THE MALTING AND BREWING POTENTIAL OF A LOCAL VARIETY OF PEARL MILLET (PENNI SETUM TYPHOIDES)

A THESIS SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF MASTER OF SCIENCE

BY

MR ADEWALE OLATUNDE OLOYEDE

[B.Sc. HONS]

TO THE

DEPARTMENT OF BIOCHEMISTRY

FACULTY OF SCIENCE

AHMADU BELLO UNIVERSITY

ZARIA, NIGERIA

JULY, 1991

CERTIFICATION

This project thesis entitled Malting and Brewing Potential of a local variety of pearl millet (Pennisetum Typhoides) by Adewale Olatunde Oloyede meets the regulations governing the award of the degree of Master of Science of Ahmadu Bello University, and is approved for its contribution to knowledge and literary presentation.

Prof. C. O. Ikediobi
Professor of Biochemistry
Dept of Biochemistry
A.B.U., Zaria
[Major Supervisor]

Date

Prof. A. I. Ukoha
Professor of Biochemistry
Dept of Biochemistry
A. B. U., Zaria
[Co-Supervisor]

Date

Prof. G. H. Ogbadu
Professor of Biochemistry and
Head of Department
Dept of Biochemistry
A. B. U., Zaria

Date

Dean, Postgraduate School
A. B. U., Zaria.

2/1/92
Date

DEDICATION

To all those who persevere.

ACKNOWLEDGEMENT

I wish to express my most sincere gratitude to God and to all those who have been instrumental in one way or the other to the success of this project.

Prominent amongst these are my employer, Kaduna Polytechnic, who sponsored this programme; the project supervisor, Professor C. O. Ikediobi, who in spite of his busy schedule provided relevant reading materials, rare reagents and constructive critisms; the co-supervisor Professor A. I Ukoha and Mrs (Dr) Obagaiye for their critical appraisal of the draft copy; the Head of Department of Biochemistry, Dr. G. H. Ogbadu for his prompt attention to the incessant requests necessitated by this work; Dr. Elegbede and Mr Achoba who offered many useful suggestions regarding the sensory evaluation of the beer.

I am also greatly indebted to Mr. C.O. Ajayi, Mr. J.T. Dawodu, Mr. Ukeji; Mr. P. Okugba and the entire staff of the Food and Drug Administration Kaduna; the Research and Development Manager Jos International Breweries, Mr. Arri; the Quality Control Manager of I.B.B.I. Ltd, Kaduna; the Head of Department of Applied Science, Kaduna Polytechnic Alhaji M.B. Bappa, the Head of Department of Food Technology, Kaduna Polytechnic Mr. J.A. Egamana; Mr. H. Ade and Mr. P. C. Onyenekwe (fellow post-graduate students).

Last but not the least I wish to thank my wife Mrs Adetutu Oloyede and children Balarabe, Olufemi, Mojisola and Ikechukwu for providing me with a love nest, without which this work could not have been possible.

DECLARATION

I declare that this work is wholly the author's and has not been part of any presentation for any other qualification.

Mr A. O. Oloyede (Author) Date

Professor C.O. Ikediobi

[Supervisor]

Professor A. I. Ukoha

[Co-Supervisor]

Professor G. H. Ogbadu

[Head of Department]

TABLE OF CONTENTS

Page No

CERTIFICATION i
DEDICATION ii
ACKNOWLEDGEMENT iii
DECLARATION iv
TABLE OF CONTENTS v
ABSTRACT ix
Chapter 1 1
INTRODUCTION 1
Chapter 2 3
LITERATURE REVIEW 3
2.1 CHOICE OF LOCAL RAW MATERIAL FOR THE MALTING INDUSTRY IN NIGERIA 3
2.2 MILLETS: ORIGIN AND DISTRIBUTION 4
2.3 THE MILLET KERNEL 5
2.4 CHEMICAL COMPOSITION 6
2.5 ENZYME ACTIVITY 7
2.6 MALTING 8
2.7 UNIT OPERATIONS IN MALTING 8
2.7.1 GRAIN ASSESSMENT 8
2.7.2 GRAIN CLEANING 10
2.7.3 STEEPING 10
2.7.4 GERMINATION 12
2.7.5 KILNING 15
2.8 ASSESSMENT OF MALT QUALITY 17
2.9 SURVEY OF MALTING ACTIVITY IN NIGERIA 18
2.10 BREWHOUSE PROCESS 19
2.10.1 MILLING 19
2.10.2 MASHING 21
2.10.3 WORT PRODUCTION 24
2.10.4 WORT FERMENTATION 26
2.10.5 SENSORY EVALUATION 28
Chapter 3 31
3. MATERIALS/REAGENTS 31
3.1 MATERIALS/REAGENTS 31

Chapter 3 31
3. MATERIALS/REAGENTS 31
3.1 MATERIALS/REAGENTS 31
3.2 SAMPLE PRETREATMENT 31
3.2.1 CLEANING: 31
3.2.2 MILLING: 31
3.2.3 PARTICLE SIZE ANALYSIS: 31
3.3 MALTING PROFILES OF MILLET GRAIN 32
3.3.1 DETERMINATION OF PERCENTAGE FLOAT 32
3.3.2 DETERMINATION OF PERCENTAGE OF FOREIGN MATTER AND BROKEN KERNELS 32
3.3.3 DETERMINATION OF GERMINATION ENERGY 32
3.3.4 DETERMINATION OF GERMINATION CAPACITY 33
3.3.5 DETERMINATION OF ONE THOUSAND KERNEL WEIGHT 33
3.3.6 MOULD DETECTION 33
3.4 PHYSICAL AND BIOCHEMICAL ANALYSIS OF MILLET GRAIN 34
3.4.1 MOISTURE DETERMINATION 34
3.4.2 PROTEIN DETERMINATION 34
3.4.3 FAT DETERMINATION 34
3.4.4 STARCH DETERMINATION 35
3.4.5 TOTAL FREE AMINO ACID: 36
3.4.6 DETERMINATION OF POLYPHENOL 36
3.4.7 ESTIMATION OF TANNIN CONTENT 37
3.4.8 PROTEOLYTIC ACTIVITY: 37
3.5 EFFECT OF DURATION OF STEEPING ON GERMINATION ENERGY 38
3.6 PRODUCTION OF MILLET MALT AT VARYING DURATION OF STEEPING . 38
3.7 ANALYSIS OF MILLET MALT 38
3.7.1 MOISTURE: 38
3.7.2 DETERMINATION OF MALTING LOSS 39
3.7.3 DETERMINATION OF HOT WATER EXTRACT 39
3.7.4 DETERMINATION OF COLD WATER EXTRACT (CWE) 40
3.7.5 DETERMINATION OF DIASTATIC POWER 40
3.8 DETERMINATION OF ALPHA AND BETA AMYLASE ACTIVITIES AT VARYING DAYS OF GERMINATION 41
3.9 EFFECT OF GERMINATION DURATION (DAYS) ON MALT QUALITY 42
3.10 EFFECT OF ADDITIVES ON MALT QUALITY 42

