water at room temperature for 15 minutes, kneaded gently under a stream of water till all starch and water-soluble components of flour were removed. Starch free gluten was then pressed as dry as possible between hands and left to stand for 1 hour before weighing. The percentage gluten was calculated as

$$\% \text{ Gluten} = \frac{\text{Weight of dry gluten} \times 100}{\text{Weight of flour sample.}}$$

3.2.1.2 Protein concentration (Modified Folin- Lowry, Holme and Peck, 1998).

To 0.5 grams of each sample powder prepared (Table 3.1), 10 mls of 50 mM Tris-HCl buffer, pH 8.0, containing 3 % Triton X-100 was added and the samples homogenized. The homogenate was then centrifuged at 4000 r.p.m. for 30 minutes and the proteins precipitated from 2 mls of supernatant with 4 mls of 10 % TCA. The protein was then pelleted by centrifugation and then hydrolyzed in 2 mls of 1 M NaOH at 37⁰ C overnight. 0.6 mls of distilled water and 6 mls of a reagent A (described below) were added to 0.4 mls of the hydrolysate. The mixtures were kept at 37⁰ C for 10 minutes. 0.5 mls of Folin-phenol reagent diluted from the stock Folin-phenol at a ratio of 1:1 was then added. The mixtures were then kept for 30 minutes at room temperature for the blue colour to develop. The intensity of the blue colour was measured spectrophotometrically at 660 nm and then expressed as standard protein (Bovine serum albumin, BSA) in µg/ml. This

involved the measuring of the absorbance of different protein concentrations varying from 0-150 µg/ml BSA. A protein standard curve was drawn with the absorbance on the vertical ordinate and the concentration of the BSA on the horizontal ordinate. The measured absorbance of different blends of Chozi and Kwale were then read from the standard protein curve. The different absorbances were converted to protein concentrations and then multiplied by a 100 % to obtain percent protein content.

Reagent A consisted of: 2 % Sodium bicarbonate (NaHCO_3) in 100 mls distilled water (solution 1) and 0.5 percent $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ in 1 % Sodium-potassium tartarate (solution 2). 50 mls of solution 1, was mixed with 1 ml of solution 2 following the method of Holme and Peck, (1998).

3.2.1.3 Protein profile analysis

Extraction of storage proteins in the two wheat flours was done using Singh *et al*, (1991) procedures to obtain the HMW-GS and LMW-GS. Gliadins were extracted first, by heating 0.2 grams of the crushed powder at 60 °C in a water bath for 1 hour in 300 µL of 70 % aqueous ethanol. 1 ml 50 % n-propanol added followed by vortexing briefly and incubating at 60 °C in a water bath for 30 minutes washed the residue in each eppendorf tube. This step was carried out two times. Centrifugation at 10000 r.p.m. / 2 minutes and sucking off of all n-propanol followed. The HMW-and LMW-GS was extracted from the gliadin free residue by incubating in a 60 °C water bath in 120 µL of extraction buffer (50 % n-propanol in 0.08 M Tris-HCl containing freshly added 1.25 % (w / v) dithiothreitol). After a brief initial vortexing, samples were again incubated in extraction buffer containing 0.17 % 4-vinyl-pyridine. After centrifugation, the supernatant was collected in a new tube and finally mixed with an equal volume of sample loading buffer (0.08 M Tris-HCl, (pH 8.0), 20v % glycerol, 1.6 % (w / v) SDS and 0.016 % (w / v) bromophenol blue. Polyacrylamide gel electrophoresis method described by Maartens *et al*, (1999), was performed on a vertical slab gel electrophoretic unit. 30 % acrylamide (29.2g) and 2.65 % Bis-acrylamide (0.8 g) was dissolved in a total volume of 100 mls of water. The composition of the separating and the stacking gels are shown in Table 3.2.

The buffers used were prepared according to the method of Laemmli, (1970). Table 3.3 shows the buffers' preparation for polyacrylamide gel electrophoresis (PAGE). The reservoir buffer was at a pH of 8.3 .The sample buffers were mixed with the samples and loaded on to the gel. The power settings were a constant 10mA for 18 hours.

Table 3.2 Electrophoresis buffer preparation

Name of buffer	Component	Weight in grams	Final volume of buffers for stock solution
Separating gel buffer pH 8.3, 3.0 M	Tris (dissolved in water and pH adjusted with 1 M HCl)	36.3 grams 48 mls	100 mls
Stacking gel buffer pH 6.8, 0.5 M	Tris (dissolved in water and pH adjusted with 1 M HCl)	6 grams	100 mls
Tank buffer pH 8.3 (diluted 4 times to use)	Tris Glycine SDS (10%)	30.3 grams 144 grams 10 grams in 100 mls water	1000 mls
Sample buffer pH 8.0, (0.08 M)	Tris (dissolved in water and pH adjusted with HCl) Glycerol (20% w/v) SDS (1.6%) Bromophenol blue (0.016 % w/v)	0.8 grams 20 mls 1.6 grams 1.6 milligrams	100 mls

After the electrophoretic run the gels were stained following the procedure of Wrigley, (1992). Gels were immersed for at least 4 hours in a fixing solution (acetic acid, methanol and distilled water at 1:4:5 ratio). The gels were stained overnight with a solution of 0.58 % (w / v) Coomassie Brilliant Blue G250 in a 14 % (w / v) TCA containing 5 % methanol and 200 mls distilled water. Destaining was carried out using the 14 % (w / v) TCA containing 5 % methanol and 200 mls distilled water. The acid was then rinsed 5 times using 500-ml portions of distilled water. After destaining the banding pattern, was then, photographed.

Table 3.3 Composition of gels

Constituent	Separating gel at 10%)	Stacking gel at (5.7%)
30% acrylamide solution	14.3 ml	6.0 ml
0.05M Tris-HCl	10ml (pH8.8)	3.0ml (pH8.8)
Water	16.2 ml	6.8 ml
TEMED	0.02 ml	0.05 ml
10 % Ammonium per sulphate	0.1 ml	0.05 ml
Total	40.62 ml	15.9 ml

3.2.2 Dough characteristics

Dry moisture Content was determined by weighing 10 grams of flour and placing in a moisture meter followed by heating at 130 °C for 3 hours. The moisture content. was then read from the dial to the nearest 0.1 scale division. (Nutritional lab manual, 1995).

3.2.2.1 Farinograph test

Farinograph tests were carried out at the NPBRC according to constant flour weight (CFWP) method. (AACC, 1995). The thermostat and the circulating pump were switched on one hour before use, at 30 °C.

The amount of flour to be used was calculated as:
$$\frac{86 \times 50 \text{ grams}}{(100 - \text{FMC})}$$

Where FMC is the flour moisture content;

86 g is Flour weight – standard m.c. (100-14)

50 g is the average weight of the composite flour used.

These quantities were placed in a farinograph bowl and the machine turned on. Water was then added from the burette and the machine ran till an adequate curve centred at the 500BU was achieved. The water absorption value was calculated at 14 % m.c. using the equation,

$$\text{Absorption (\%)} = 2(x + y - 50)$$

Where x is the amount of water that produces the curve within the maximum consistency centred on the 500 BU line.

y is the grams of flour used calculated on 14% moisture basis.

3.2.3 Baking tests:

The AACC, 1993 pup loaf method was used for loaf preparation by making the following:

Preparation of the yeast: - 1.2 g was added to 25ml-distilled water heated to 40 °C.

Salt preparation: - non-iodised salt was added at the rate of 1.0-g salt / 25 ml distilled water.

Sugar preparation: - 2.5 grams of sugar was added to the water also containing 1.0 g salt.

Oxidising agent: - 1.5 grams of potassium bromate (KBrO₃) and 100 grams of ammonium dihydrogen phosphate was added to water to make it 1000-ml solution in a volumetric flask. one millilitre of the prepared oxidising agent was added during the dough preparation.

Malt solution preparation: - 3.5 grams fungal enzyme (malt) in 100 ml distilled water.

One millilitre of the prepared oxidising agent was added during the dough preparation.

Shortening: - 3.0 grams were used per loaf (Kimbo®).

Milk powder: - 3.0 grams were used per loaf (Miksi®).

The amount of water to be added to the solution was obtained at the farinograph absorption. All solids were placed in the mixing bowl, followed by the addition of 25 mls of the salt / sugar solution, the required amount of water as per the Farinograph titration, 25 mls of yeast suspension into the ingredients in the mixing bowl. Also added is one millilitre each of the oxidising agents and malt solutions preparations. These were mixed to achieve maximum consistency and finally removed from the mixer and the dough proofed. Punching, moulding and baking. - after 105 minutes, the doughs were punched through 9 /32" and returned to the fermentation / proofing cabinet, then punched again, through the same setting after a further 50 minutes. They were then fermented for a further 25 minutes and moulded after punching through 9 /32", placed in a greased, labelled baking tin and proofed for 55 minutes at 30 °C and 85 % relative humidity. The doughs were baked at 425 °C / 25 minutes with the oven previously turned on 1 hour before baking containing some water in it. Scoring the loaf volume followed 10 minutes after removal from the oven and after weighing the loaf.

3.2.3.1 Loaf volume

It was determined by the rapeseed displacement method (AACC, 1993).

- I. Volume of standard wood = 400 cc,
- II. Volume of wood +rapeseed = 565 cc,
- III. Volume of rapeseed = 165 cc
- IV. Volume of bread +rapeseed = x cc
- V. Volume of bread = x -165 cc

3.2.3.2 Sensory (organoleptic) evaluation

Descriptive testing was used; a 10 people panel rated the breads after a brief sensory training. Bread was served at random mid-morning and mid-afternoon for scoring. The descriptions of rating the composite bread samples followed the method of (Haglund, and Johansson, 1998).

3.3 Statistical Analysis

The statistical significance test of a treatment was tested using Duncan's multiple range tests (DMRT) at $p < 0.05$. This was done to extract further information from the data after the analysis of variance (ANOVA) calculation. The treatment in this investigation was the blending of wheat with non-wheat flours. There were eight non-wheat flours derived from maize, sorghum, cassava, sweetpotato, soybean, arrowroots, cowpea and banana representing the different factors being investigated so as to analyse the effect of the blending treatment. The two wheat varieties were considered in this investigation to represent statistical blocks, namely the block for Chozi and its blends (representing the drought resistant statistical block) and Kwale and its blends (representing the non-drought resistant bread wheat variety block) of statistical comparison. The research design is illustrated in Table 3.3 below. Simple correlations between all the tests were calculated where $p \leq 0.05$ was significant. The completely randomised block design used in this investigation was derived from Gomez and Gomez, (1984).

Table 3.4 The completely randomised block design for Chozi, Kwale and their blends.

Treatment (T)	Block 1 {Chozi}			Block 2 {Kwale}		
	Replications (R)			Replications (R)		
	R1	R2	R3	R1	R2	R3
T1	1	6	7	1	6	7
T2	2	3	9	2	3	9
T3	7	8	6	7	8	6
T4	4	5	1	4	5	1
T5	9	2	8	9	2	8
T6	5	4	3	5	4	3
T7	6	1	2	6	1	2
T8	3	7	5	3	7	5
T9	8	9	4	8	9	4

Key:

T represents the different flour blends entered in the table as number 1 to 9 representing the different blends tested for different properties in a randomised research design.

R represents the number of replications (three in this investigation) per sample

CHAPTER FOUR

4.0 RESULTS AND DISCUSSION

4.1 Protein properties analysis

4.1.1 Gluten content

The gluten content values of the two wheat varieties and their blends using various non-wheat flours are presented in Table 4.1. The values that are significantly different from each other for each variety, by separation using DMRT method at $p=0.05$, have different superscripts. Gluten levels in the two wheat varieties and their blends ranged from 7.9 % to 10.08 % for Chozi and 3.86 % to 6.3 % for Kwale.

Table 4.1 Percent gluten content of Chozi, Kwale and their blends

Sample (Chozi)	Gluten content, DMRT at $p=0.05$	Kwale	Gluten content, DMRT at $p=0.05$
Unblended Chozi, (Cpl)	10.08 ^a	Unblended Kwale, (Kpl)	6.30 ^a
Chozi/cowpea, (Ccp)	9.64 ^{ab}	Kwale/ soybean, (Ksoy)	6.08 ^a
Chozi/cassava, (Ccas)	9.28 ^{ab}	Kwale/ sorghum, (Ksor)	5.87 ^a
Chozi/banana, (Cbn)	9.21 ^{ab}	Kwale/ cowpea, (Kcp)	5.79 ^a
Chozi/sweetpotato, (Csp)	9.04 ^{ab}	Kwale/ sweetpotato, (Ksp)	5.7 ^a
Chozi/ soybean, (Csoy)	8.92 ^{ab}	Kwale/ maize, (K mz)	4.85 ^{ab}
Chozi/arrowroot, (Carr)	8.32 ^{ab}	Kwale/ cassava, (Kcas)	4.79 ^{ab}
Chozi/sorghum, (Csor)	8.00 ^{ab}	Kwale/ arrowroots, (Karr)	4.77 ^{ab}
Chozi/maize, (Cmz)	7.90 ^b	Kwale/ banana, (Kbn)	3.86 ^b

The higher gluten content (7.9-10.08 %, Table 4.1) in Chozi and its blends compared to the values in Kwale and its blends, (3.86-6.3 %) was attributed to the differences in the environments they were grown and also due to their genetic makeup. Chozi a drought resistant variety can cope in the wheat growing areas of the country and therefore attain a higher gluten content in the event of drought (as experienced in the year 2000 in many wheat growing areas including those that KARI was using in the trials). Kwale a non-drought resistant variety cannot achieve high protein and gluten content in the event of drought noted from the lower protein content and the gluten contents. Gluten constitutes of 85 % of total grain and flour protein, lower protein levels in flour usually indicates a lower gluten content and therefore poorer dough properties. The higher gluten and protein

in correlation analysis shows strong positive correlation since the two values in grains are closely linked.

In both Chozi and Kwale, unblended wheat flour had the highest gluten content as expected. Addition of non-wheat flour lowered the gluten content, through the dilution of the total gluten in flour. The decrease in the gluten content was not proportional to the amount of wheat flour removed for the same level of replacement with a specific type of non-wheat flour and was different in both Chozi and Kwale.

The lowest gluten content in Chozi blends is 7.9% which is much higher than the gluten content of 6.38% in plain Kwale wheat. Since the bread baking quality is dependent on gluten content and quality (Bushuk, 1985, Manley *et al*, 1992) then Chozi gluten may be further diluted by replacement with non-wheat flours and still produce acceptable bread. Chozi from the gluten content values reported is superior compared to Kwale, the NDR bread wheat variety. High gluten content in Chozi indicated good dough characteristics measured by farinography as did the low tolerance, this led to a negative correlation between the gluten content and tolerance. Cpl reported a significant negative correlation at $r = -0.95$ at $p = 0.05$, (Appendix 2) between the tolerance and the gluten content thus indicating good dough characteristics. In farinography tests a low tolerance in flour indicates good dough strength while in the measurement of the gluten content higher gluten levels indicates better dough strength. The high gluten content, the low tolerance and the significant negative correlation in cpl agreed with the classification by KARI, 2001 which has classified Chozi as a bread wheat variety. The strong positive correlation between the gluten content and stability reported in ksoy at $r = 0.99$ at $p = 0.05$, (Appendix 3) was due to the low level of dilution. High gluten content in a bread wheat variety indicates good dough properties and acceptable baking qualities, as does the high stability. Despite the blending which diluted the gluten content both the stability and the gluten content remained high and produced a strong positive correlation. Gluten levels were not detrimentally lowered and the good dough strength expressed as high dough stability measured in ksoy was attained.

4.1.2 Protein content

The protein content of Chozi, Kwale and their blends are reported in Table 4.2. The protein content of the flours from Chozi varied from 9.36 % in the Chozi/maize blend (cmz) to 15.52 % in the unblended Chozi flour. In Kwale the values ranged from 6.4 % in the Kwale/maize (kmz) blend to 12.08 % in the Kwale/ soybean (ksoy) flour blend. Most

blends were significantly different from unblended wheat flour in protein content except the soybean derived flour at $p=0.05$ level of significance by the analysis of variance (ANOVA). The samples sharing the same superscript are not significantly different and vice versa when the means of the percent protein contents are separated using the DMRT method at $p= 0.05$. Table 4.3 shows the absorbances of different blends of the two wheat varieties while Figure 4.1 shows the standard protein curve.

The protein content of unblended wheat ranged from 11.67 % in Kwale to 15.52 % in Chozi. This observation agrees with the analysis by (KARI, 2001) for the protein content of various bread wheat varieties from the wheat crop of the year 2000, the protein content varied from 11.8-17 %. While wheat protein content varies from 6-25 % in grains in different wheat varieties, bread wheat varieties have wheat protein contents of 11-14%, (Blackman and Payne, 1987). The protein content of Chozi (cpl) was higher than that of kpl and was found to be significantly different. Significant differences were found between the protein content of every blend in Chozi (and cpl) with it's equivalent in Kwale (and kpl). All blends of Chozi had higher protein contents than their equivalent non-wheat flour blends in Kwale.

Table 4.2 Percentage protein content for Chozi, Kwale and their blends

Sample (Chozi)	Protein content DMRT at $p=0.05$	Sample (Kwale)	Protein content, DMRT at $p=0.05$
Cpl	15.52 ^a	Ksoy	12.08 ^a
Csoy	15.44 ^a	Kcas	11.96 ^a
Ccp	13.68 ^a	Kpl	11.67 ^a
Ccas	13.68 ^a	Kbn	9.36 ^{ab}
Csor	12.76 ^{ab}	Kcp	9.184 ^{ab}
Csp	12.24 ^{ab}	Ksor	8.84 ^{ab}
Cbn	10.06 ^b	Karr	7.34 ^b
Carr	9.54 ^b	Ksp	6.72 ^b
Cmz	9.36 ^b	Kmz	6.40 ^b

The significant difference between the flour protein content in kpl and cpl, (Table 4.2), was explained in part by the genetic variation. While Climate and the soils influence the protein content more than the influence of the genes (Williams, 1975) genetic variation in the genes that code the synthesis of storage proteins also influence the protein content in different proteins. This variation has a 5 % influence on the protein content (Johnson and

Mattern, 1978). The two being different wheat varieties while showing all the properties of bread making varieties may have been influenced variably by their respective storage protein coding genes leading to significant difference between the protein content of Chozi and Kwale. Comparison of different wheat varieties invariably leads to the recommended protein content levels for bread wheat varieties. Haglund and Johansson (1998), has established that levels of 8 % and above are required to produce acceptable bread from the fermentation process. Blackman and Payne (1987) have stated that the minimum amount of protein required to produce acceptable bread in standard tests is 11-12 %.

Blending decreased the protein content in most blends in comparison to the unblended wheat flour except ksoy and kcas that had higher protein concentrations than kpl, while the lowest concentrations were recorded in karr, ksp and kmz. Soy flour had higher protein content in both wheat flours, (Table 4.2). In Kwale, this was higher than even kpl. Soy seeds like other grain legumes have higher protein contents that make up to 40 % of dry matter content (Norton, *et al*, 1985) and even at this low replacement level raised the protein content of the flour. Ccp's significant correlation to both the dough development time (DDT) and tolerance at $r=0.97$ and $r=-0.97$, (Table 8 in Appendix 2) respectfully is mainly because of the high level of quality protein due to the low level of protein dilution at 10 % replacement level. Dried cowpea grains contain up to 22 % protein (Norton *et al*, 1985), protein content is a major factor affecting the bread making quality (Khan *et al*, 2000). Haglund and Johansson, 1998 established the threshold of at least 8 % protein content for the production ofv acceptable bread. Apart from ccp, ccas, and csoy both of which had high protein contents reported strong or significant positive correlation between the DDT and the protein content an indication of good dough characteristics. Other correlation values that revealed the dough characteristics included the correlation between the tolerance and the protein content, where in csp and ccp (at $r = -0.9$ and $r = -0.99$ (Appendix 2),). Low tolerance coupled with high protein as noted in the two samples is a strong indicator of good dough characteristics. The two properties of flour are usually negatively correlated because the indicator of one quality is high in one parameter while the other is low. Strong negative correlation between the two properties indicates better dough properties in flour. The low tolerance produced a strong negative correlation was attributed to the high protein content in the two blends. A low protein content in kmz led to a fast attainment of optimal DDT. This low protein content directly influenced the low DDT leading to a positive and significant correlation at $r = 0.99$. Within a specific type of

wheat, there are a specific number of bonds formed either as HMW-HMW-GS, LMW-LMW-GS and LMW-HMW-GS to achieve optimal consistency at peak time.

Table 4.3 Spectrophotometric values of the protein of Chozi, Kwale and blends

Sample (Chozi)	Absorbance at 662 nm	Sample (Kwale)	Absorbance at 662 nm
Cpl	0.975	Ksoy	0.794
Csoy	0.970	Kcas	0.783
Ccp	0.935	Kpl	0.771
Ccas	0.935	Kbn	0.587
Csor	0.830	Kcp	0.576
Csp	0.818	Ksor	0.565
Cbn	0.667	Karr	0.454
Carr	0.613	Ksp	0.437
Cmz	0.587	Kmz	0.411

The lower the protein content the lower the number of HMW-HMW-GS formed, with a specified amount of potassium bromate terminating bond formation, the time spent in this reaction will be shorter than when the levels of proteins are higher.

Table 4.4 The protein concentration – optical density values of Bovine Serum Albumin

Protein concentration in µg/ml	Absorbance at 662 nm
0.0	0.00
0.05	0.325
0.1	0.485
0.2	0.565
0.3	0.615
0.4	0.860
0.5	0.930

The low loaf volumes recorded in most Kwale blends was in tandem with the observation by Haglund and Johansson, (1998), that protein contents below 8 % produces bread of unacceptable volume and quality. The loaf volume of kcas with a protein content of 11.96 %, only lower than the ksoy was the only sample that was comparable to the loaf

volumes of Chozi and their blends. The high protein content in cassava-based blends was attributed to the single cell protein added by fermentation during the flour preparation (Omune *et al*, 2001).

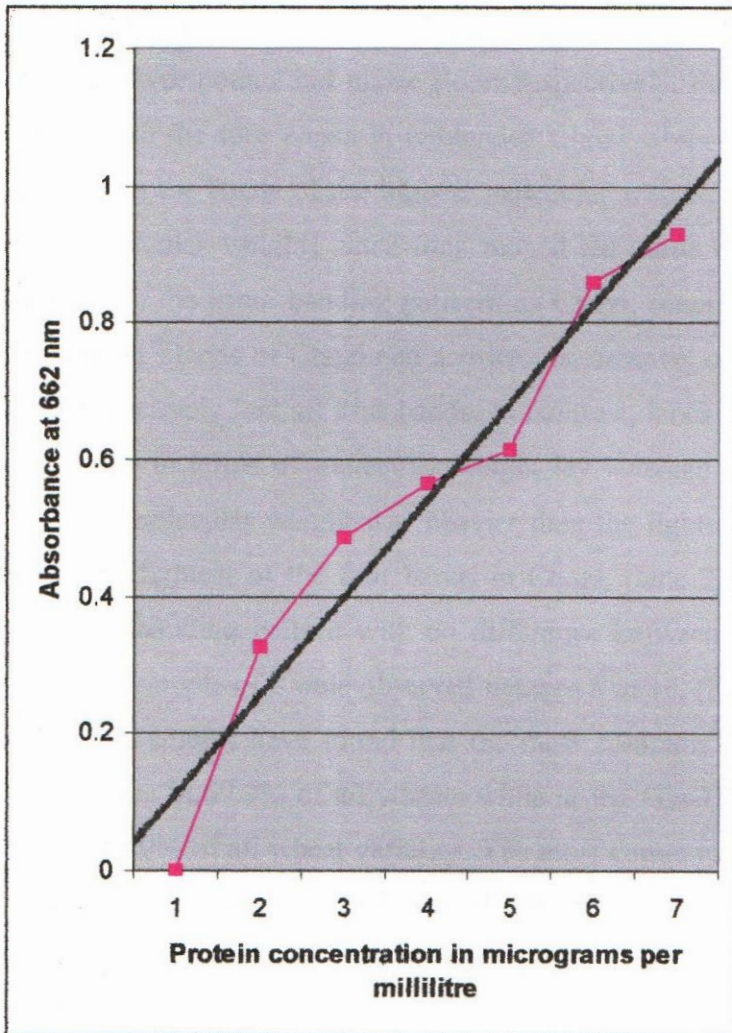


Figure 4.1 The standard protein curve of Bovine Serum Albumin

Key

Y-axis- 0-7 represents the protein concentrations from 0- 210 µl/ml with each graduation representing 30 µg/ml.

4.1.3 Protein profile analysis

SDS-PAGE of glutenin proteins revealed the differences in the two bread wheat varieties. Differences in the banding patterns in both LMW-GS and the HMW-GS in Chozi and Kwale have been shown in figure 4.2 and Table 4.5. The determination of the protein profiles of the two wheat varieties and their blends showed the presence of five HMW-GS (bands) in Kwale (lane 10) and four bands in Chozi (lane 2). This agreed with

the observation by Manley *et al*, (1992) that each bread wheat variety has between three and five HMW-GS. The subunits in each of these wheat varieties was however different observed from the electrophoretogram, (Figure 4.2), from the different distances moved, revealing allelic differences between the glutenin subunits of Kwale and Chozi.

In Chozi, lanes 17, 15, 13 and 12 representing the blends of Chozi and banana, sorghum, sweetpotato and maize flours respectively, three bands (subunits) were present compared to the four bands in unblended Chozi, observed in lane 2 in Figure 4.2. With reference to the bands of the highest molecular weight, the first two bands had the same size (molecular weight) since they moved the same distances in all blends of Chozi. Blends with the same banding patterns as Chozi, observed in lanes 18, 16, 14, 2 and 1, representing blends of Chozi and arrowroots, cassava, cowpea, unblended Chozi and soy flour, respectively had all four bands. In contrast, lanes 17, 15, 13 and 12, lightest of the three bands in terms of molecular weight lay between the last two bands in unblended Chozi. Its molecular weight was heavier than the lightest band in Chozi but lighter than the second lightest of the four bands in Chozi, (lane 2, Figure 4.2). Kwale in contrast revealed a banding pattern with no difference between the unblended flour or blended flour. Each sample of Kwale observed in lanes 8 to 16, (Figure 4.2) had five HMW-GS.

Previous studies have found that the most common subunit at the Glu-1A locus is 2 which occurs in 60.9% of all wheats while at the Glu-1B locus, the most common is the 20+20y in 50% of all wheat varieties. The most common subunits at the Glu-1D locus are 2+12, which are present in 89.2 % of all wheat varieties Rodriguez *et al*, (1998). The good dough properties reported from farinography and the loaf volumes showed that Chozi the better HMW-GS profile or proportion. Manley *et al*, 1992 reported strong correlations between the presence of specific HMW-GS and good or poor bread making qualities exists.

Differences between the LMW-GS of both Chozi and Kwale presented in Figure 4.2 were in line with the reports of Rodriguez *et al*, (1998) that great allelic differences in the LMW-GS exists and Amsal *et al*, (1995) reported that gliadins and LMW-GS were unique for all cultivars and lines, respectively. A distinctive banding pattern of the LMW-Gs was noted in the two wheat varieties. The most remarkable difference in the glutenin proteins of the two cultivars noted was the number of LMW-GS present in the two wheat varieties. Chozi had fourteen LMW-GS (observed as bands in the gel) while Kwale had twelve. Some bands present in unblended flours but absent in blended flour in both wheat varieties were detected. In Chozi, lanes 15, (csor), 13, (csp) and 12, (cmz) lacked two band each

that were present in cpl. Levels below 20 µg/ml are not detectable by the coomassie brilliant blue dye. They had one subunit that was absent in cpl. In Kwale, lanes 9, 7, 6 and 4, (Figure 4.2), representing karr, kbn, ksp and ksor respectively also lacked the same banding pattern as unblended Kwale or other blends of Kwale with reference to the LMW-GS banding pattern. The four blends had a faint (low) banding intensity relative to kpl.

The presence of some LMW-GS and some HMW-GS in cbn, csp, cmz and csor (Figure 4.2), compared to cpl was partly attributed to the presence of non-wheat glutelins. Cereals including maize and sorghum have glutenin-like proteins but varying from wheat in their proportions of the prolamin /glutelin (Payne, 1987). The study did not conclusively establish whether extra LMW-GS expressed in the two cereal varieties, cmz and csor were derived from non-wheat cereal sources. The absence of some bands in blends Kwale, observed in karr, kbn, ksp and ksor, (Figure 4.2), as in Chozi was also attributed to loss during glutenins extraction. The structural similarities especially with maize and sorghum glutelins to those of wheat possibly led to their forming of bonds with sequestering components like tannins to result in wheat gluten /non-wheat glutelins/ tannin complexes which were removed from the aqueous glutenins /glutelins supernatant during the extraction. This was considered one of the reasons why some bands were absent in the gel when compared to the profile of unblended flours. The sodium dodecyl sulphate breaks hydrogen and hydrosulfryl bonds between protein subunits but not covalent bonds as may exist in tannin glutelin protein complexes. Tannins are present in most sorghum varieties in large quantities and the method of preparation, (Table 3.1), did not adequately remove them since the outer cover was milled together with other parts of sorghum grains.

Previous reports have revealed a large size variation among HMW-GS and LMW-GS. Different gluten proteins have either an enhanceive effect on the dough properties or a detrimental effect. They affect the water absorption, reduction/oxidation, rheology, gas retention and ultimately the final quality of bread (Coultrate, 1996). The probable presence of subunits that confer superior bread making qualities to Chozi (which was better ranked in terms of the bread loaf volume) was likely.

The link between the gluten proteins to the osmoregulation gene was not established for the two wheat varieties. While the protein quality is primarily genetically determined (Finney *et al*, 1987), it may also be affected by growth conditions after anthesis (Huebner *et al*, 1990). A relationship between the weakening of the dough as a result of heat stress and an increase in the proportion of gliadin in the gluten has been reported (Blumenthal *et al*, 1994). The dough properties noted from the high stability, the good loaf volume, the

high dough development time and the low tolerance in Chozi led to this study's suggestion that the HMW-GS in Chozi conferred to its flour the dough strength in line with the report by Wrigley *et al*, (1994). The good dough properties in Chozi were also explained in part by its better capacity for drought resistance taking in consideration that the cultivar is grown within the ME5B region.

Ehdaie and Waines (1997), reported that chromosome 1 in wheat at the complex locus 1D might carry genes that decrease relative water loss, genes inherited together with these genes influence the overall baking quality and drought tolerance of different cultivars of bread wheat. This genetic inheritance was not proved at Chozi's *Glu-D1*, however living organisms have multiple processes to cope with stress, this association may have been present helping decrease water loss. Their association would prevent the events associated with water loss including the decrease in the level of synthesis of glutenin proteins and the altering of the gliadin / glutenin proportion occurring and thus negatively impacting the dough properties. This positive influence arising from the presence of such a HMW-GS and its association with these sorts of genes would be absent in Kwale.

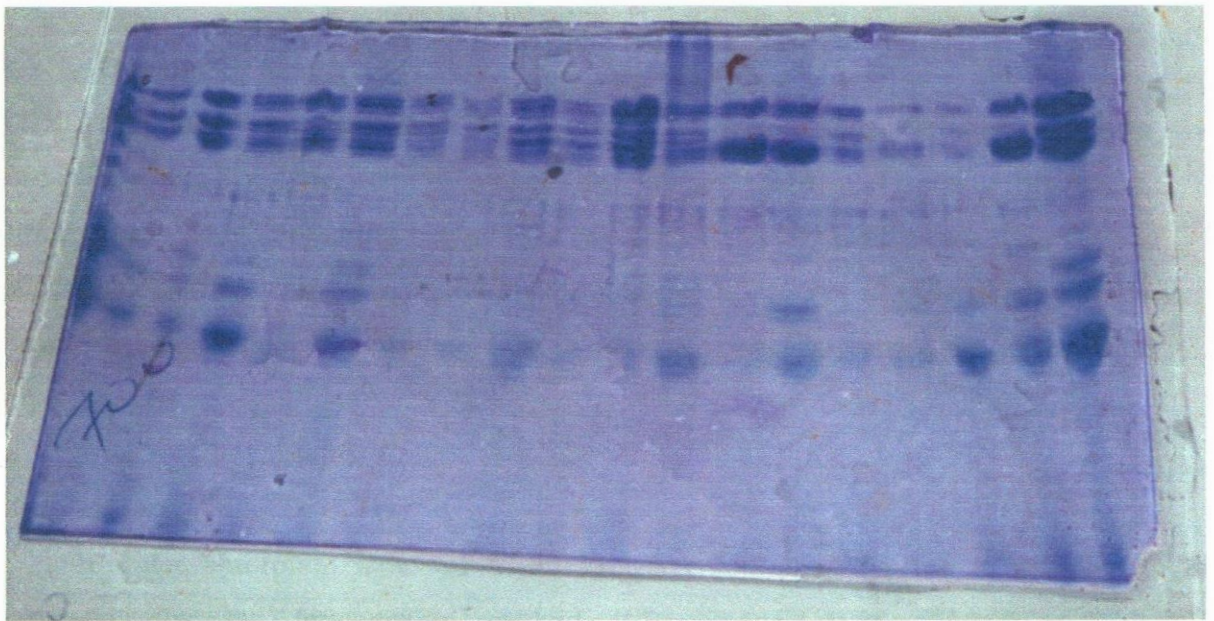
Thirteen different loci have been identified and chromosomal location determined in wheat varieties. The HMW-GS are assigned three loci namely the *Glu-A1*, *Glu-B1* and the *Glu-D1* mapped close to the centromeres on the long arm of the homoeologous chromosomes 1A, 1B and 1D, (Payne *et al*, 1987). The LMW-GS, ψ and ω - gliadins are controlled by loci *Gli-A1*, *Gli-B1* and *Gli-D1* located distally on the short arms of each of the chromosomes of the homoeologous group 1 (Payne, 1987). Many of the loci are genetically complex as they are tightly linked genes that rarely recombine. Over 100 genes have been recognized and assigned on chromosome arms mainly isoenzymes. One study has found that because of the position of the gene that controls the production of endosperm peroxidase (*Per-4*) relative to the osmoregulation gene (*or*), differences exist in drought resistance between different wheat varieties (Morgan J.M, 1999). These differences have been found to influence the dough properties with bread wheat varieties which have high levels of production at the *or* loci leading to high levels of endosperm peroxidase which improves the dough properties in bread wheat varieties.