Chapter 4 45
RESULTS AND DISCUSSION 45
4.1 QUALITY ASSESSMENT OF UNGERMINATED MILLET 45
4.2 GERMINATION STUDY/MALT QUALITY (MILLET AS AN ALTERNATIVE FOR BREWING) 48
SUGGESTIONS FOR FURTHER WORK 74
Chapter 5 75
REFERENCES 75
TABLE 1: DATA ON PEARL MILLET GRAINS (PROXIMATE, PHYSICAL AND BIOCHEMICAL PROPERTIES) 47
TABLE 2: RESULTS OF CHI-SQUARE ANALYSIS 61

LIST OF FIGURES

FIGURE I: CHANGES IN ALPHA AND BETA-AMYLASE ACTIVITY DURING MALTING 49
FIGURE II: CHANGES IN PROTEOLYTIC ACTIVITY DURING MALTING (i.e GERMINATION) 50
FIGURE III: NUTRIENT CHANGES DURING MALTING OF A LOCAL VARIETY OF MILLET 51
FIGURE IV: MOISTURE PERCENT VERSUS STEEPING TIME (HRS) 53
FIGURE V: STEEPING DURATION VERSUS GERMINATION ENERGY (%) 53
FIGURE VI: MALTING LOSS VERSUS STEEPING DURATION 54
FIGURE VII: STEEPING DURATION VS COLD WATER EXTRACT 55
FIGURE VIII: HOT WATER EXTRACT VERSUS STEEPING DURATION 56
FIGURE IX: WORT FILTRATION TIME VS STEEPING DURATION 58
FIGURE X: PATTERN OF MALT CHARACTERISTICS OF A LOCAL MILLET VARIETY 59
FIGURE XI: EFFECT OF ADDITIVE (IN STEEPING LIQUOR) ON MALT XTERISTICS 63

LIST OF APPENDIXES

APPENDIX 1: STANDARD CURVE FOR FREE AMINO ACID ESTIMATION 64
APPENDIX 2: STANDARD CURVE FOR POLYPHENOLS ESTIMATION 65
APPENDIX 3: STANDARD CURVE FOR MALTOSE 66
APPENDIX 4: STANDARD CURVE FOR TANNINS 67
APPENDIX 5: EFFECT OF STEEPING DURATION ON MALT QUALITY 68
APPENDIX 6: EFFECT OF ADDITIVES ON MALT QUALITY 68
APPENDIX 7: RESULT OF PROXIMATE ANALYSIS OF MILLET MALT USING DIFFERENT ADDITIVES AS STEEPING LIQUOR 69

APPENDIX 8: EFFECT OF GERMINATION DURATION ON MALT QUALITY 69
APPENDIX 9: PROXIMATE ANALYSIS OF MILLET MALT AT VARYING GERMINATION DURATION 70
APPENDIX 10: ORGANOLEPTIC ASSESSMENT OF LAGER BEER 71
APPENDIX 11: RESULT OF THE SENSORY EVALUATION OF LAGER BEER (CONTROL ie. STAR LAGER BEER) 72
APPENDIX 12: RESULT OF THE SENSORY EVALUATION OF MILLET LAGER BEER 72
APPENDIX 13: CHI-SQUARE ANALYSIS OF THE RESULTS OF SENSORY EVALUATION 73

ABSTRACT

The malting and brewing potential of a local variety of pearl millet (LPMV) was studied. Quality assessment of ungerminated grain indicated absence of mould and insect infestation and gave 0.6% foreign matter, 3.6% float an average one-thousand kernel weight of 7.83g, a germination energy of 66.7%, germination capacity of 69.3%, cold water extract of 2.32 lit. Deg. Kg^{-1};Diastatic power of $4.56^{O}L$ B-amylase activity of 55 µg maltose/ml/min, -amylase activity of 45 µg maltose/ml/min, proteolytic activity of 184.55 gm/100 1/07 µg phenol/100g and 2.48% tannin. Proximate analysis of the ungerminated grain gave 10.03% moisture, 1.87% Ash, 4.25% Fat, 66.27% Starch 11.06% protein and 0.0075% reducing sugars.

ß-amylase activity rose during grain germination reaching a peak of 220 Ug/ml on the fourth day and thereafter declined. α-Amylase activity on the other hand, rose progressively throughout the seven day germination period with an activity of 750 Ug/ml on the seventh day as compared to 45 µg/ml in ungerminated grain. Proteolytic activity rose from 184.55 mgN/100g in dormant grain to a peak value of 572.73 on the fourth day of germination.

Although millet malt produced by steeping grains for 6 hours gave the highest hot water extract of 171.38 Lit. Deg. Kg^{-1},the highest diastatic power of $18.65^{O}L$was obtained after 18 hours of steeping. The highest cold water extract of 7.5% was obtained for grains steeped for 24 hours . The inclusion of different additives affected the malt characteristics significantly. $Ca(OH)_2$(0.2%) produced the best effect followed by hydrogen peroxide. Grain steeped for 24 hours and germinated for 5 days with 0.1% $Ca(OH)_2$in steeping liquor gave the best malt characteristics, except for the high malting loss (28.4%).

Lager beer produced from millet malt was well accepted in terms of the brilliance, colour, bitterness, bodyfulness, odour and sourness ($P > 0.05$). However, a few attributes of the beer such as foam stability and sparkling were not accepted ($P<05$). It is implied that the use of millet malt for lager beer production in the presence of exogenous enzymes is technically feasible.