Flour protein, gliadin/glutenin proportion, dough breakdown, dough extensibility and maximum resistance for seven cultivars from fifteen diverse locations showed that the proportion of glutenin was dependent on cultivar. Performance at *Glu-D1* locus was associated with environment and cultivar (Panozzo and Eagles, 2000). With Chozi, a recognized drought resistant wheat variety showing good dough properties and producing

acceptable baking qualities, analysis led to the conclusion that the differences in Chozi and Kwale were to an extent influenced by differences in drought resistance during their culture.

Table 4.5 –The HMW-GS compositions of Chozi, Kwale and their blends.

	Blend	Number of HMW-GS bands	Number of LMW-GS bands
18	Carr	4	14
17	Cbn	3	14
16	Ccas	4	14
15	Csor	4	13
14	Ccp	3	14
13	Csp	3	13
12	Cmz	3	13
11	Kmz	5	12
10	Kpl	5	12
9	Karr	5	12
8	Kcas	5	12
7	Kbn	5	12
6	Ksp	5	12
5	Ksoy	5	12
4	Ksor	5	12
3	Kcp	5	12
2	Cpl	4	14
1	Csoy	4	14



1_b 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18

Figure 4.2 The glutenin profiles of Chozi, Kwale and blends

Blends key

- 18 Carr
- 17 Cbn
- 16 Ccas
- 15 Csor
- 14 Ccp
- 13 Csp
- 12 Cmz
- 11 Kmz
- 10 Kpl
- 9 Karr
- 8 Kcas
- 7 Kbn
- 6 Ksp
- 5 Ksoy
- 4 Ksor
- 3 Kcp
- 2 Cpl
- 1 Csoy
- 1_b Gluten

4.2 Dough characteristics

Different flours vary in dough characteristics as measured in farinograph (water absorption, stability (consistency), dough development time and mixing tolerance). Differences in dough properties between the two flour types and their blends were largely reflections of differences between the glutens and the differences in their starches water absorptive indices since the milling of the two wheat flours was carried out in the same way.

4.2.1 Absorption

Flour water absorption index of 57-60 % is recommended for use in the dough preparation by the fermentation method (Chamberlain, 1995). Based on this recommendation cpl had the more acceptable absorption index. Blends derived from cowpea, sorghum, maize and cassava produced flours with acceptable absorption indices in both Chozi and Kwale. Arrowroots, banana and sweet potato blends had higher absorption indices than the recommended levels, while soy flour's water absorption was within the acceptable range in Chozi but unacceptably high in Kwale.

Some blends were significantly different in their absorptive indices from the unblended flour, reported showing different superscripts in Table 4.6. Cmz and ccas were significantly different from cpl in absorption. There was no significant difference between blends from the same non-wheat source blended at the same level of replacement by DMRT at $p=0.05$ except with regard to the sweet potato derived flour, where csp (at 65 %) and ksp at (50%) showed great difference, (Table 4.6) in absorption values.

Generally the tubers had the highest levels of percentage absorption followed by legumes (Table 4.6) then cereals were reported. Significant difference was reported between cpl (59.7%) and kpl (50.4%) by ANOVA. No Significant difference in values was reported between one type of non-wheat flour blend and any other by ANOVA, while the DMRT revealed differences in the mean percentage absorption indices in different blends. Table 4.6, below shows the % Absorption for Chozi, Kwale and their blends. Kpl and ksp had absorption levels below the recommended 57-60 % level. There was similarity in the percent absorption between specific type of blend in Chozi and Kwale. Kwale based blends had higher indices. The difference in the absorption index was narrow at a range of 1 % (sor) to 5 % (soy), (Table 4.6). The absorption of cpl (59.7) was higher and significantly different from that of kpl (50 %), (Table 4.6). This was due to the

differences in the proteins that make up the gluten in the two wheat varieties, noted when mixed with water to reveal different diffraction patterns (Hoagland, 1987).

Table 4.6 Percent absorption for Chozi, Kwale and their blends

Samples (Chozi)	% Absorption, DMRT at p=0.05	Samples (Kwale)	% Absorption, DMRT at p=0.05
Carr	73.60 ^a	Karr	76.50 ^a
Cbn	65.20 ^b	Kbn	68.30 ^b
Csp	65.00 ^b	Ksoy	66.30 ^b
Ccp	63.40 ^{bc}	Kcp	62.70 ^{bc}
Csor	62.80 ^{bc}	Ksor	61.90 ^{bc}
Csoy	61.20 ^{bc}	Kmz	61.10 ^{bc}
Cpl	59.70 ^{bc}	Kcas	57.30 ^{cd}
Cmz	57.30 ^{bc}	Ksp	50.60 ^d
Ccas	55.00 ^c	Kpl	50.40 ^d

Depending on the type of wheat and the type of proteins in it, the level of starch damaged is differentially noted after milling. Wheat varieties of higher strengths have higher starch granules damage (Blackman and Payne, 1987). Damaged starch granules absorb water at twice their weight of water compared to undamaged starch (Blackman and Payne, 1987). The higher DDT, better dough stability and lower tolerance all indicate better dough strength in Chozi when compared to Kwale, (Tables 4.7, 4.8 and 4.9). The higher protein content in Chozi explains the higher absorptive index since absorption increases as protein content increases and gluten quality improves (Pomeranz and Shellenberger, 1975).

With the two cereal blends from maize and sorghum, the similarity in their average carbohydrates content at 73 % for both compared to 70 % for wheat (Hegsted *et al*, 1954) led to their flours reporting water absorption rates similar to those of the wheat varieties which were used in blending them. The low water absorption in kpl compared to other cereals was probably attributed to a lower level of damaged starch in Kwale. Wheat varieties with lower grain hardness (poorer dough strengths) also have lower levels of starch grain damage and also lower level of water absorption.

Higher water absorption than the unblended flour was noted in both Chozi and Kwale. The cowpea and soybean had lower absorptive indices than those of tubers because of the lower levels of carbohydrates and the higher protein content. Starch has a higher absorptive index than protein (Riganakos and Kontominas, 1994). Tubers are rich in

carbohydrates, which give higher flour absorptive indices, as observed in the arrowroot blend, carr (73.6 %), karr (75.8 %) compared to cpl (59.7 %) and kpl (50 %). Starch makes up to 50-70 % of dry matter of sweet potato and other tubers and fruit based flours, influencing properties such as the swelling power, solubility, gelatinization, enthalpy and pasting properties as they are analysed by the Brabender amylograph (Sakamoto and Bouwkamp, 1985). Absorption of water by starch varies depending on the temperature and the type of starch.

Chozi based flours produced breads with acceptable loaf volumes while Kwale based breads had low loaf volumes due to the higher protein content as well as the better protein quality enabling the holding of water more efficiently than in Kwale. High water absorption due to a high level of damaged starch has been found to result in poor bread quality if the protein is not capable of holding additional water (Meeds, 1995). The low stability, DDT and the high tolerance of Kwale based blends bolstered this observation and argument.

The low absorption in cassava-based blends compared to other tubers was attributed to the possible presence of fibre in the flour that has been associated with the inhibition of the water uptake in previous studies (Balagopalan *et al*, 1988). Kpl reported a strong negative correlation ($r=-0.9$) between its absorption and DDT while ksor, kcp and kbn reported strong positive correlation values at ($r=0.96$, $r=0.99$ and $r=0.99$, respectively mainly because of the differences in their starch water absorption capacities. The non-wheat components in blends raised the water absorption capacity.

4.2.2 Stability

The stability was recorded for the different blends of Chozi and Kwale in Table 4.7. The means of their stability values were separated using DMRT method at $p=0.05$. Replacement of wheat flour with non-wheat flour in both Chozi and Kwale led to a decrease in the dough stability. Unblended Chozi flour and the flours blended at low levels of wheat flour replacement had the highest stability. There was a significant difference between the dough stabilities in Kwale and Chozi.

Different trends were reported in Chozi and Kwale. The stability in Chozi ranged from 4.25 minutes in cmz to 27 minutes in cpl. The values in Kwale range from 2.25 minutes in ksor to 5 minutes in kpl. Chozi based blends had higher stability at the 500 BU, (Table 4.7) compared to Kwale implying that certain gluten protein components present in Chozi but are absent in Kwale that imparted to the dough more stability also partly attributed to

the high level of gluten. Chamberlain, (1995), reported that flours with acceptable dough strength in bread making have dough stability values ranging from 3-20 minutes. A higher dough stability time indicates good dough strength especially if the farinogram curve has more thickness.

Table 4.7 Stability for Chozi, Kwale and their blends

Chozi	Minutes, DMRT at p= 0.05	(Kwale	Minutes, DMRT at p= 0.05
Cpl	27 ^a	Kpl	5 ^a
Cbn	25 ^a	Kbn	4.25 ^a
Csoy	17 ^b	Ksoy	4 ^a
Csp	10 ^c	Kcas	3.5 ^a
Ccas	9.5 ^c	Ksp	3 ^a
Csor	7.5 ^c	Kcp	2.5 ^a
Ccp	7 ^c	Karr	2.5 ^a
Carr	5 ^c	Kmz	2.5 ^a
Cmz	4.25 ^c	Ksor	2.25 ^a

Glutenin confers to a flour the elasticity and resistance to breaking during mixing. Good and poor bread making quality has been associated with two allelic pairs at the *Glu-D1* complex locus. The probable presence of protein subunits that confer good dough properties possibly contributed to the better stability and the better baking quality noted with regard to the loaf volume, (Table 4.10). Chozi had the higher level of gluten when unblended and even in the blended flour at any level of replacement.

In Chozi, only the stabilities of cpl and cbn were not significantly different and higher than other Chozi blends, showing a better capacity than the rest. The high dough stability of cpl, (Table 4.7), compared to those of its blends is due to the high levels of gluten. The low-level replacement in soy and banana, (Table 3.1), led also to a high stability.

In the legumes, a decrease in stability relative to the unblended flours was attributed to the dilution of the gluten proteins. The stability of the Chozi/ legume blend was more acceptable than the Kwale/ legume blend where the dough stability was very low. The low stability in part explains the low loaf volume in Kwale/ legume blends, (Table 4.7), because low stability usually causes overmixing in standard loaf making procedures like the one used in this study. Oomah, (1983) reported that the increase in the protein concentration without an increase in the gluten, concentration decreased not just the

stability but also the DDT and the absorption in wheat/ oat blend. The results of this investigation were found with regard to the legumes to agree to this observation.

In the tubers and cereals, there was a decrease in stability compared to unblended wheat at the given replacement level, from Table 3.1. The decrease in the dough stability was attributed to the increase in the starch content and the dilution of the gluten quantity in flour especially in samples with higher non-wheat flour replacement levels (Oomah, 1983)

Replacement with either maize or sorghum in both Chozi and Kwale decreased the dough stability more than any other type of flour, (Table 4.7). The prolamin like proteins at 55 % in maize and 70 % in sorghum, of the total storage proteins in the two cereals (Johnson and Mattern, 1978), were considered to have negative impact on the dough stability unlike the glutenins in unblended wheat. The effectiveness of glutenin influence to the dough properties, particularly the HMW-GS is high (Hoseney *et al*, 1986). Prolamin proteins lead to the formation of HMW-LMW bonds, which are detrimental to the dough strength (Wood, 1995) because they confer less elasticity, no resistance and weak extensibility leading to poor gas holding and low loaf volumes.

Kpl with a dough stability of five minutes was acceptable for bread making but poor compared to Chozi (27 minutes), (Table 4.7), drought stress was considered to be the probable cause of this relatively poor dough strength. Kwale is a wheat variety classified by KARI, 2001 as a bread wheat variety but not drought resistant unlike Chozi. Acceptable dough stability is achieved when the glutelins and prolamins ratio is 1:1, raising the levels of LMW-GS or gliadins lowers the stability. The presence of tannins in sorghum which chelate proline-rich proteins, prevents optimal dough stability achievement, (Butler, 1982), explaining the lower loaf volume relative to the volumes of unblended wheat or even maize based blends.

Low tolerance and high stability in flour indicates in flours good dough properties, on analysis the two parameters are correlated negatively. In csoy and csor (both at $r=-0.99$, Appendix 2), the high protein content positively influenced the dough properties. Strong correlation reported between the two properties in kpl ($r=-0.92$) and kmz ($r=0.84$), Appendix 3 showed the differences in dough properties due mainly to the differences in protein and gluten levels.

4.2.3 Tolerance

The tolerance as a indicator of dough strength in Chozi ranged from no recorded tolerance (zero tolerance) in both cbn and coy to 100 B.U.s in carr. Tolerance values with the same superscripts were not significantly different. Kwale recorded tolerance values ranging from 70 B.U.s to 160 B.U.s in karr. Replacement with non-wheat flour raised the tolerance of the flours, except the soy flour in both Kwale and Chozi where the tolerance was not significantly different from that of the unblended (pure) flours. This was also the case in cbn which was statistically similar to cpl. Blends with higher levels of replacements had higher tolerances than blends at low levels of replacements. The tolerance values are reported in Table 4.8. For the same level of replacements with a specific type of non-wheat flour, the tolerance values were higher in Kwale than in Chozi. The cowpea-based blend was the exception to this observation being significantly different from the unblended wheat flour while substituted at only ten percent. Chamberlain, (1995), reported that flours with acceptable tolerance have values ranging from 0-120 Brabender units. Low tolerance value of flour indicates good dough strength.

Arrowroots based blends had the highest tolerances, while soy and banana blends had the lowest tolerances in Kwale and Chozi, (Table 4.8). The high tolerance in arrowroots based flour blend, (Table 4.8) 100 BUs (carr) and 160 BUs (karr), was due to the low protein content and the gluten content and the high absorption leading to poor dough strength. Protein contents below 8 % lead to production of unacceptable loaf volumes (Haglund and Johansson, 1998). There were significant differences between cpl and Chozi blends and between kpl and Kwale blends. Significant difference was found between the cpl and other cereals and tubers, (Table 4.8).

The tolerance was higher in comparison to the unblended Chozi, the high tolerance was attributed to the dilution of the gluten; low level of gluten confers weakness because the glutenin proportion in the blended flour is lowered. Glutenin forms the HMW-HMW bonds that confer to a dough resistance and elasticity, the two properties that result when an optimal three-dimensional protein network that supports the dough structure is formed (Wood, 1995). Tolerance was not significantly different from that of cpl in ccas, csoy and cbn, (Table 4.8), implying that the blends were not significantly different from cpl at their respective non-wheat flour replacement level to the wheat flour.

High replacement levels, (Table 3.1) can produce breads that are acceptable with these two non-wheat flours in Chozi. The low tolerance in csoy and cbn was attributed to the low replacement levels. The level of glutenin was not considerably lowered and the dough

structure of the flours from these blends was not significantly different from that of unblended Chozi flour.

Table 4.8 Tolerance for Chozi, Kwale and their blends

Chozi	Tolerance in BUs, DMRT at p=0.05	Kwale	Tolerance in BUs, DMRT at p=0.05
Carr	100 ^a	Karr	160 ^a
Csor	70 ^b	Kcas	130 ^b
Cmz	60 ^{bc}	Kmz	120 ^{bc}
Ccp	60 ^{bc}	Ksp	120 ^{bc}
Csp	45 ^c	Kcp	110 ^{bcd}
Ccas	15 ^d	Kbn	100 ^{de}
Cpl	10 ^d	Ksor	90 ^{de^f}
Csoy	0 ^d	Ksoy	80 ^{ef}
Cbn	0 ^d	Kpl	70 ^f

Only in karr and kpl was significant difference recorded at $p < 0.05$ DMRT level, (Table 4.8), with karr showing unacceptably high tolerance levels. Increase in tolerance has been attributed to the dilution of the gluten proteins that leads to a reduction in cohesiveness and also due to the inelasticity of non-wheat flour proteins (Oomah, 1983). Only with arrowroots was the replacement level found to impact negatively on the tolerance, (Table 4.8).

Significant differences between Chozi (and Chozi blends) on one hand and Kwale (and Kwale blends) on the other in the 0.05 points of the F-distribution were also noted. The high tolerance of Kwale and its blends when compared to Chozi and its blends was attributed to the poorer quality due to the presence of gluten subunits in Kwale that led to the formation of weak dough structure. The rise in tolerance especially in blends derived from Kwale was due to the dilution in wheat gluten. Tsen, 1979, reported that this rise with concurrent fall in DDT and stability in Kwale was due to dilution of the gluten by the addition of non-wheat flour.

The strong but negative correlation reported between the DDT and tolerance in cpl, csor and ccp at $r = -0.87$, $r = -0.99$ and $r = -0.89$, (Appendix 2), respectively showed acceptable dough properties since the two parameters reported as high DDT and low tolerance indicated good dough strength.

4.2.4 Dough development time (DDT)

Unlike other farinograph parameters, for most blends DDT was not a good parameter for this analysis in the two wheat varieties as the values were similar. Chamberlain, (1995), reported that flours with acceptable DDT in bread making have values ranging from 3-10 minutes. The means of the DDT of the two varieties and their blends are shown in Table 4.9. The alphabetical figures recorded as superscripts show the DDT values that are significantly / not significantly different from each other. The DDT values ranged from 3 minutes in ccas to 7.5 minutes in cbn in the Chozi-based samples. In Kwale, the DDT ranged from 1.2 minutes in Kcas to 5 minutes in kpl. A general decrease in the DDT was noted with the blending of wheat with non-wheat flour in both Chozi and Kwale.

Table 4.9 Dough development time for Chozi, Kwale and their blends

Sample (Chozi)	Minutes, DMRT at p=0.05	Sample (Kwale)	Minutes, DMRT at p=0.05
Cbn	7.5 ^a	Kpl	5 ^a
Csp	7 ^{ab}	Kbn	4 ^{ab}
Cpl	6 ^{abc}	Karr	3.5 ^{abc}
Csoy	6 ^{abc}	Ksor	3 ^{abc}
Ccp	5.5 ^{abc}	Ksp	3 ^{abc}
Csor	5 ^{bcd}	Ksoy	3 ^{abc}
Carr	5 ^{bcd}	Kcp	3 ^{abc}
Cmz	4 ^{cd}	Kmz	2.5 ^{bc}
Ccas	3 ^d	Kcas	1.2 ^c

The blends with the highest non-wheat flour replacement, (Table 3.1), had the lowest DDT in Chozi, where as with the exception of csp, blends with DDT values that were not significantly different from cpl, (Table 4.9), were substituted at low levels. The maize and cassava based flour which had the highest replacement levels produced the shortest DDT.

Maize blends had the most different value when correlation values were calculated for DDT, (Table 4.9), and other parameters, Appendix 2 and 3. The influence of maize zein, similar to wheat gliadin and which is detrimental dough properties in bread when present at a higher proportion in flour was considered the cause of the differences in farinograph properties in maize/wheat flour blend when compared with other flours. In maize based blends, the lower DDT was also attributed to the decrease in the proportion of glutelins (HMW-GS) relative to the levels of prolamins resulting in the increase of prolamins

contributed to the dough by the maize flour in the blend. Relative proportions of glutenins and gliadins are a major factor in the bread making character of flour (Coultate, 1996). In each specific blend, Chozi based blends had higher DDT values. The narrowest difference in DDT was recorded between cpl (6 minutes) and kpl (5 minutes), representing no significant difference by ANOVA or by DMRT value, (Table 4.9).

The DDT was significantly different in ccp, csor, carr, cmz and ccas compared to the rest, all of which with the exception of ccp had high replacement levels, (Table 4.9). In the rest of the blends the dilution of the HMW-GS proportion in the flours was thought to be the cause of the lowered DDT. With samples ground to a 60-mesh fineness, the DDT could have been influenced by the dilution of the gluten content mainly. The DDT value in wheat/ cassava (wcas) blend, (Table 4.7), was the one of the lowest in both Chozi and Kwale due to the low gluten content resulting from the high replacement level at 20%, (Table 3.1). The higher DDT in Chozi was attributed to the better HMW-GS profile as well as the presence of these HMW-GS at higher levels. Higher gluten content represents higher levels of HMW-GS since wheat storage proteins exist at a ratio of 1:1 between glutenins and gliadins.

Four samples including cpl, csp, csor and carr all had an acceptable optimal DDT and all produced acceptable loaf volume. The two parameters have an enhance complementary effect in bread wheat noted as positive correlation with r-values at $r=0.96$, $r=0.95$ and $r=0.99$ for csor, csp and carr respectively. The optimal DDT achieved in ksoy and its acceptable loaf volume led to a significant positive correlation at $r=0.99$. The correlation value was attributed to high gluten content relative to other Kwale blends.

4.3 Baking quality evaluation

4.3.1 Loaf volume

The means of the samples in Tables' 4.10 and 4.11 were arranged in descending order. Table 4.10 reported the general loaf volume comparison in Kwale and Chozi and their blends. Table 4.11 tabulated the loaf volume comparisons within each bread wheat variety comparing samples within the Chozi block and within the Kwale block. The means show the calculated average of the loaf volume from three replications of the baking. The loaf volumes varied from 480 c.c in ccas blend to 685 cc in ccp in Chozi, the volumes in Kwale ranged from 315 c.c to 550 c.c in kcas. Significant differences were noted in the loaf volumes derived from the same non-wheat flour in the two wheat varieties except with regard to the cassava based variety where the loaf volumes of kcas (550c.c) and ccas (480c.c) were not significantly different, (Table 4.10).

Table 4.10 Loaf volume for Chozi, Kwale and their blends 1

Sample	Variety	Blending ratio	Volume, DMRT at p=0.05
Ccp	Chozi	90:10:00	685 ^a
Cbn	Chozi	90:10:00	660 ^{ab}
Csoy	Chozi	95:05:00	650 ^{ab}
Cbn	Chozi	80:20:00	585 ^{bc}
Kcas	Kwale	80:20:00	550 ^{cd}
Csor	Chozi	85:15:00	530 ^{cd}
Carr	Chozi	85:15:00	525 ^{cd}
Cpl	Chozi	100:00:00	515 ^{cde}
Csp	Chozi	85:15:00	500 ^{cde}
Ccas	Chozi	80:20:00	480 ^{def}
Ksoy	Kwale	95:05:00	435 ^{efg}
Kmz	Kwale	80:20:00	400 ^{fgh}
Kcp	Kwale	90:10:00	400 ^{fgh}
Ksp	Kwale	85:15:00	390 ^{gh}
Karr	Kwale	85:15:00	375 ^{gh}
Kpl	Kwale	90:10:00	355 ^g
Kbn	Kwale	90:10:00	350 ^g
Ksor	Kwale	85:15:00	315 ^h

Significant loaf volume differences between Chozi (and Chozi/non-wheat flour blends) and Kwale and (Kwale/ non-wheat flour blends) were reported by DMRT for example in

Table 4.10 where ccp, cbn, csoy, cmz, csor, carr, csp were significantly different in the loaf volume values compared to their equivalents in Kwale.*

Table 4.11 Loaf volume for Chozi, Kwale and their blends 2

Chozi	DMRT at p=0.05	Kwale	DMRT at p=0.05
Ccp	685 ^a	Kcas	550 ^a
Cbn	660 ^{ab}	Ksoy	435 ^{ab}
Csoy	650 ^{abc}	Kmz	400 ^{ab}
Cmz	585 ^{abcd}	Kcp	400 ^{ab}
Csor	530 ^{abcd}	Ksp	390 ^{ab}
Carr	525 ^{abcd}	Karr	375 ^b
Cpl	515 ^{bcd}	Kpl	355 ^b
Csp	500 ^{cd}	Kbn	350 ^b
Ccas	480 ^d	Ksor	315 ^b

Loaf volume has been consistently correlated previously to the grain protein content, the flour protein content and alveograph components of bread flour originating from fermentative and thermal reactions (Martinez, 1996). Chozi blends produced the more acceptable bread in terms of the loaf volumes and specific gravity (mostly greater than 3.5 g/ml) compared to Kwale because of the higher levels of protein and the gluten content and better gluten profile or a better glutenin/gliadin ratio when compared to Kwale. Specific gravity values below 3.5 g/ml in loaves prepared by the fermentation method are considered of low and the flour, of poorer breadmaking quality. KARI uses the standard wood weight (400c.c) in loaf volume determination. Loaves with volumes below 400 c.c by the rapeseed displacement method have low loaf volume. The high loaf volume in cpl was attributed to the high dough stability and the long dough development time, (Table 4.11). Loaf volume response is more related to the protein content than the gliadin/glutenin ratio according to Agenbag and DeVillers, (1995), this observation was confirmed by the correlation values between the protein content and the loaf. The replacement level influenced the loaf volume in Chozi where high replacement levels in Table 3.1 led to lower loaf volumes. The loaf volume varied with the type of non-wheat flour and the level of replacement. Many blends had higher loaf volumes than the unblended wheat flour in both Chozi and Kwale, this observation was agreeable to the reports of various proposers of acceptable levels of replacements of the blends wheat flour with non-wheat flour, recorded in Table 4.10 and 4.11. The amylase content of the dough

has an effect on the rate of gas production by the yeast. The higher loaf volumes in cbn, cmz, csor, carr than cpl in Chozi and ksp, kmz, kcas, karr and kbn were partly attributed to the addition of dextrin and soluble starch, which increases gas production. It has been shown that 42-95 % of starch that makes 80-90 % of sweet potato dry weight is converted to sugars and dextrans during baking (Garcia and Palmer, 1980).

Kwale produced breads of lower loaf volumes except in kcas and ksoy because of the lower protein and gluten content, (Tables 4.1 and 4.2). Kwale has been classified as being of bread making quality. The poor bread making qualities attributed to the cultivar in this study were thought to be due to the effects of drought which may have reduced the glutenin levels during the samples anthesis. Glutenin contributes considerably to the strength properties of dough (Wrigley *et al*, 1994). Reduction in glutenin levels alters the gliadin/glutenin proportion even if the gluten content is not affected (Agenbag and DeVillers, 1995). Low intensity water stress reduces the LV (Simmonds, 1989). High LVs hinted at the ability of non-wheat flour to replace wheat flour acceptably in blends of ccp, cbn and csoy at higher levels of replacements than those used in this study.

Low bread volume in sorghum based blend in both Kwale and Chozi was attributed in part to the presence of polyphenols especially tannins. These bind to proteins rich in proline (prolamins in sorghum and wheat), their binding inactivates tannins and overcome their anti nutritional effect but also limits the dough development and may make them unavailable in diet (Butler, 1982).

The maize-based blends produced loaves with acceptable volumes in both Kwale and Chozi at 400c.c and 585 c.c respectively, despite the low gluten content and the poor dough stability. The loaf volume of kbn was unacceptably low because of the low gluten levels at 3.86 %. The quantity of gluten in dough overrides any other consideration of its intrinsic quality (Evan and Peacock, 1981). In Chozi, legumes had the highest values followed by cereals and then tubers. Most blends of Kwale were significantly different from those of Chozi when compared by the DMRT statistical method at $p=0.05$.

High loaf volume coupled with low tolerance indicated good baking qualities, flours that showed strong negative correlations had good dough characteristics and baking qualities and vice-versa. The strong negative correlation reported in cpl at $r=-0.9$ and csor at $r=-0.99$, Appendix 2 agreed with this observation and was attributed to the high protein content and also due to high protein quality. The contrast was true between the correlation of the loaf volume of a flour and its dough stability where strong positive correlation indicated good quality. Poor dough characteristics indicated by the r -value between the

stability and loaf volume in cmz ($r=-0.93$) was due to the low protein content in cmz at 7.9 %.

4.3.2 Sensory scoring

The loaves from different blends were subjected to sensory scoring using a panel of 10 assessors; the results were recorded in Table 4.12. The results were the average of the attributes reported in Table 4.13 from all the assessors. The calculated sensory score of each sample (blended or unblended) in Chozi revealed a significant difference between the samples. Score based on a hedonic scale of, (1-dislike very much, 9-like very much, Appendix 1). The trend in acceptance was described as moving from the highest acceptability in cereals > tubers > legumes in Chozi. Significant difference was found between samples of cereals (except cmz) and the rest of the blends (except carr) recorded in Table 4.12, in Chozi.

In Kwale, unblended flour produced bread of the highest acceptability, Table 4.12. Analysing the sensory scores revealed different relationships between blends and the unblended flour as well as differences between Chozi (and blends) and Kwale (and blends). The trend in Kwale was less clear than in Chozi when classifying blends generally as legumes, non-wheat cereals, fruit or tuber. On a scale of one to nine, all the breads at these replacement levels were organoleptically acceptable, with the lowest acceptable in both wheat varieties, the cbn (5.95) and 5.98 (kbn) indicating '6' in the hedonic scale, (appendix 1), as the term 'like slightly'.

All crumbs were cream coloured, the more aesthetically acceptable colour for the crumb with the exception of the Chozi /arrowroot blend (carr) in Chozi, which was brownish. The whiter crumb colour of loaves in soy based blends compared to the crumb colour of loaves from tuber based flour blends was attributed to the presence of lipoxgenases in the full fat soy flour used. Lipoxgenases are active in full fat soy flour and leads to the production of a whiter crumb colour through bleaching when added to bread dough (Chamberlain, 1995).

In terms of the structure (texture) of the crumb, the majority of the blends were classified as soft except in csoy, ccp, cmz that were classified as good. The ccas was however classified as semi soft in texture. There was no significant difference between Chozi (and blends) and Kwale (and blends) at the 1 % point, differences at 5% were found between kpl and kmz. Significant difference was found between cpl and both cmz and cbn. Significant difference was also found between kpl and both ksoy and karr.

Table 4.12 Sensory scores in graduations of 1 to 9.

Sample (Kwale)	Sensory score, DMRT at p=0.05	Sample (Chozi)	Sensory score, DMRT at p=0.05
Wheat (Kwale)	7.23 ^a	Wheat: maize	6.97 ^a
Wheat: sorghum	6.6b ^{bc}	Wheat (Chozi)	6.58 ^{ab}
Wheat: arrowroots	6.41 ^{cd}	Wheat: cassava	6.5 ^{ab}
Wheat: sweetpotato	6.28 ^{de}	Wheat: sweetpotato	6.48 ^{ab}
Wheat: soybeans	6.23 ^{de}	Wheat: soybeans	6.47 ^{ab}
Wheat: banana	6.15 ^{de}	Wheat: sorghum	6.1 ^b
Wheat: cassava	6.02 ^c	Wheat: arrowroots	6.1 ^b
Wheat: maize	5.98 ^e	Wheat: cowpea	5.96 ^b
Wheat: cowpea	5.98 ^c	Wheat: banana	5.95 ^b

Texture quality in bread has been linked to the elasticity and extensibility of the dough, these are dependent on the gluten content and quality (He and Hoseny, 1992). Grain size is an important part of texture make up of the crumb in bread. Addition of non-wheat cereal flours in wheat flours to blend prevents the retention of gas in doughs leading to heaviness and coarseness in texture, (Hoagland, 1987). This was noted in ksor and kmz as (heaviness), as small grains in csoy, csor, csp, as coarseness in carr and cmz, and as big grains in ccp, cmz and cbn. Larger quantities of non-wheat flour diluted the gluten and protein, this reduced the grain size and led to the hard texture, observed in carr, kmz, kcp, kbn, ksoy, ksor and kcas.

Sour taste was noted in cassava blends and was attributed to the higher cassava levels, which produced traces of prussic acid. Gumminess in sweet potato blends breads was attributed to the presence of pectic derivatives (Hoagland, 1987).

The high levels of fibre in tubers especially those of sweet potato and arrowroots is believed to have cause the roughness of the crust in blends with non-wheat flour from these two tubers. High crust colour (dark brown) in tubers and cereal substituted flours rather than a golden brown colour was attributed to the low protein contents especially in arrowroots based blends loaves. The crust colour in carr, ksor, karr and kbn was darker than the rest, (Table 4.13). The darker colour was probably due to the combination between lysine and sugars present in their doughs during Maillard reactions (Kent, 1983).

No significant difference between blends of Kwale and Chozi was observed except between ccas and kcas. Cmz and ccas were significantly different from cpl as were carr

from karr and csor from ksor with regard to the internal colour of samples. There was no significant difference in blends when compared with the unblended sample in terms of smell. Significant difference was found between cmz, csoy and carr from cpl at the 5 % point of the F-distribution in the general acceptability of the samples. Cmz, cbn and cep were significantly different from kmz, kbn and kep respectively at the 5 % points of the F-distribution.

The sensory scores for each attribute in both Chozi and Kwale are shown in Table 4.13. Bitterness in the crust and crumb of cowpea and soybean blends was reported in both Chozi and Kwale flours. A beany flavour was present in blends of cowpea and soy in both Chozi and Kwale. This flavour is difficult to remove and hence a limiting factor to the use of these two legumes (Johnson and Mattern, 1978).

Cmz, csp and cbn reported strong positive correlation between their dough stabilities and their sensory scores at $r = 0.99$, $r = 0.98$ and $r = 0.91$ respectively because both their parameters were reported as high. High dough stability indicates the presence of high levels of protein that forms the dough superstructure that must be adequate so as to hold the air and starch producing a large volume. Without this achievement, the loaf volume is low and the structure may be hard or too soft, the crumb may be heavy and coarse instead of fluffy. The strong positive correlation reported in the three blends indicates the presence of high levels of HMW-GS of the size that positively affects the dough and the bread quality.