Chapter 1

INTRODUCTION

The ban on barley importation by the Federal Government in 1988 and the structural adjustment policy has changed the pattern of raw material utilization in the brewing and allied beverage industries. The brewing industry witnessed a phenomenal growth between 1977 and 1983, rising from 8 to 33 breweries. Concurrently, the production capacity of the industry rose from 2.72 million hectolitres in 1970 to 24 million hectolitres per annum in 1983 (Beer Sectoral Group, 1986). This resulted in a proportionate increase in barley malt importation into the country. Thus the import bill required to support this huge barley importation was enormous relative to the country's foreign exchange position. It has been estimated that the industrial sector requires a staggering ₦5.1 billion per annum in foreign exchange for malt and enzyme importation to meet their annual malt and enzyme supply of 450,000 and 7,500 metric tonnes respectively (Ikediobi, 1989). This experience helped a great deal to fuel research efforts into the local souring of industrial raw materials which has now become a national policy.

Unlike barley which is not cultivated or grown naturally in Nigeria, cereal grains such as sorghum (guinea corn), maize and millet are cultivated in abundance. The estimated annual production of sorghum is 3.8 million tonnes, millets, 3.2 million tonnes, maize 2 million tonnes and milled rice 0.9 million tonnes (Nwasike and Aba, 1986). It has been estimated that sorghum and millet alone account for 70% of the total area under cereal cultivation in Nigeria (Obigbemi, 1987).

The use of sorghum for beer production is well documented. As far back as 1943, sorghum beer was produced in the United States. The poor yield and bitter taste of the early sorghum beer has been attributed to the use of cracked and partially dehulled sorghum (Canale, et al., 1976). However, modern brewing techniques have changed this situation drastically and acceptable sorghum beer has been produced at Laboratory and industrial level from 100% sorghum (Ogundiwin, 1989). In fact most breweries in Nigeria now use over 60-100% local raw materials such as sorghum grain and grits, maize grits and sorghum malt for beer production (Chukwura, 1988).

Kashim Ibrahim Library,

The problems associated with sorghum malt are quite diverse. They include mould growth, cyanogenesis high malting losses, insufficient modification, uneven germination, high polyphenol-tannin levels, (Ikediobi, 1989) and the high gelatinization temperature of sorghum starch (Arri, 1989). The absence of husk in sorghum has also been reported to pose filtration problem when lauter tun is used for mash filtration (Chukwura, 1989). Furthermore, the high gelatinization temperature of sorghum starch accounts for the poor modification of sorghum and the need for expensive exogenous enzymes when sorghum malt is used for beer production (Ikediobi, 1989). It is obvious from the foregoing, that research efforts should be intensified to perfect the use of sorghum and other potentially suitable local cereals, particularly millet, not only for the production of brewers malt or distiller's malt but also for speciality malt. (Pomeranz, 1975).

Though the small size of millet (½ to 1/3 the size of sorghum) raises some technical problems in large scale malting, both pearl and finger millet can give diastatic activities equivalent to sorghum (Hulse, et al., 1981). Literature indicates that certain millet varieties are very high in malting or modification enzymes compared to sorghum in which the diastatic power is only about one third to half of barley (Ikediobi, 1989).

Also the starch content of millet is comparable to that of sorghum and barley. Though the relatively high content of protein, fat and probably tannin (Aisen, 1989, Pomeranz, 1975) are disadvantageous in terms of its possible uses for the production of brewers or distillers malt, the development of an appropriate malting technology could reduce their levels in millet malt. The use of millet malt in 'beer' production has been reported in India and other countries (Ikediobi, 1989). However, little work has been done to determine what physical and chemical factors are related to grain quality and millet consumers can only subjectively distinguish between 'good' and 'poor' quality grains for particular endues (Briggs et al., 1981).

Therefore, the objectives of this study is the quality assessment of millet and millet malt produced under defined set of conditions coupled with the organoleptic assessment of lager beer brewed from the millet malt which gives the best characteristics. These findings could suggest directions to which future research efforts should be geared to meet the raw material specifications for millet malt for lager beer production.

Chapter 2

LITERATURE REVIEW

2.1 CHOICE OF LOCAL RAW MATERIAL FOR THE MALTING INDUSTRY IN NIGERIA

The use of tropical crops such as maize, millet and sorghum or rice has been suggested as substitute for barley (Olaniyi, 1984). The possibility of producing malting barley locally has also been reported (Asenime, 1985).

The main problem with maize, rice, sorghum and millet is their low level of enzymes which necessitates the use of exogenous enzymes for mash break down. Barley on the other hand contains sufficient enzymes that can be substituted only up to 50% with local grains without the use of exogenous enzymes. However, for 100% substitution with sorghum, a combination of exogenous enzymes, namely, beta-glucanase, proteinase and maltogenic fungal amylase are required in large quantities (1 Kg/100 Kg of sorghum) to arrive at the correct wort composition (Sugris and Hope, 1987; Aisen, 1988).

It has been demonstrated that high quality malting barley can be produced in Nigeria if suitable barley cultivars are grown on a well irrigated nutrient - rich soils, preferably deep sandy clay loames in the coldest period of the year (15th November to 1st December); (Sugris and Hope, 1987). They reported the successful cultivation of 50ha of barley on an experimental farm in Lake Chad Basin by Nigerian breweries Ltd. and the Guinness Nigerian Ltd, during the 1983/84 dry season, and at two other experimental farms in Savannah and Kaduna with a grain yield of 1192Kg/ha respectively. None-the-less, the writers questioned the economic feasibility of barley production in Nigeria and predicted that Nigeria lacks the capability to produce enough barley to meet her requirement for the next seven years.

Haase, 1985 demonstrated that with appropriate modification of the equipments used in barley malt production, sorghum malt which can be used to brew a normal European lager beer without addition of artificial enzymes can be produced. Haase's sorghum malt lager beer gave a high acceptability score of 3 compared to 3.5 for the German export lager beer when graded by professional beer tasters. As indicated in the introduction some Nigerian workers have also reported the use of 100% sorghum for the production of lager beer without the use of exogenous enzymes.