Table 4.13 The sensory scores of in Chozi, Kwale and their blends

CHOZI							KWALE					
	Structure	Smell	Taste	Crust colour	Internal colour	General Acceptability	Structure	Smell	Taste	Crust colour	Internal colour	General Acceptability
Wheat	7.3	7.9	6.7	6.6	7.3	7.16	5.8	6.9	6.4	6.4	6.4	6.67
Wheat: maize	6.1	6.6	5.6	5.8	6	6.02	5.8	6.2	6.8	6.3	6.3	6.18
Wheat: sorghum	6.8	6.6	6	6.7	6.9	6.6	6.7	5.9	5.9	5.6	5.6	6.19
Wheat: cassava	7.2	6.8	6.5	6.8	6.9	6.84	6	5.6	5.8	6.4	6.4	6.34
Wheat: arrowroots	6.9	6.1	5.5	5.9	7.4	6.36	6.3	6.2	6.1	5.4	5.4	6.06
Wheat: sweetpotato	6	6.1	5.7	6.9	6.7	6.28	5.8	6	6.3	6.8	6.8	6.32
Wheat: cowpea	6.7	5.9	5.6	5.4	6.6	6.04	5.3	6.4	5.8	7.3	7.3	6.28
Wheat:soybean	5.4	5.8	6.6	6.1	6.1	6	6.3	6.1	5.6	5.8	5.8	5.95
Wheat: banana	7.1	6	4.9	6.3	6.6	6.18	6	6.5	6.9	7.3	7.3	6.57

CHAPTER FIVE

5.0 CONCLUSIONS AND RECOMMENDATIONS

5.1 Conclusions

The gluten proteins in Chozi (the DR wheat variety) had the more positive influence to the bread quality because of either the better gliadin / glutenin proportion in Chozi flour or due to the presence of specific gluten subunits that confer to flour the ability to produce loaves of higher acceptability.

The link between the aspect of drought resistance and the protein quality was not conclusively arrived at. It was however noted that Chozi, DR wheat variety produced more acceptable breads due to higher protein and gluten contents in environments predisposed to drought stress.

The dough characteristics, namely 3-20 minutes (stability), 3-10 minutes (DDT), 55-65 % (absorption), 0-120 B.U.s (tolerance) and the protein content (8 %) for most samples was within the acceptable range for the production of acceptable loaves.

The high molecular weight glutenin subunits of the two wheat varieties revealed differences seen from the banding pattern of the samples. Four HMW-GS were identified from the banding pattern of Chozi while five were identified in Kwale. Regarding the dough characteristics, the LMW-GS are less important to the bread baking quality than the HMW-GS.

5.2 Recommendations

The study of the gluten proteins of Kenyan wheat, including, the established and the newly developed or introduced varieties using biochemical markers rather than the traditionally used morphological characteristics is recommended so as to overcome the complexities.

The study of storage proteins especially in cereals which have close evolutionary relationships with wheat and which are similar in certain properties and their influence to the dough properties so that blending can be studied in a more reproducible way.

The study of the effect of non-wheat flour to the wheat gluten proteins (and other vital components) in bread making flour, especially the effect at the molecular level to identify detrimental effects of non-wheat flours in blends has to be investigated.

Studies should be carried out to help overcome the detrimental effects of antinutritional factors. The effect of non-wheat proteins and other components in these flours and the effect on the sensory perception indicating the same level of acceptance to products made

from the blended wheat flours as those from unblended good quality wheat flours should be studied.

Investigation involving differences in the dough properties of other wheat flour blends at different replacement levels should be conducted. Chozi revealed the possibility of good dough properties at higher replacement levels than those now acceptable, generally for wheat varieties. Other wheat varieties that can produce higher replacement levels than those now proposed as the highest that produce acceptable bread should be studied and classified to determine where there are differences generally in blending based on the drought resistance aspect of wheat varieties.

The differences in technological parameters and the protein profile led to the recommendation for further studies of differences in individual wheat varieties to arrive at optimum blending rates for various wheat varieties with different non-wheat flours. Blends of cowpea, banana, soy, maize, sorghum and arrowroots can probably replace Chozi flour acceptability at higher levels based on their loaf volumes, which was even higher than that of unblended Chozi flour.

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APPENDICES

APPENDIX 1

The descriptive scale for sensory evaluation

Judge's name

Date Time

Please assess the samples for general acceptability using the most appropriate numerical value to you on a score of 1 – 9 entering scores in the table no.3 below.

Table 3 Score chart

Sample	Crust colour	Flavour	Texture	Internal colour	General acceptability
Wheat					
Wheat/ maize					
Wheat/ sorghum					
Wheat/ cassava					
Wheat/ sweet potato					
Wheat/ arrow root					
Wheat /banana					
Wheat/ cow pea					
Wheat/ soybean					

HEDONIC SCALE

Quality factor scale

1. Very much desirable
2. Very desirable
3. Moderately desirable
4. Less moderately
5. Neutral
6. Less moderately undesirable
7. Moderately undesirable
8. Very undesirable
9. Very much undesirable

General acceptability

1. Dislike extremely
2. Dislike very much
3. Dislike moderately
4. Dislike slightly
5. Neutral
6. Like slightly
7. Less moderately
8. Less moderately
9. Like extremely

APPENDIX 2

Correlation tables for various properties in Chozi and blends

Table 1 cpl									Table 2 cmz								
	tl	ab	st	dt	ss	lv	gc	pc		tl	ab	st	dt	ss	lv	gc	pc
PC	0.13	-0.95*	0.58	0.38	0.52	0.89	-0.43		PC	0.99**	-0.3	0.99**	0.99**	0.99**	-0.9	-0.3	
GC	-0.95*	0.12	0.67	0.67	-0.99**	0.48			GC	-0.38	0.99**	0.29	-0.47	-0.39	-0.29		
LV	-0.9	-0.59	0.89	0.76	0.08				LV	-0.97*	-0.6	-0.93	-0.98	-0.96*			
SS	0.92	-0.22	0.68	0.5					SS	0.58	0.57	0.99**	0.99**				
DT	-0.9	-0.66	0.97*						DT	0.99**	-0.4	0.99**					
ST	-0.7	-0.81							ST	0.99**	-0.3						
AB	0.19								AB	-0.3							
TL									TL								

Table 3 ccsa									Table 4 csp								
	tl	ab	st	dt	ss	lv	gc	pc		tl	ab	st	dt	ss	lv	gc	pc
PC	0.2	0.5	-0.3	0.99**	-0.3	0.8	-0.6		PC	-0.9	-0.96*	-0.9	-0.25	-0.9	-0.56	0.6	
GC	-0.2	0.94	-0.99**	0.47	0.39	-0.99**			GC	-0.2	-0.8	-0.2	-0.9	-0.69	-0.2		
LV	0.14	0.93	0.36	0.5	0.35				LV	0.14	0.76	-0.5	0.95*	0.71			
SS	-0.98**	0.68	-0.63	0.53					SS	-0.6	0.87	0.98**	-0.6				
DT	0.99**	0.99**	-0.3						DT	-0.2	0.5	-0.19					
ST	0.33	0.99**							ST	-0.99**	0.76						
AB	-0.5								AB	0.76							
TL									TL								

Table 5 csoy									Table 6 csor								
	tl	ab	st	dt	ss	lv	gc	pc		tl	ab	st	dt	ss	lv	gc	pc
PC	0.97*	-0.3	-0.9	0.87	-0.99	-0.7	0.22		PC	-0.7	0.3	0.21	-0.4	-0.7	-0.6	-0.5	
GC	0.97	-0.99**	-0.99**	0.67	-0.72	-0.54			GC	0.97*	-0.69	-0.99**	0.99**	-0.29	-0.54		
LV	-0.6	-0.5	0.5	-0.3	0.99**				LV	-0.99**	-0.3	-0.9	0.99**	0.99**			
SS	0.31	0.67	0.04	-0.2					SS	-0.99**	-0.41	0.87	-0.78				
DT	0.96*	-0.7	-0.99**						DT	-0.99**	0.72	-0.99**					
ST	-0.99**	0.6							ST	-0.9	-0.9						
AB	-0.5								AB	0.5							
TL									TL								

Table 7 carr									Table 8 ccp								
	tl	ab	st	dt	ss	lv	gc	pc		tl	ab	st	dt	ss	lv	gc	pc
PC	-0.99**	-0.96*	-0.91	0.59	-0.99	0.34	-0.99**		PC	-0.97*	0.29	0.47	0.97	0.28	-0.6	0.36	
GC	0.99**	0.94	0.93	-0.6	0.99**	0.93			GC	-0.7	-0.6	0.66	0.14	0.99**	0.66		
LV	-0.24	-0.05	-0.69	0.96*	-0.24				LV	0.77	0.59	-0.4	-0.01	-0.9			
SS	0.99**	0.98**	0.44	-0.8					SS	-0.5	-0.84	0.5	7293				
DT	-0.5	0.33	0.87						DT	-0.9	0.5	-0.7					
ST	0.87	0.76							ST	0.25	-0.99**						
AB	0.98*								AB	0.06							
TL									TL								

Table 9 cbn

	tl	ab	st	dt	ss	Lv	gc	pc
PC	0.6	-0.5	-0.9	0.76	0.97**	0.66	0.99**	
GC	-0.68	-0.59	-0.92	0.72	0.95*	-0.9		
LV	-0.99**	-0.99**	-0.93	0.01	0.46			
SS	-0.4	-0.3	-0.91	0.72				
DT	0.02	0.13	-0.38					
ST	0.92	0.87						
AB	0.99**							
TL								

APPENDIX 3

Correlation tables for various properties in Kwale and blends

Table 1 cpl								Table 2 kmz									
	DT	TL	AB	ST	SS	LV	GC	PC		DT	TL	AB	ST	SS	LV	GC	PC
PC	0.84	-0.99	-0.7	-0.09	0.8	0.99**	0.66		PC	-0.99**	0.01	0.19	0.52	0.97**	-0.37	0.66	
GC	0.73	0.73	0.8	0.99**	0.54	-0.94			GC	0.91	-0.5	-0.61	-0.29	0.81	-0.94		
LV	-0.38	-0.99**	-0.99**	0.98**	-0.99**				AB	-0.2	-0.67	0.84	-0.57	-0.99**			
SS	0.87	-0.94	0.5	0.12					SS	-0.65	-0.23	0.03	0.32				
ST	0.99**	-0.92	0.84						ST	-0.92	0.84	0.94					
AB	-0.9	-0.99**							AB	0.96*	-0.9						
TL	0.84								TL	0.4							
DT									DT								

Table 3 ksp								Table 4 kcas									
	DT	TL	AB	ST	SS	LV	GC	PC		DT	TL	AB	ST	SS	LV	GC	PC
PC	-0.41	-0.86	0.14	-0.91	-0.85	-0.97**	-0.99**		PC	0.41	-0.82	0.28	-0.61	-0.98**	-0.44*	-0.72	
GC	0.84	-0.6	-0.1	-0.9	0.8	0.95*			GC	0.91	-0.47	-0.61	-0.29	0.81	0.94		
LV	-0.6	0.46	-0.4	0.95*	0.5				LV	-0.6	0.1	-0.74	-0.8	0.63			
SS	-0.83	0.7	-0.64	-0.55					SS	0.22	-0.87	-0.07	0.76				
ST	-0.01	0.94	0.6						ST	-0.05	0.94	0.6					
AB	0.96*	0.6							AB	-0.9	-0.96*						
TL	0.94								TL	-0.7							
DT									DT								

Table 5 ksoy								Table 6 ksor									
	DT	TL	AB	ST	SS	LV	GC	PC		TL	AB	ST	DT	SS	LV	GC	PC
PC	0.02	-0.3	-0.41	0.48	-0.35	0.99**	-0.38		PC	-0.34	-0.3	0.69	0.06	0.99	0.67	0.41	
GC	0.77	0.73	0.64	0.64	0.99**	-0.36			GC	0.89	0.29	0.53	-0.8	0.92	-0.3		
LV	-0.9	-0.5	-0.59	-0.79	-0.32				LV	-0.9	-0.9	-0.99**	0.59	0.96*			
SS	0.65	-0.7	0.65	-0.65					SS	-0.9	-0.9	-0.99**	-0.8	0.59			
ST	-0.9	-0.7	-0.7						ST	-0.34	-0.24	-0.04	-0.32				
AB	-0.87	-0.9							AB	-0.99**	-0.7	0.9					
TL	-0.99**								TL	0.96*	0.99**						
DT									DT	0.99**							

Table 7 karr

	DT	TL	AB	ST	SS	LV	GC	PC
PC	-0.08	-0.01	-0.04	-0.08	0.72	0.99**	-0.99**	
GC	0.99**	0.29	-0.5	0.89	0.61	0.99**		
LV	-0.8	-0.1	0.66	0.8	-0.98*			
SS	-0.02	0.73	-0.74	-0.73				
ST	0.99**	-0.93	-0.38					
AB	-0.99**	0.66						
TL	-0.69							
DT								

Table 8 kcp

	DT	TL	AB	ST	SS	LV	GC	PC
PC	-0.1	0.96*	0.64	-0.07	0.07	-0.5	-0.83	
GC	-0.61	0.92	-0.11	0.61	-0.64	-0.05		
LV	0.08	-0.43	-0.99**	-0.76	-0.85			
SS	-0.73	0.99**	-0.76	0.99				
ST	0.99**	-0.93	-0.08					
AB	0.99**	0.66						
TL	0.68							
DT								

Table 9 kbn

	DT	TL	AB	ST	SS	LV	GC	PC
PC	0.8	0.96*	0.43	0.52	0.91	-0.8	-0.99**	
GC	-0.61	0.92	-0.11	0.61	0.61	-0.99**		
LV	-0.32	-0.94	-0.87	-0.78	0.51			
SS	0.98**	0.51	0.01	-0.18				
ST	-0.38	0.72	-0.9					
AB	0.99**	0.7						
TL	0.61							
DT								

KEY: For appendix 2 and 3;

* Represents correlation is significant at $p < \text{or} = 0.05$

** Represents correlation is significant at $p < \text{or} = 0.01$

DT dough development time

PC protein content

GC gluten content

SS sensory scores

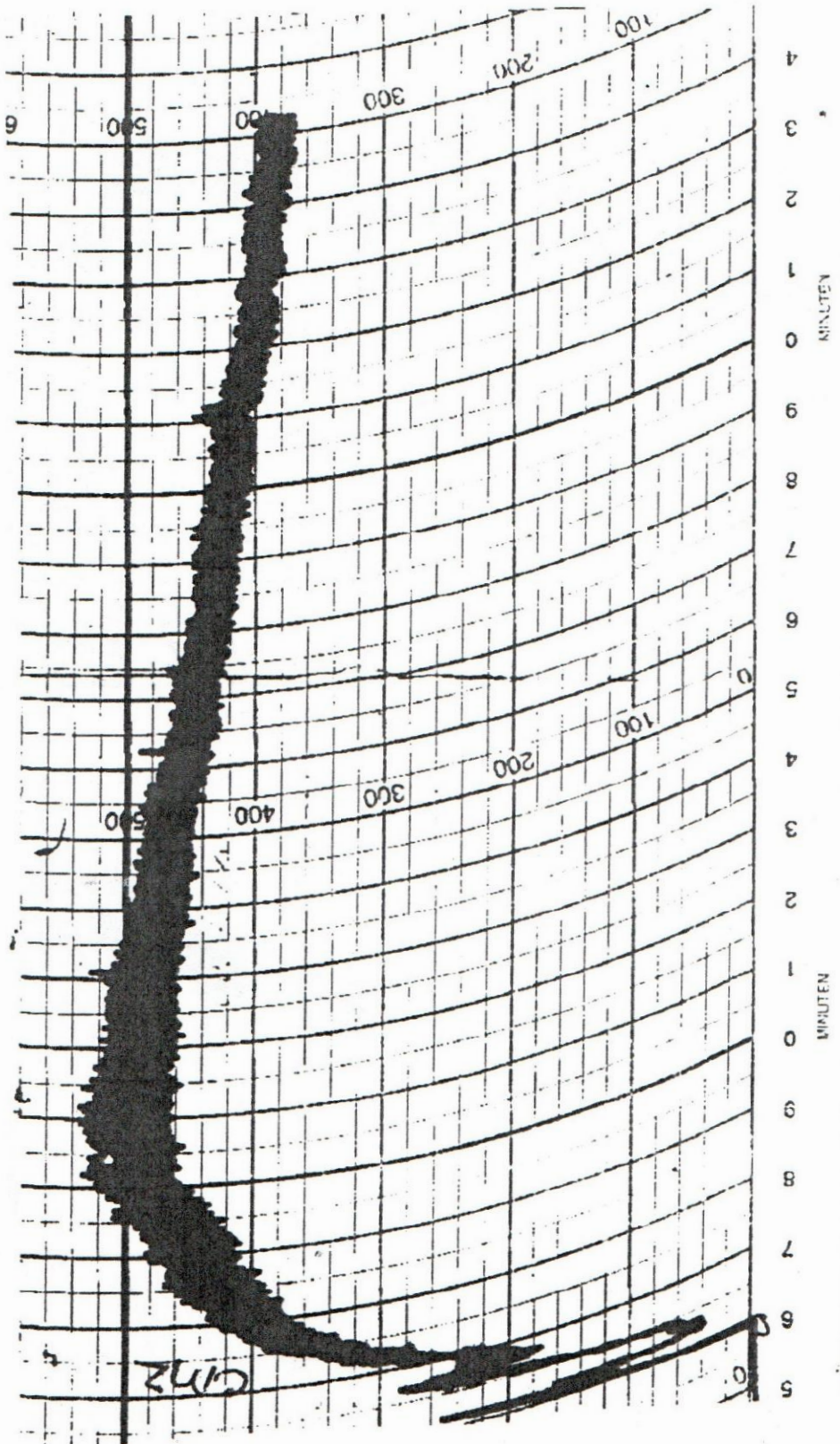
ST stability

TL tolerance

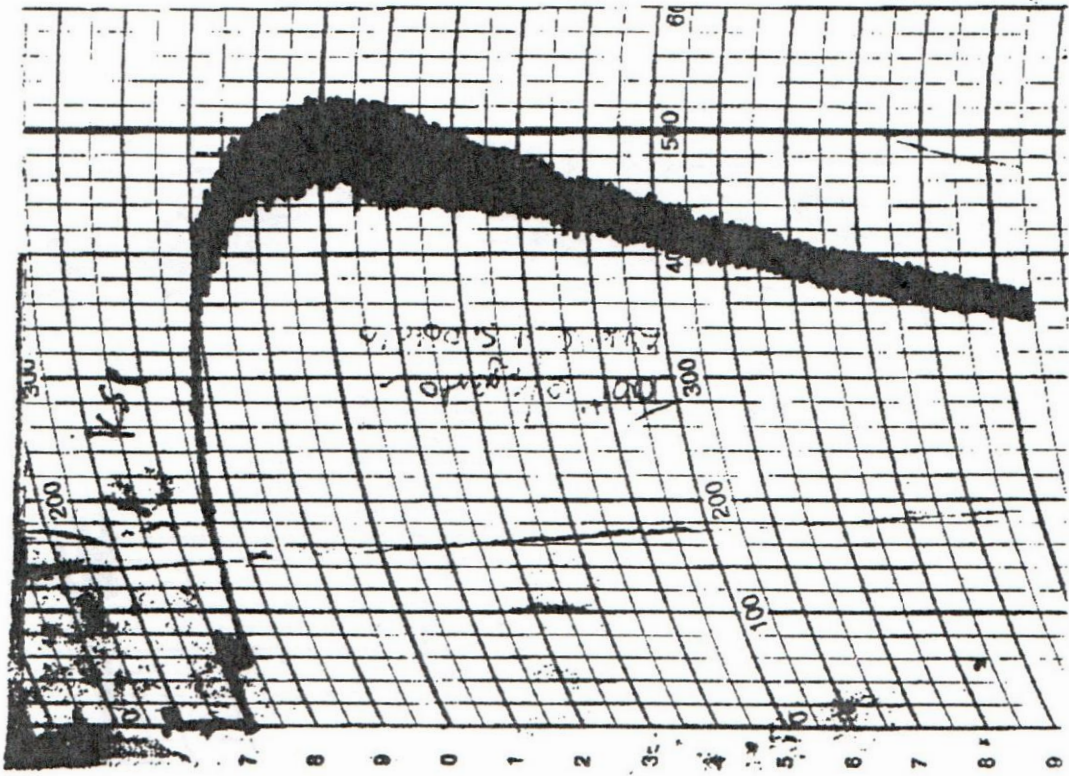
LV loaf volume

AB absorption

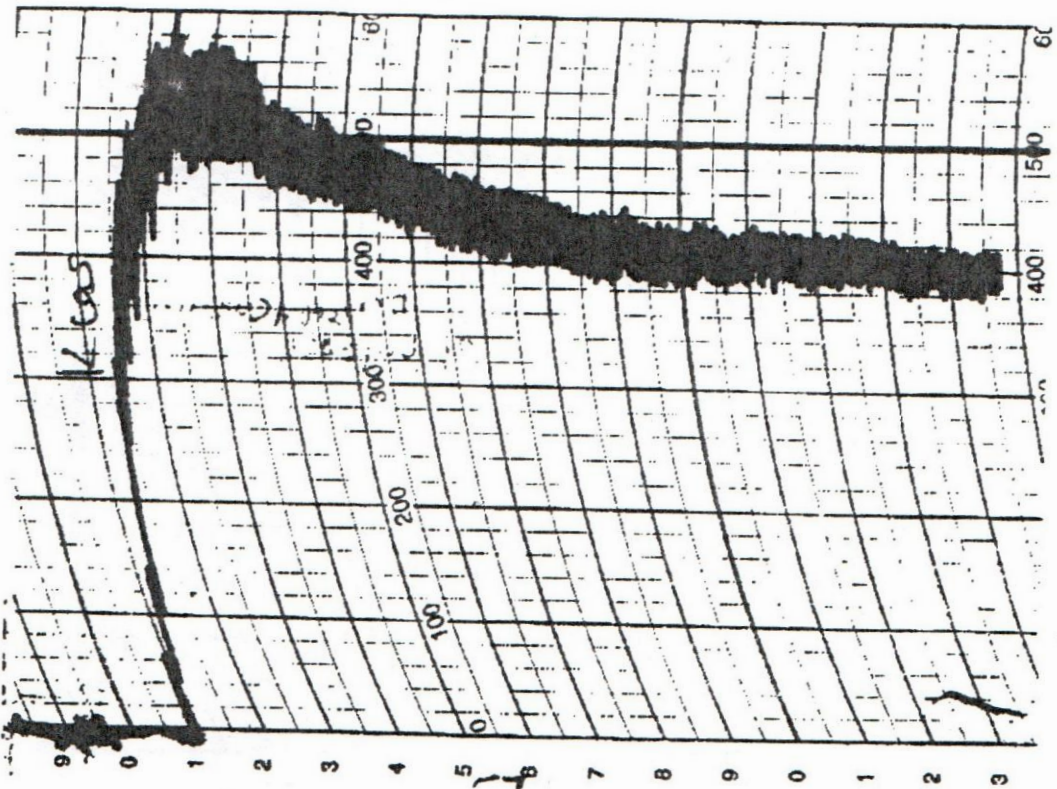
APPENDIX 4:
FARINOGRAPHS



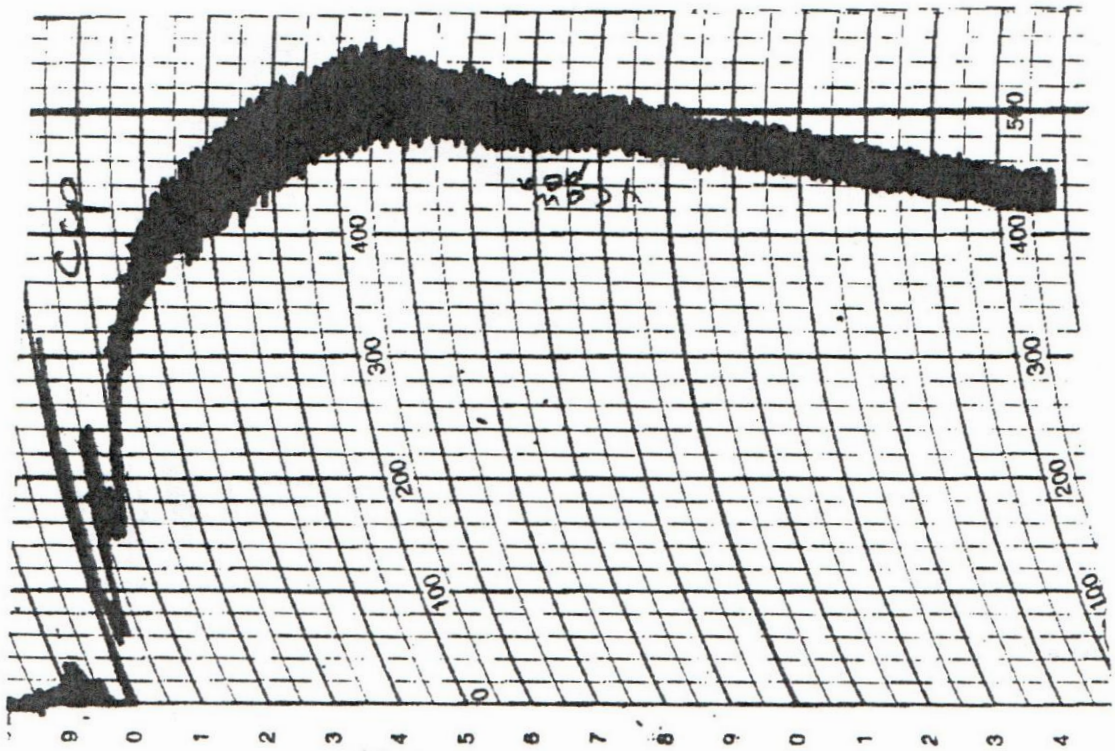
Farinograph 1: Chozi/maize (cmz)



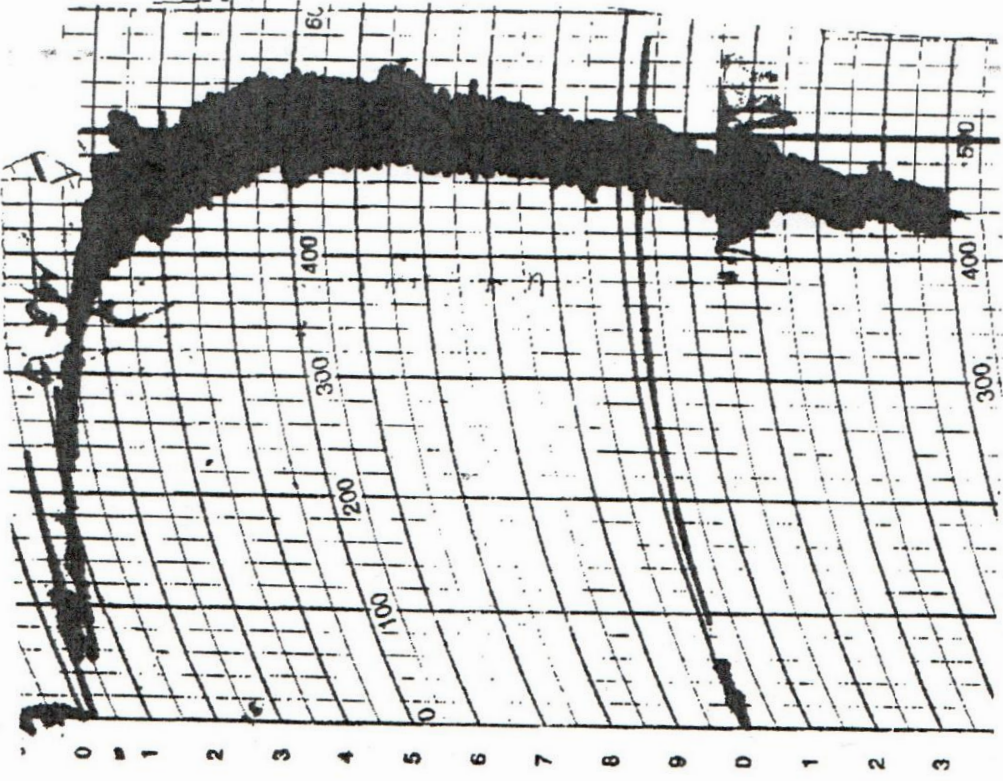
Farinograph 2: Kwale/ sweetpotato (ksp)



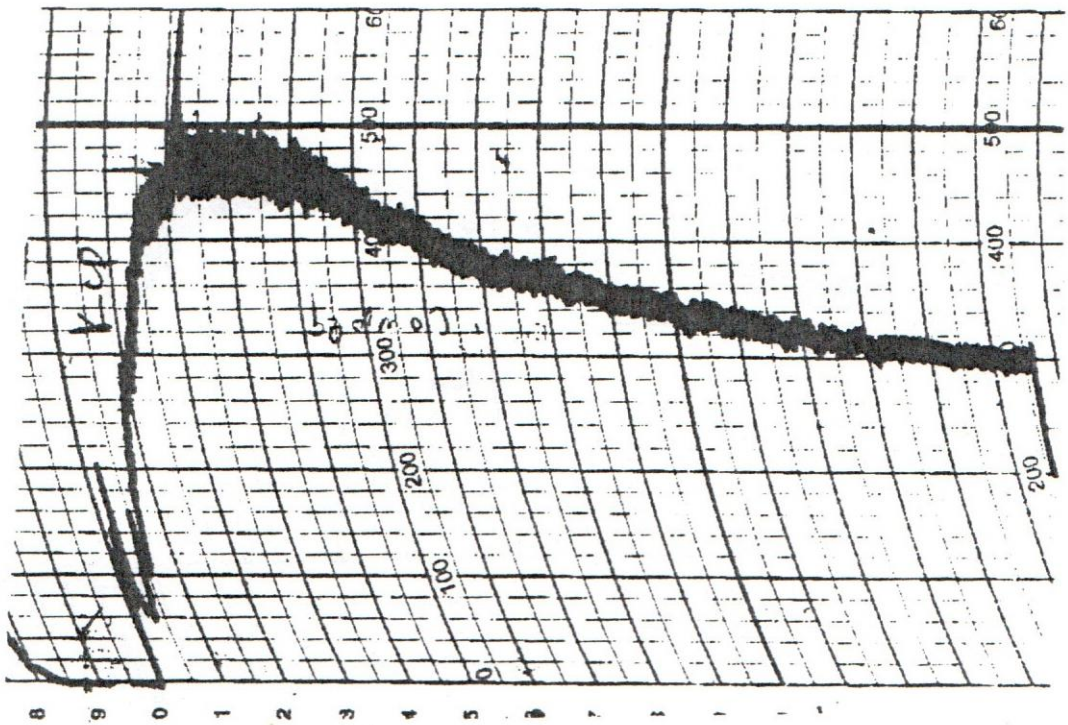
Farinograph 3: Kwale /cassava (kcas)



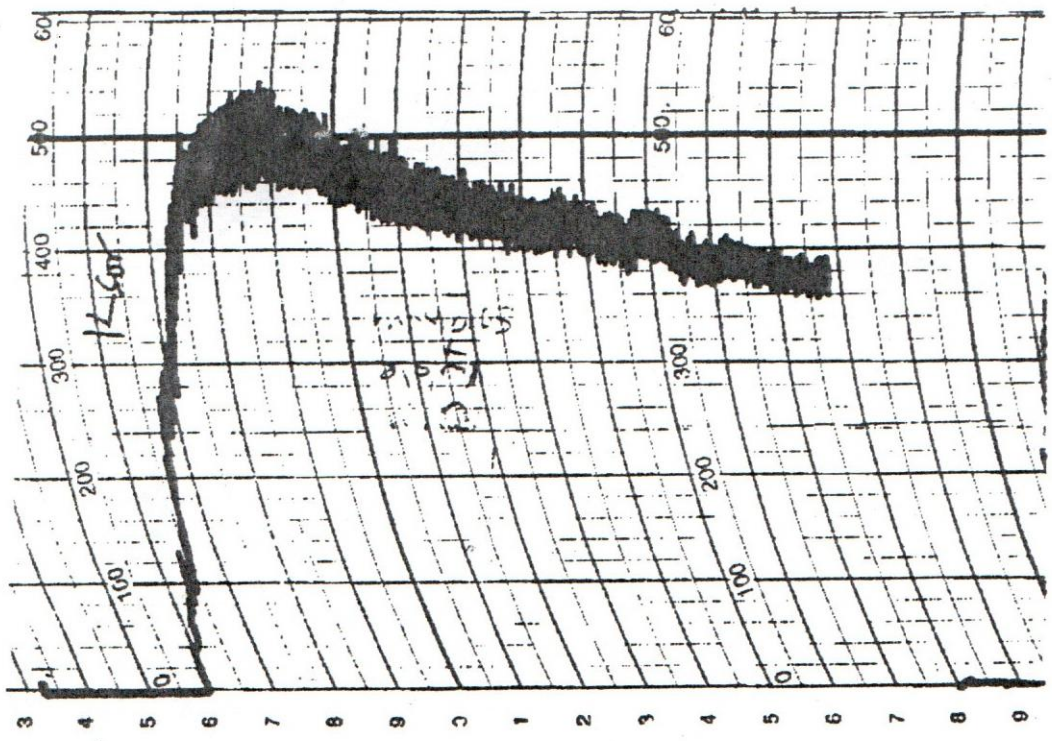
Farinograph 4: Chozi/Cowpea (ccp)