Thus most breweries in Nigeria have opted for the use of local cereals either as malt or adjunct. Most current research efforts are directed at the production of sorghum cultivars with a high diastatic power, low polyphenol, soft endosperm, loosely packed starch granules and low rate of cyanogenesis (Ikediobi, 1989).

Since the choice of sorghum is based largely on the relative food use of sorghum compared to maize or rice, there is need for intense research into the feasibility of millet malt production because of its availability, and promising biochemical potential.

2.2 MILLETS: ORIGIN AND DISTRIBUTION

Millets are small-seeded annual cereal grasses many of which are adapted to hot climates. The species includes the following genera pearl millet (Pennisetum typhoides) Proso millet (pennicum milliaceum), foxtail millet (setaria italica), finger millet (Eleusince coracana), the Japanese barnyard Echinochoa crusgallivar frumentaca) and the koda millet of India (Paspallum scrobiculatum) (Hulse, et al., 1980).

The pearl millet is the most important of all the millets and miscellaneous cereals. It is commonly known as bulrush, pearl, cattail, spiked millet (English), bajra (Hindi), kambu (Tamil), dukin (Arabic) and mila' chandelles (West and North Africa). The millets commonly grown in Nigeria are Geromaiwa and Dauro. They are given special names based on where they are grown. Thus 'Ex-Benue', 'Ex-Riyom' refer to those grown in Borno, Benue and Plateau State respectively Eghgrevba 1979).

The millets represent the most important food crops, second only to sorghum, in the semi-arid tropics. Their distribution covers countries surrounding the Sahara, many parts of East-Africa, a large area of central India and some regions of South-east Asia and South America (Hulse, et al., 1980). In Nigeria the millets are grown in twelve states of the Federation, namely; Bauchi, Benue, Borno, Cross River, Gongola, Kaduna, Kano, Katsina, Kwara, Niger, Plateau and Sokoto state (Obigbemi, 1987). However, in Nigeria most millets are grown mixed by farmers (Egharevba, 1979).

2.3 THE MILLET KERNEL

The shape of the millet kernel varies from globose to subcylindrical to conical. They are usually ashgray although varieties having purple, greyish brown, yellowish-brown or amber white colour are also known. The size of the millet kernel is about one-third to half the size of the sorghum kernel (which ranges in size between 8 - 35 mm in diameter). Briggs, et al., 1981.

The kernel is composed of three major portions, the outer covering or pericarp, the storage tissue or endosperm and the germ or embryo. As in sorghum, the pericarp consists of three distinct layers: The epicarp, mesocarp, and endocarp. The husk is absent in both sorghum and millet, unlike in barley. The significance of this is brewing has been mentioned. Millet pericarp is thicker and denser than that of sorghum and each of the layers of the pericarp comprises of 2 cell layers. The endosperm comprises two distinct layers; The outer or aleurone layer of the endosperm, extends around the entire cell. There is no testa between the aleurone layer and the endocarp as in sorghum and barley, although a thin tissue membrane is present. The starchy endosperm next to the aleurone layer consists of a peripheral corneous and a floury region, both of which contain starch granules embedded with protein matrices which contain protein bodies (Hulse, et al., 1980).

2.4 CHEMICAL COMPOSITION

Millet comprises 8.9 - 19.62% protein, 59-65% starch, 2.0 - 6.5% fat 1.7 - 3.7% Ash and 2.0 - 2.7% total sugars as compared to sorghum which comprises 9.5 - 12.5% protein, 71.73% starch, 2.8 - 3.5% fat, 1,2-4.4 Ash or barley which comprises 8-13% protein, 63-65% starch, 2-3% fat, and 1.55-2.65% Ash. (Aisen, 1989, Hulse et al., 1980, Nwasike et al., 1986, Pomeranz, 1975).

The proteins of pearl millet consists of 43% prolamine (ie, the 70% ethanol extract) and 20% globulin. The protein bodies are largest in peripheral endosperm and decrease's towards the centre (Hulse et al 1980). The starch granules are smaller compared to those of sorghum and vary in shape from polygonal (in the corneous zone of the endosperm) to spherical. Millet starch has a high amylopectin content, consisting of 68% amylopectin and 32% amylose (Purseglove, 1972). High amylopectin is undesirable in a malting grain. The difficulty of enzyme hydrolysis of sorghum starch has been attributed to the high amylopectin content (Twagirmukiza, 1983), and this causes wort filtration problem (Subramanian and Jambunathan 1989).

The millets are rich in potassium, magnesium and phosphorous (Nwasike and Aba, 1986). The level of iron varies from 26 - 153 mG/100g depending on variety. Some millet varieties also have a high calcium content (Indera, et al., 1981). They also contain different vitamins. A vitamin level of 0.19mg/100g of thiamine, 2.5mg/100g of riboflavin and 0.19mg/100g of niacin has been reported for certain millet varieties (Hulse et al., 1980).

The colour of the millet kernel is due to the presence of phenolic compounds in the peripheral endosperm region of the kernel. The major one being the glucosyl flavonoide (590mg/100g) (Hulse et al., 1980). High polyphenol grains are considered unsuitable for brewers and distillers malt. This is due to the polyphenol-protein interaction which results in the formation of precipitates ('breaks' or 'trubs') during wort boiling and also the formation of nonbiological haze (Briggs et al., 1981). The first step in the mechanism whereby phenolic compounds attach themselves to protein is the oxidation of the phenol to a highly reactive quinone. For example 0-benzboquinone reacts with the thioester group of methionine to yield 3,4 - dihydroxyphenyl - N - acetyl methionine sulphonyl - chloride (Vithayathil, et al., 1972). High polyphenol is also inhibitory to brewing enzymes (Dalber, 1975).

Another important class of compounds found in millets are the tannins. The word 'tannin' originally applied to concentrates of vegetable polyphenols used to treat hide, so tannin it and converting it to leather (Briggs, et al., 1981). The more polymerized polyphenols have higher tannin ability than the simpler ones.