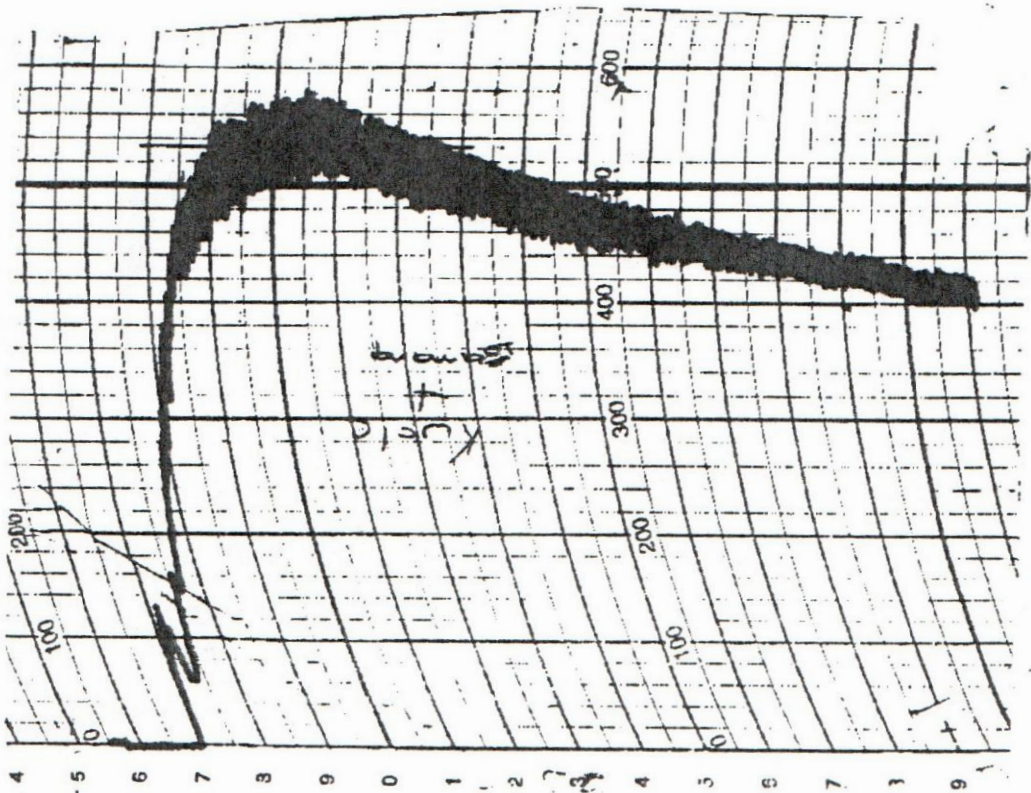
Farinograph 5: Chozi/Sorghum (csor)



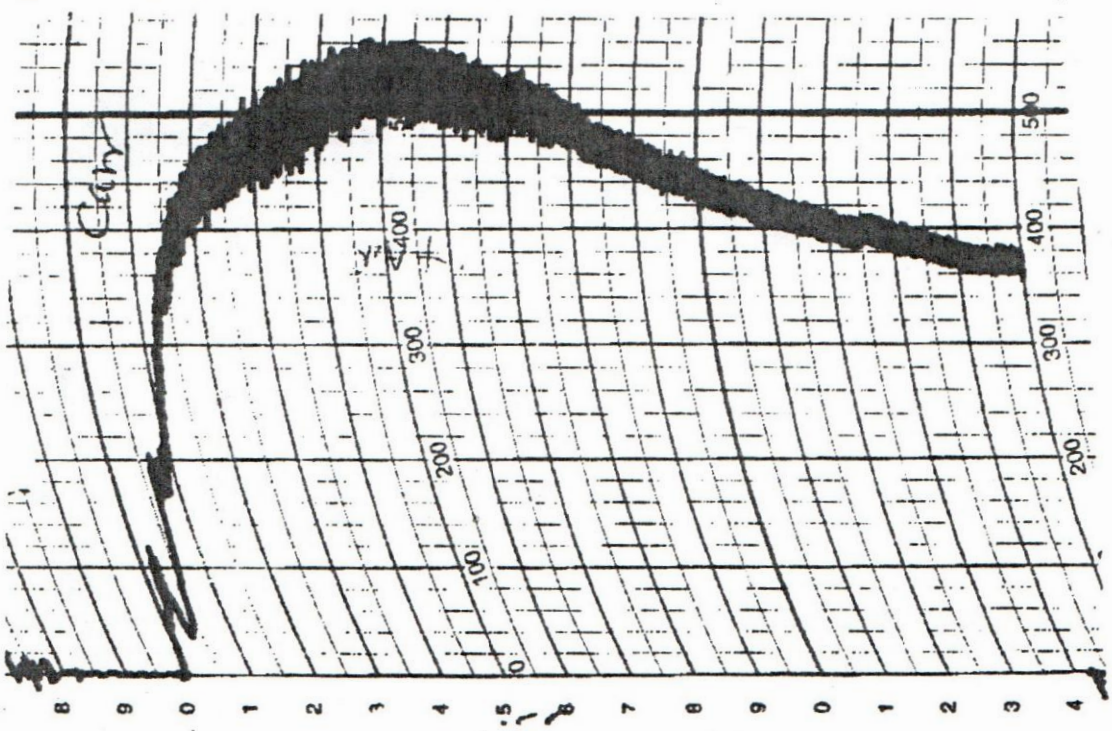
Farinograph 6: Kwale/ cowpea (kcp)



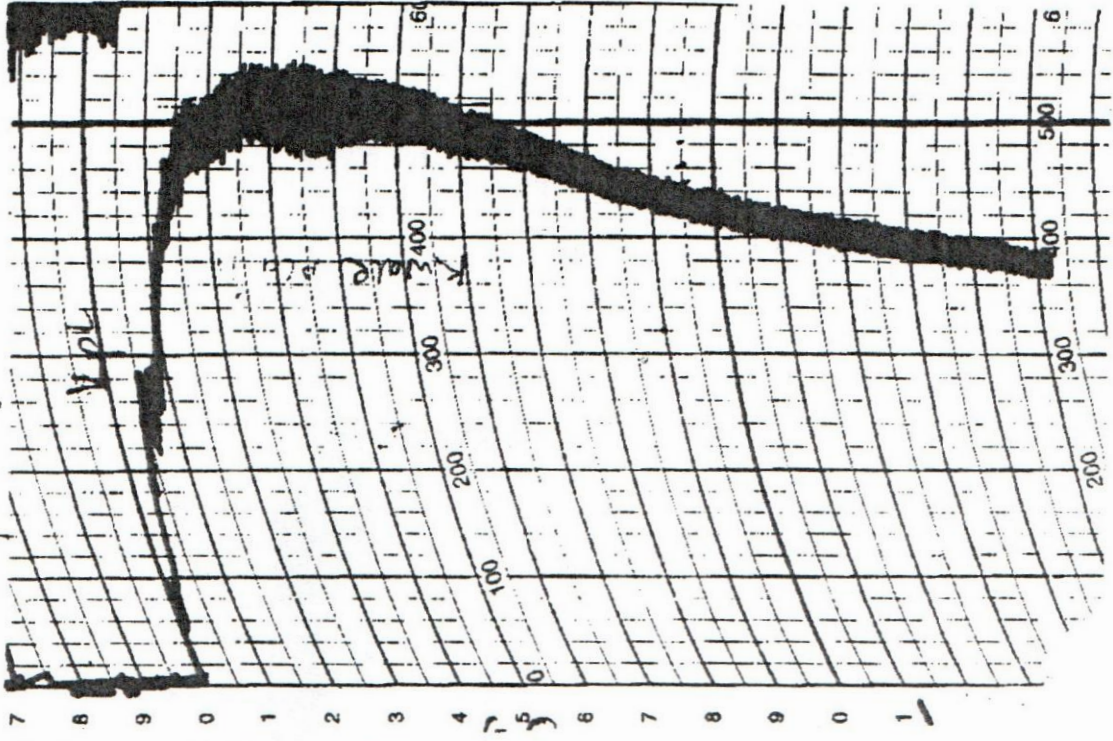
Farinograph 7: Kwale/ Sorghum (ksor)



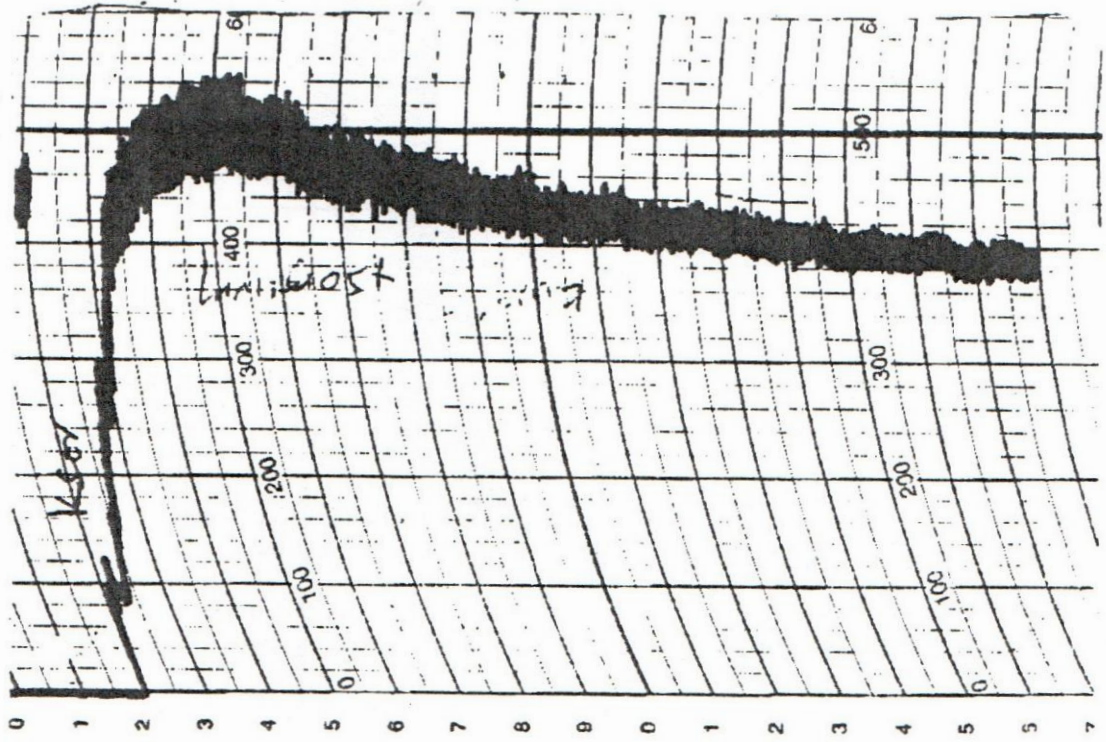
Farinograph 8: Kwale / Banana (kbn)



Farinograph 9: Chozi / Arrowroots (carr)

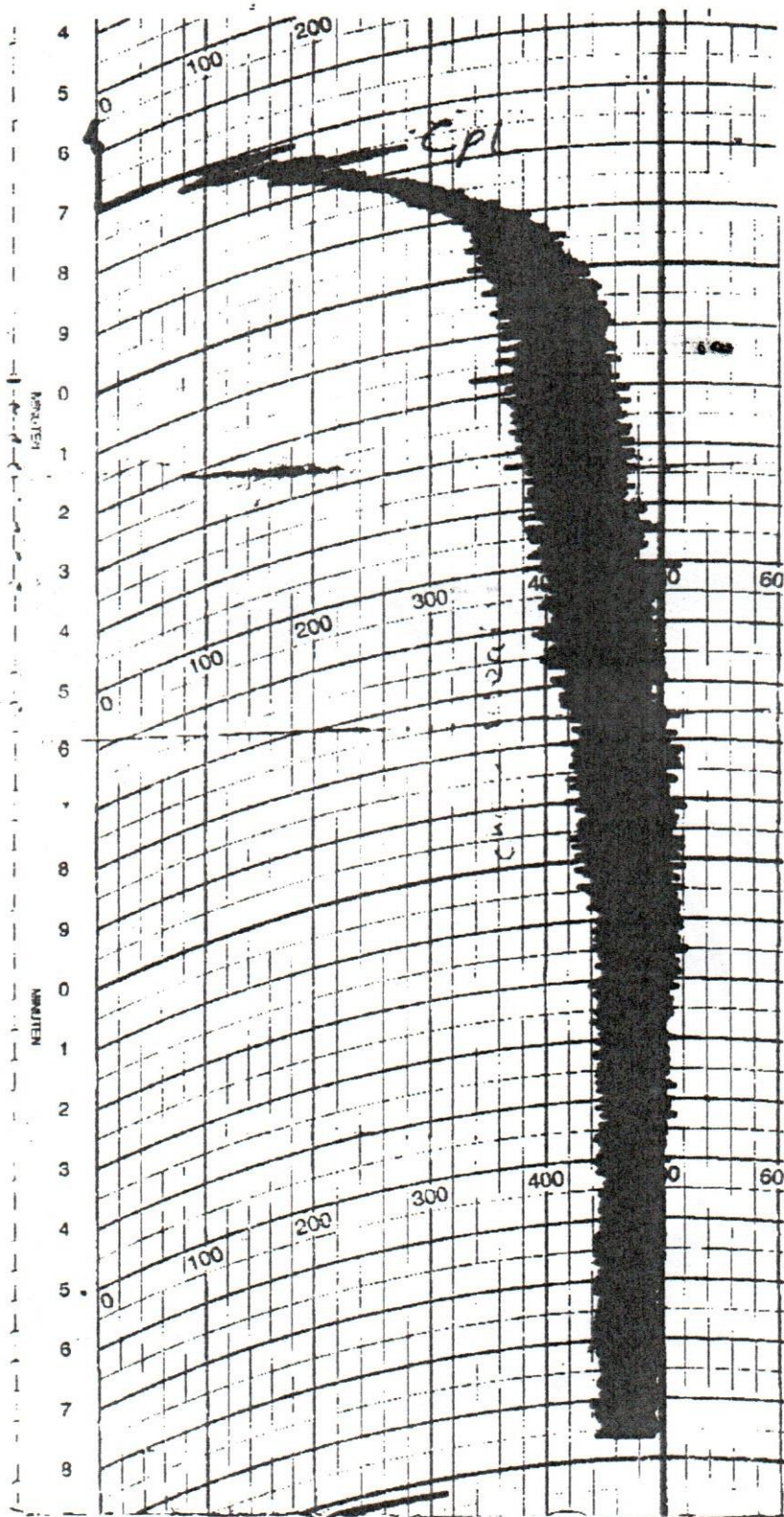


Farinograph 10: Kwale (kpl)

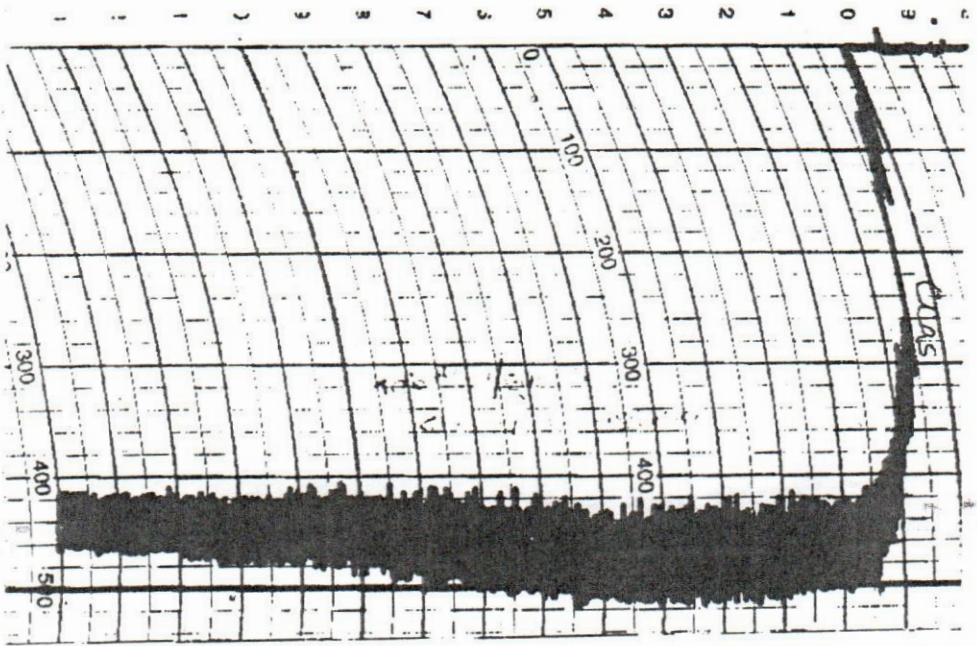


Farinograph 11: Kwale / sorghum (kSor)