The seed colour also varies with tannin levels. The tannin content declines as the seed colour changes from brown to red, to yellow and to white. Tannin content was also found to correlate negatively with posts-harvest moulding and is apparently related to germination and grain resistance to disease (Hulse, el al., 1980). The tannin and polyphenol level has been correlated with invitro digestibility of millet (IVDP). Thus low tannin cultivars showed high in-vitro digestibility and vice-versa (Hulse et al., 1980). Ramachandra et al., 1977, whowed that polyphenol and tannin levels in certain African and Indian millet cultivars ranged from 0.54 to 2.44 and 0.50 to 3.47% respectively. Tipton 1974 gave a range of 2.36 to 7.25% for certain millet hybrids analysed.

A critical evaluation of most of the methods used in the past for tannins indicate that polyphenols were mistaken for tannins (Maxson and Rooney, 1972). The method which showed the least variation amongst the samples analysed by these workers was the one based on the formation of a coloured complex with ferric ammonium sulphate after extraction with urea.

2.5 ENZYME ACTIVITY

Whereas B-amylase is present in ungerminated cereals, ∝-amylase and protease are practically absent. A low level of glycerol -, and pyrophosphatase were also reported in certain millet varieties before germination (Hulse, et al., 1980). The presence of other enzymes such as dextrinase, -fucosidase has been reported (Pankrashkina and Zherebson, 1983).

The diastatic power of ungerminated millet also vary widely with variety. Chandrasekara, et al., 1939 (as cited by Hulse, et al., 1980) reported low diastatic power in certain varieties. However millet varieties with high diastatic power such as SIE composite (16.6°L) and Hairy composite (10°L) SDU, Ex-Borno, 18.35°L) have been reported (Novellie, 1959). Another important class of enzymes in malting grains are the ß-glucanases. These hydrolyse ß-glucans to glucose. Since ß-glucosan contributes to wort filtration problem, malt with low ß-glucanase activity could give wort filtration problem, if exogenous ß-glucanase is not used during mashing (Briggs et al., 1981).

2.6 MALTING

Malting is the controlled germination of grain; under a well defined set of conditions (of moisture, aeration and temperature) sufficient to cause limited degradation of the endosperm and to mobilize the enzymes (Olaniyi, 1984). Malt is therefore a cereal grain which has been germinated and then dried under a defined set of conditions (Briggs, et al., 1981).

Germination is therefore the consecutive number of steps which cause a quiescent seed with a low water content to show a rise in its general metabolic activity and to initiate the formation of seedling from the embryo (Mayer and Poljakoff 1963).

2.7 UNIT OPERATIONS IN MALTING

The operations involved in malting include grain assessment, grain cleaning, steeping, germination, kilning or drying sprout removal and packaging. (Ikediobi, 1989; Briggs, et al., 1981; Palmer and Pathgate, 1976).

2.7.1 GRAIN ASSESSMENT

The parameters for assessing the suitability of malting grain are quite diverse. They include the determination of one thousand kernel weight, tests for broken kernel and foreign matter, germination energy and capacity; floatation, insect and mould infestation. Physical and biochemical tests carried out on malting grains include hot and cold water extract, α- and ß-amylase, ß-glucanase, diastatic power, ß-glucans and pentosan estimation. (Ikediobi, 1989; Aisen, 1989; Briggs, et al., 1981).

The one thousand kernel weight gives the maltster precise information on the grain size. It was originally used for barley study. Its use has however been extended to cover the study of other cereal grains such as sorghum and millet. Adrian and Sayerse, 1957, established that the average one thousand corn weight of pearl millet grown in Senegal is 6.5. It has been reported also that amongst ICRISAT lines the 1000 corn weight for millet ranged from 2.5 to 14.0g with a mean of 7.96g (Hulse, et al., 1980).

The germination energy measures the proportion of the seed that will germinate under the test condition while the germination capacity measures the capacity to germinate. The difference between the two tests is a measure of the dormancy of the grain sample. (Briggs, et al., 1981). It has been demonstrated that a malting grain should have a viability or germinative capacity of at least 96% for barley and 95% for sorghum (Palmer and Pathgate 1976; Ikediobi, 1989). However, grains giving low values in germination tests could often be malted on a pilot scale. Therefore, tests closer to commercial practice were devised. These include the use of small-scale steeps involving test-tubes (micro-malting) or by tests in which 0.6-1.0 Kg grain samples held in nylon net bags and steeped and germinated in contact with a large bulk. The values obtained from these tests, though higher, still represent a pessimistic estimate of the samples readiness for malting (Briggs, et al., 1981).

The floatation test determines the percentage of immature seeds. Immature seeds are not viable and constitute a loss to the maltster (Ikediobi, 1989).

The presence of mould is evidenced by grain discolouration and endosperm destruction, hence the mould infestation test. According to Ikediobi, (1989), most of the mould species encountered during malting are either field derived or picked up in ware-houses during storage. In sorghum malting in Nigerian the presence of mould is usually determined by visual inspection, since signs of fungal contamination can easily be seen. Insect infestation also destroys the endosperm and reduces grain viability.

The higher the percentage of broken kernels and foreign seeds, the less the suitability of a particular variety of grain for malting, since unwanted grain will not produce the same kind of malt. (Briggs, et al., 1981). A high percentage of broken kernel is also suggestive of poor post-harvest handling and is generally associated with insect and rodent attack (Ikediobi, 1989).

Moisture determination in a malting grain is very important since the germination rate and modification intensity of grains are controlled by regulating the moisture content and temperature. Besides the storage stability of the grain is a function of moisture (Briggs et al., 1981).

Malting grains are also required to possess certain physical and biochemical characteristics such as low fat, tannin and polyphenol content as well as high level of starch, soluble protein and extract yield (Aisien, 1989). According to Ikediobi (1989), the desired properties for an ideal sorghum required for malting purpose includes low gelatinization temperature of the starch (65 - 68°C), soft endosperm, loosely packed starch granules, easily assessable protein bodies in association with starch in the endosperm, high endogenous ß-amylase, low content or complete absence of polyphenol, high protease and ß-glucanase activity and a low rate of cyanogenesis.

2.7.2 GRAIN CLEANING

After grain reception and storage which require a thorough grain assessment as described, the first unit operation in malting is grain cleaning. The grain is cleaned with water to separate the dirts, including sticks, stalks, stone/pebbles, sand, dust, papers/leaves and other extraneous matter. All these dirts carry micro-organisms that would render the malt useless if not removed. The cleaned grains are then graded to different sizes. This is very important, since lack of uniformity in grain size results in uneven germination (Palmer and Pathgate, 1976; Ikediobi, 1989; Briggs et al., 1981).

2.7.3 STEEPING

A well worked out steeping schedule is very essential for each grain variety to ensure adequate modification of grain without at the same time incurring heavy malting losses (Ikediobi, 1989).

When grain is immersed in water, its moisture content increases rapidly at first but progressively slows down, and eventually seizes, if no germination follows (Briggs, et al., 1981). Water imbibition is a physical process. The extent to which imbibition occurs during steeping is determined by seed composition, seed coat permeability, availability of water (either in the liquid or gaseous form) in the seed envornment (Mayer and Poljakoff, 1963) the presence of additives in the steep liqour and the volume of water used. The steeping technique employed also affect the rate of water imbibition (Briggs, et al., 1981). The steep volume in industrial maltings is quite variable. Palmer and Pathgate (1976), reported a steep volume of 2.5 w/tonnes, while Briggs, et al., 1981 reported a volume of 27.90 W/tonnes for barley. According to Briggs et al., (1981), additives are

employed during steeping for various reasons. Formaldehyde (0.1%) is employed in steep liquors to control microbial growth, to reduce dormancy and to suppress germination, thereby reducing the malting loss. Formaldehyde steeps also kill Fusaria and other groups of fungi on grains. It also helps to reduce polyphenol content. Hydrogen peroxide (0.1%) steeps increase oxygen pressure, thereby helping to overcome dormancy. It also helps to promote a rapid and even grain germination. The same could be achieved by the use of ozone and giberellic acid. Giberellic acid also helps to break dormancy and to accelerate grain modification. Bromates and salts of octanoic acid are used to control the development of soluble nitrogen. Alkalis such as sodium hydroxide and calcium hydroxide are also employed in steep liquors to reduce microbial population on grain as well as the phenolic contents while acetate or chloroform are used to increase the rate of water imbibition.

Steeping systems vary in different maltings. These include rectangular cisterns or tanks or concrete steeps without aeration or emptying facilities, conical steeps with aeration and gravity discharge systems and those flat bottomed steeps with automatic aerating and discharge systems (Briggs, et al., 1981). In most modern maltings, steeping systems are not separated from germinating systems (Palmer and Pathage, 1976). In traditional techniques or immersion steeping, grains are soaked in water (usually fresh well water) for a specific duration as in U.K. with or without air rest, the steep liquor being changed at least once in twenty-four hours. However steeping techniques have developed, over the years from immersion to immersion/air rest steeping, to running water steep to spray steep to immersion/aeration steep. The immersion-air rest steep is often used for large scale malting as it helps to overcome the need for specialized equipment as in modern aerated steeps, whilst still ensuring a rapid and vigorous germination (Briggs, et al., 1981). It has been demonstrated that spraying steeps with nitrogen or carbondioxide improve malt yield by I and 2½%, respectively, and that oxygenated steeps result in uncontrolled germination and high malting loss (Briggs, et al., 1981). Generally grains loose 0.5 - 15% of its dry weight as dust and dissolved materials. Growth occurs during steeping with a resultant oxygen consumption by steeped grains. (ie, Biological oxygen demand, B. O. D). This results in pH increase, hence the need to change the steep liquor at least once during steeping (Briggs, et al., 1981).

The ultimate moisture content which a particular variety of malting grain must attain during steeping is of utmost importance in controlling its germination rate and modification intensity. For barley, this lies between 42-48% while for sorghum, it lies between 40-60% (Palmer and Pathgate 1976; Ikediobi, 1989). Thus a strict steeping schedule must be worked out for each grain variety. Little wonder then that many maltsters use secret steeping schedules which they have worked out to give the best germination for a particular seed (Ikediobi, 1989).

2.7.4 GERMINATION

Seed germination may be regarded as the consecutive number of steps which cause a quiescent seed with a low water content to show a rise in its general metabolic activity and to initiate the formation of a seedling from the embryo (Mayer and Poljakoff, 1963).

Before germination, grains from the steep are first drained of water for some time (6-10 hours for barley) and then made up into a heap or couch to build up heat (ie, casting) (Briggs, et al., 1981). The subsequent treatment to which grains are subjected during germination depends on the type of malting technique employed. In floor malting, the grain is spread out on a very clean floor at a predetermined thickness (6-9 inches for Sorghum and about 30 cm for Barley) and left to germinate or sprout. This process is carried out under optimum conditions of room temperature, relative humidity aeration and malting duration. (Ikediobi, 1989). The floor temperature varies from 13-16.5°C for barley and 25 - 30°C for sorghum malting (Briggs, et al., 1981; Ikediobi, 1989); while the relative humidity varies from 95 - 100% for barley and 85-95% for sorghum (Palmer and Pathgate 1976; Ikediobi, 1989). In traditional malting the grain is turned regularly with shovels. Turning of germinating grains also prevents root matting (Palmer and Pathgate, 1976; Briggs, et al., 1981).

In pneumatic malting system, the steeped grains are germinated in a bed one metre or more in depth and are attemperated with a forced flow of air and turned mechanically. The air is saturated with water vapour, hence, the relative humidity is rigidly controlled at 100% (Briges et al., 1981). In compartment malting, germination is carried out in open-topped containers (saladin boxes) of various structures. As in pneumatic malting germinating grains are turned mechanically and are attemperated by conditioned air. In drum malting steeped grains are germinated in drums through

which attemperating air could be forced. In both cases steeping and germination are sometimes carried out in the same compartment and some are designed for both steeping, germination and even killning (by replacing the attemperating air with a flow of hot dry air from the furnace). The use of a semi-continous and continous malting plant in which the germinating grains move down a tower with successive compartments (in which conditions can be adjusted to suit the germination stage) has been proposed (Briggs, et al., 1981). Though labour intensive the traditional flour malting is ideally suited for the developing countries beccause of its cheapness and simplicity (Ikediobi, 1989).