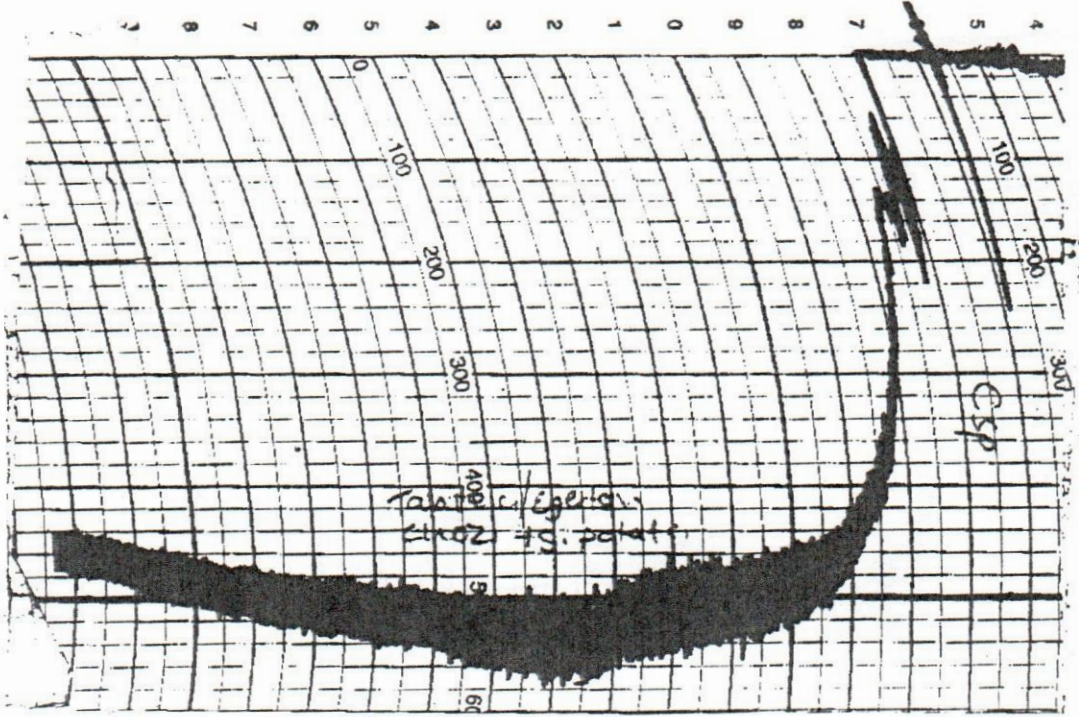
Fartnograph 12: Chozl (cpl)



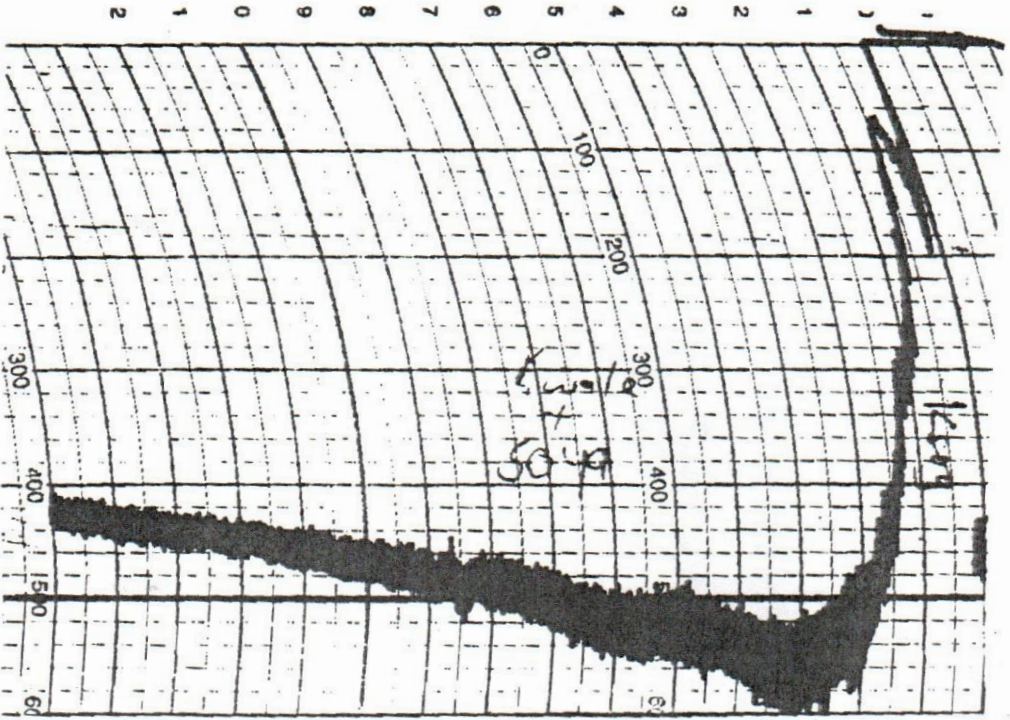
Farmograph: 14 Chozi / Cassava (ccas)



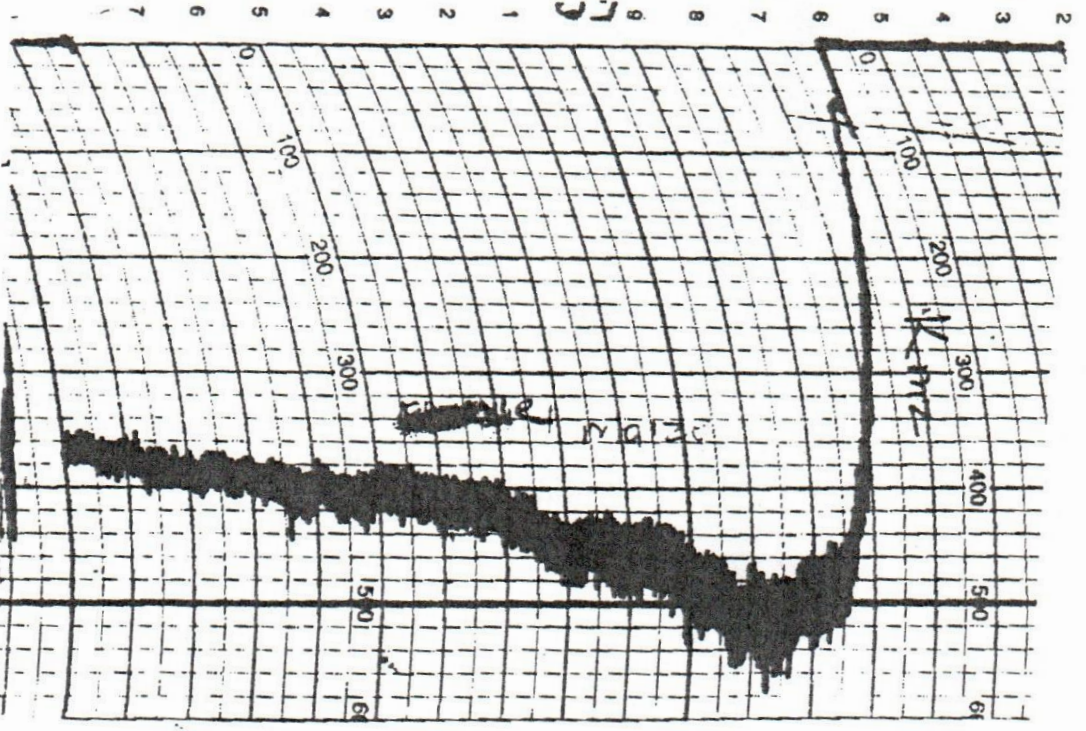
Farmograph: 13 Chozi / Sweet potato (csp)

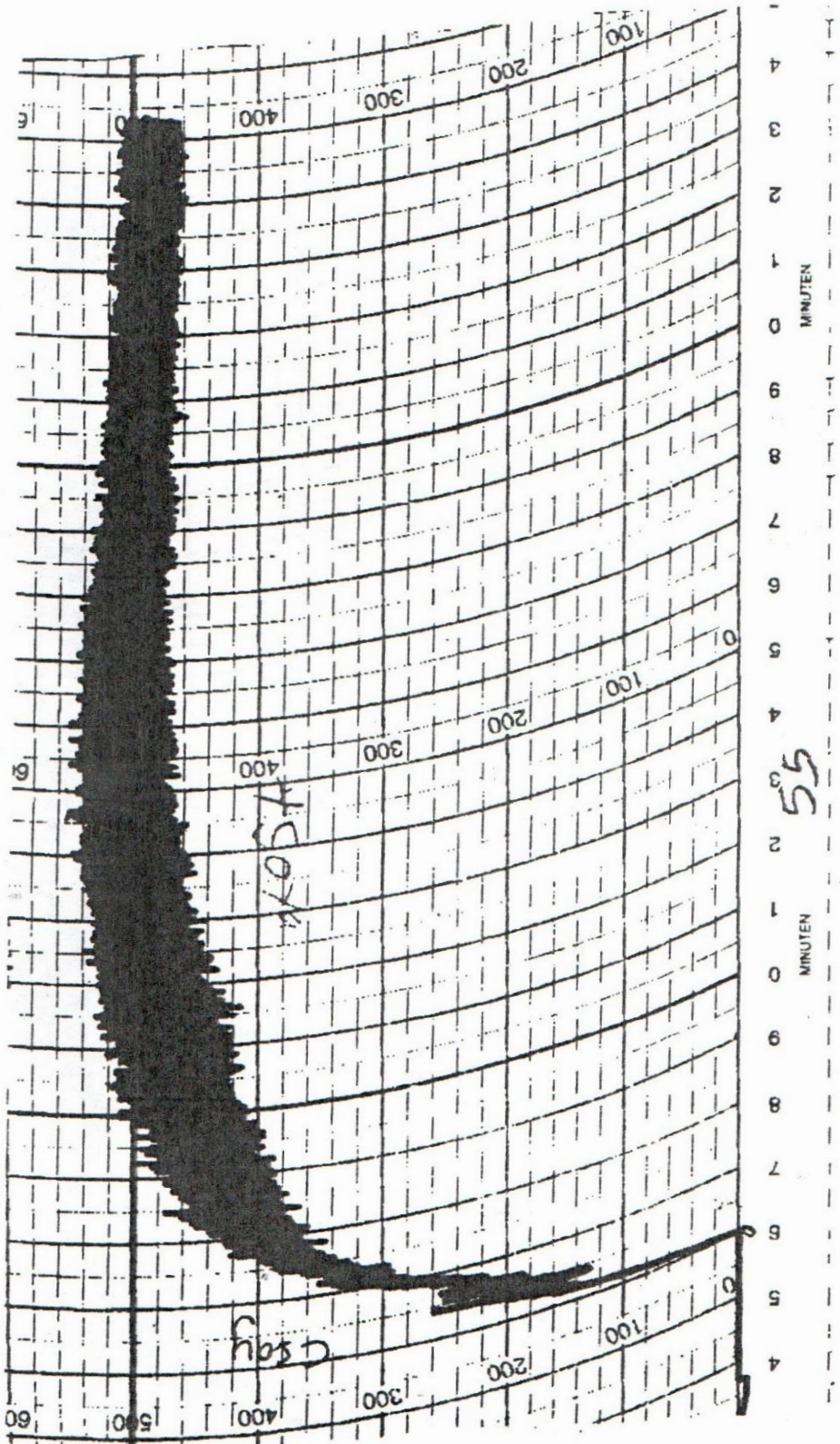


Fartnograph 16: Kwale / Soy (ksy)



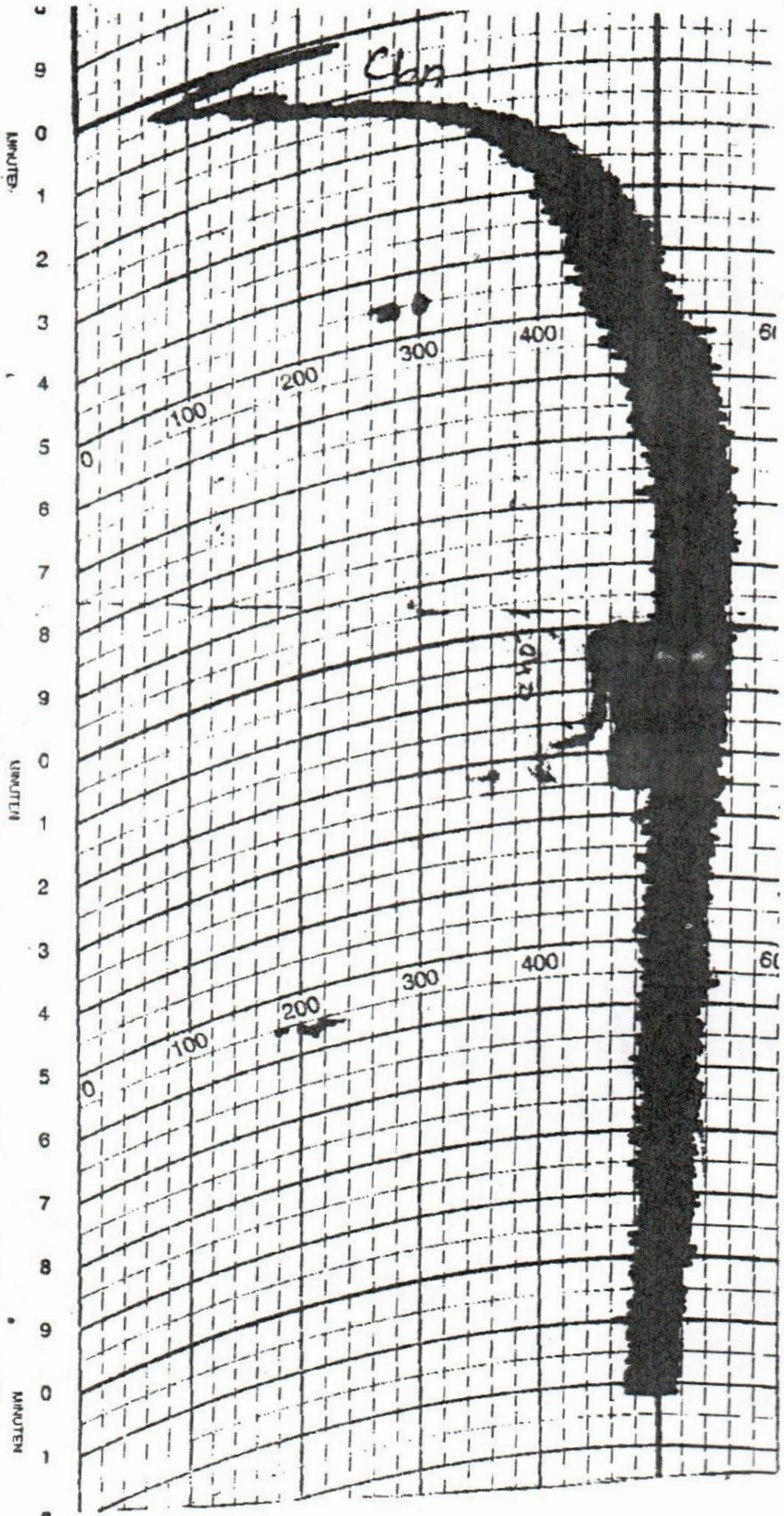
Fartnograph 15: Kwale / Maize (kmz)



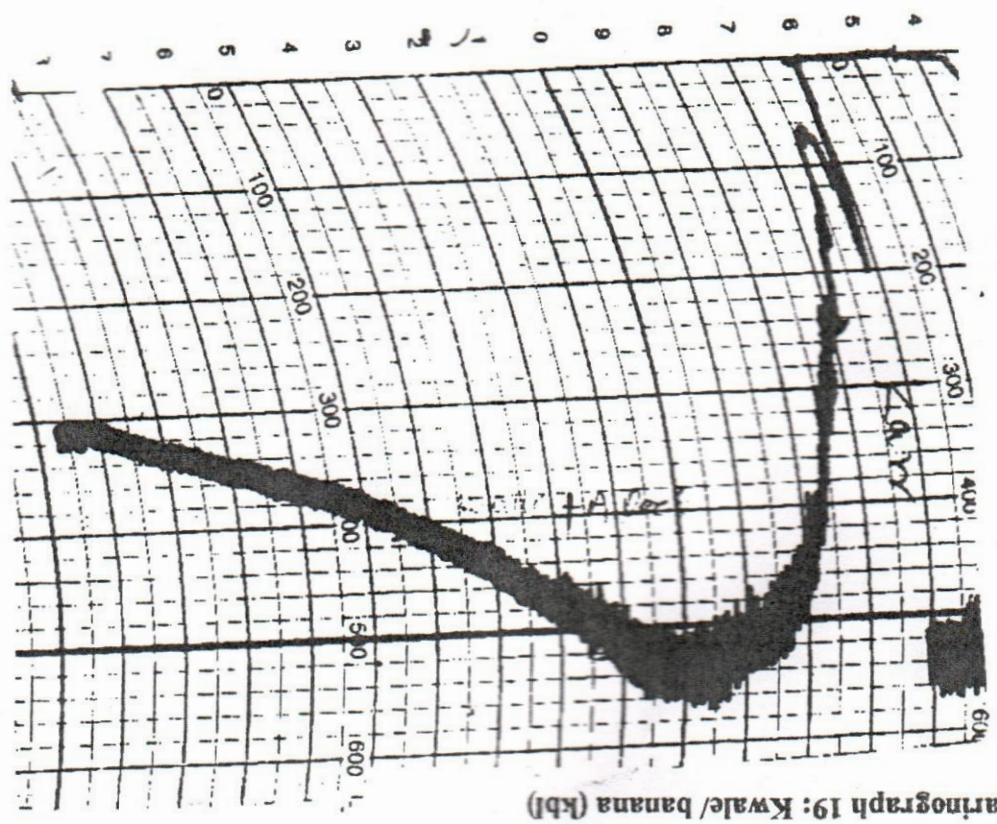


Farinograph 17: Chozi /soy (csoy)

Fathograph 18: Chozl/Banana (cbl)



Fartnograph 20: Kwale/ Aitowiroot (KART)



Fartnograph 19: Kwale/ banana (Kb)