The main objective of germination is grain modificcations. Essentially, grains modification is characterized by a large increase in the quantities of some hydrolytic enzymes present in the grain, partial enzymatic degradation of reserved substances such as cell wall, gums proteins, starch and lipids. (Hulse 1981, Palmer and Pathgate, 1976). These biochemical changes occur simultaneously with many physical changes For example, the growth of the acrospire is commonly used as a rough guide to the progress of malting, particularly in barely malting (Briggs, et al., 1981).

Germination can be carried out with or without additives. For example, the use of bromates, giberellic acid or ozonized water spray had similar effects on grain as described under steeping.

The most important biochemical change during germination is enzyme development, since enzymes are essential for grain modification. The key enzymes in malting are alpha and beta-amylase, beta-glucanase, cellulase, neutral protease and amylogucosidase. The Alhapa-, and ß-amylase hydrolyses starch (amylose and amylopectin) to maltose and dextrin; Neutral protease hydrolyses protein materials to amino-acids and peptides, beta-guanase hydrolyses beta-glucans to glucose while amyloglucosidase hydrolyses alpha-linked dextrin to glucose. (Aisen, 1989; Mayer and Poljakoff, 1963).

The most important occurrence of alpha and betamylase in higher plants is to germinated cereals. Whereas beta-amylase is present in ungerminated cereals, alpha-amylase is practically absent at this stage and the low amount present is believed to be of microflora origin. Finger millet is a good source of alpha and beta amylase and the amylase content increases with the period of germination. Malt from R009 and H-22 millet cultivate were found to be richer in alpha-amylase than comparable wheat and barely malt and higher in beta-amylase than sorghum malt. (Halse, et al.,

1980). The barley grain has relatively higher levels of beta - 1,3,1,4 - glucanase and beta 1,3 glucanase than sorghum which, however, contains equally high levels of ß-amylase. The origin of these enzymes in sorghum is the embryo, while in barley they occur in both the embryo and the aleurone layer (Aisen, 1969; Novellie, 1960, 1966). The amylase activity in aqeous extract of barely increases two to three folds during germination due to, proteolytic liberation of enzymes from latent state (Briggs et al., 1981).

Similarly, amylase activity in Reeg and H-22 millet cultivars increases with germination, reaching a climax between 72-96 hours germination time (Hulse, et al., 1980). The possibility of intensifying the saccharifying amylase activities in millet (dextrinase, alpha - and beta-amylase and alpha-glucanase) can be achieved by the use of 0.25-0.5g heteroxime/tonne of malt. During germination alpha-amylase was found to be increasing at a higher rate than do beta-amylase activity (Rachie and Majmudaz, 1980).

The diastatic power of germinated sorghum cultivars varies widely. Under laboratory conditions, the activity increased up to 96 hours of germination and later decreased. This was accompanied by an increase in the abumin-protein content and a decrease in protein content in sorghum (Subramanian and Jambunathan 1989). Novellie, (1977) reported an increase in the diastatic power of pearl and finger millet (germinated at 30°C) from 22.0 and 7.2 respectively to 65.4 and 51.2 SDU/S respectively (ie, II and 3.1 to 32.7 and 25.6°L) respectively after the 4th day of germination. The optimum diastatic power attained during germination is influenced by several factors. Also sorghum germinates ideally between 25-30°C while barley germinates between 18-20°C (warm variety or 12-13°C (cold variety) (Holdt and Brand, 1960). Low moisture levels resulted in diastatic power reduction in sorghum malt. Novellie (1962) showed decrease in diastatic power of 12.5-10°L for lower moisture levels in malted sorghum. Cutting down on watering towards the end of malting resulted in a fall in moisture level by the fourth day of germination with a concomitant retardation in diastatic power development in sorghum malt.

Proteolytic activity in millet also increased with germination although the ungerminated grain showed no activity. It has been shown that proteolytic activity could be activated by hydrogen sulphide and cyanides; slightly inhibited by ascorbic acid and completely inhibited by copper

(Chandrasekhara and swaminathan, 1957). Although ungerminated finger millet contain low level of glycerol-phosphatase a marked increase in the activities of these enzymes was reported after germination (Hulse, et al., 1980).

The increase in enzyme activity in germinating grains result in the biodegradation of the cell well and food reserves. In a well-modified barley malt, about 75% of the ß-glacan material is broken down and about 40% of the protein is solubilized. In malted sorghum up to 43% of the starch may be broken down. The higher increase in starch degrading in sorghum has been attributed to higher temperature employed in sorghum malting and a more vigorous growth of grains. (Holdt and Brand, 1960). This is of great significance in brewing, since the bulk of brewers extract derives from starch degradation products (Briggs. et al., 1981).

There is little change in the total nitrogen content during seed germination. The fat, content decreases during the first few days but some re-synthesis occur later. The resynthesis of fat and the appearance of reducing sugars is presumably the result of the transformation of the disappearing starch (Mayer and Poljakoff, 1963).

2.7.5 KILNING

The main objective of kilning is to produce a friable readily milled stable product that my be stored for long period and from which roots may be removed quite easily (Ikediobi, 1989, Briggs, et al., 1981). Drying is effected by passing heated air of low relative humidity through the germinating grains at low temperature. Removal of moisture at low temperature allows for maximal survival of enzymes and the least development of aroma and colour. It has been reported that the use of a rapid air flow (at 40°C) to dry green malt ensures' the survival of the full compliment of diastatic enzymes (Briggs, et al., 1981).

The simplest type of kiln used in traditional malting consisted of a fire basket situated in a low room, the floor of the room above being perforated. These fire baskets were later replaced by furnaces which generate hot air that is distributed throughout the hot air chamber beneath a perforated kiln floor (Briggs, et al., 1981).

The choice of fuel used in furnace is very important. Originally hardwood logs or faggots were used and a few special malts are still being made using such fuels, since smoke from burning wood gives the product their particular flavours (eg, scotch includes low-arsenic coal, natural gas, low-sulphur oils etc. (Briggs, et al., 1981). Many modern kilns consist of multiple furnaces and this helps to reduce the drying time.

The most common driers used are the bed and rotary driers which are either diesel or electricity powered. The germinating grain is fed in through one end of the drier and are carried on conveyor belts through the drier compartment which is fed with hot air of low relative humidity at 50 - 70°C (Ikediobi, 1989). A slow rate of drying is necessary to ensure uniform drying. The drying rate is influenced by the volume of air passing through the kiln bed, the depth of the grain bed, the quantity of moisture to be removed, the temperature and moisture content of the air entering and leaving the grain, temperature within the grain, the degree of shrinkage and the quantity of rootlet present (Briggs, et al., 1981).

During kilning, the diastatic enzymes are degraded to variable extends.α-glucosidase and the debranching enzymes are apparently inactivated more readily than ß-amylase, while α-amylase is least readily degraded. Consequently malts intended to have maximal diastatic powers such as distillers malt are kilned at lowest possible temperature (Briggs et al., 1981). Swaminathan and Chandrasachra (1953) reported a marked loss of diastatic power for germinated sorghum dried at temperatures above 60°C for alpha and beta-amylase in millet and showed that the diastatic power increased when germinated millet was dried in a current of air at 45-50°C.

Barley malt is normally dried to a final moisture level of about 6%, while sorghum malt is dried to 8-10% (Ikediobi, 1989). The malt is passed through a revolving perforated drum that retains the corns whilst allowing the roots to pass through, thus ensuring separation of the culms. It is then cooled before storage to reduce the chance of insect infestation, to check the development of colour and flavour and to arrest enzyme decline. It is believed that malts stored for a minimum of 4-6 weeks before mashing gives a better brew (Briggs, et al., 1981).

2.8 ASSESSMENT OF MALT QUALITY

The parameters used for malt quality assessment includes moisture determination, malting loss, cold/Hot water extract diastatic power, filtration time and dextrinizing power determination etc. These parameters estimate the brewing value of malt in the sense of the nutrients yielded by the malt, provide basis for malt evaluation and therefore serve as guide for commercial transaction (Briggs, etal., 1981).

Malting loss represents the combined losses during steeping (leaching of preformed matters into the steep liquor etc), Respiratory loss (as cerbondioxide and water) and weight of shoots/roots. A high malting loss is therefore suggestive of excessive growth during germination. The relatively higher malting loss of sorghum (20-30%) compared to barley (9-12%) has been attributed to difference in the solubility patterns of their amylases (Malomo, 1989), and also to the higher temperature employed during sorghum malting. Of the 9% malting loss in barley, steeping loss contributes 1.5% respiratory loss 3.5% and root weight loss 4%. Malting loss is increased at higher moisture levels in germinating grain. Palmer and Pathgate (1976) showed that the increased level of hydrolytic enzyme during malting results in increased yield of hot water extract.

The cold water extract represents the preformed sugars or matters soluble which is leached out into solution from ground malt under conditions which excludes the intervention of enzymes (Palmer and Pathgate, 1976) while the hot water extract (H. W. E) is a measure of the quantity of dissolved materials in a wort prepared by infusion or decoction mashing on a laboratory scale. Extract yield remains a very important index of malt quality in India, since the extract yield is an important measure of brewerhouse performance of the malt. Factors which influence the extract yield includes the type of raw material, the milling operation mash turn behaviour and filtration (Chukwura, 1989). Also samples with high nitrogen or crude protein give a malt with a lower yields of extract than low nitrogen sample (Briggs, et a]., 1981). In barley, the predicted extract yield, E is related to the per-cent nitrogen and the one-thousand corn weight (G) as follows: E = (A x II.ON + 0.22G) x 2.96, where A is a constant related to the barley variety (Palmer and Pathgate, 1976). The germination potential also has a significant effect on the level of brewers extract obtained from the malted grain. Extract yield increases with fine grinding at the expense of wort filtration rate (Palmer

and Pathgate, 1976).

Diastatic power is the degree of amylolytic activity of the malt (Bishop. 1971). According to Novellie, 1962, the diastatic power is a better index of malt quality than extract yield. Also malt with high diastatic power is best adopted for malting. The diastatic power of malt is influenced by several factors. These include temperature, moisture level germination time, type of grain etc. Novellie 1962 malted sorghum in the temperature range 15-35°L and obtained the highest diastatic power (25°L) at 30°C. He also demonstrated that low moisture levels resulted in a reduction in diastatic power and an increase of 20 - 25°L for higher moisture levels. The effect of drying temperature on diastatic power was also studied. He showed that only a negligible reduction in diastatic power occurred when germinating sorghum was dried at 40-60°C but a marked loss resulted when the drying temperature was raised to 70°C.

Mash filtration rate decides the number of brews obtainable. Factors influencing the filtration rate includes mash viscosity, particle size distribution and type of raw material. The filtration pattern of barley malt mash depends to a greater extent on the barley grain variety, the glucan content and the husk, (Palmer and Pathgate, 1976). However, replacement of barley malt with up to seventy-five percent sorghum reduces the filtration rate especially when the lauter turn is used. Replacement of the lauter turn with a mash filter improves the brew house performance of sorghum malt (Haris, 1971.

2.9 SURVEY OF MALTING ACTIVITY IN NIGERIA

Industrial scale malting is a completely new practice in Nigeria and accounts for less than 0.1% of reports or experience in this field. The bulk of the reports emanate from laboratory and pilot scale malting from institution such as the Federal Institute of Industrial Research Oshodi (F.I.R.O), The North Breweries Kano, Universities and Polytechnic (Ikediobi, 1989). A visit to the International Beverage Industry (I.B.B.I) Kaduna also revealed that the company was in the process of changing from a pilot scale sorghum malting to a commercial scale, perhaps on a scale large enough to meet the company's requirement. However, only three companies, namely, Savannah grains processing, Zaria; Benman Development Company Nsuka and Food Specialities Nigeria Limited, Agbara Lagos are currently engaged in Industrial scale malting in Nigeria (Ikediobi, 1989). These companies can

only supply a fraction of total requirement of all the breweries in Nigeria. Brauhaase, a German brewery which has many technical partners in Nigeria, has proposed the setting up of five 40,000 TPA (tonnes per annum) malting industries at an estimated cost of seven hundred million naira to provide the malt requirements of the country. It is estimated that such a project could create up to 5,000 direct jobs and several thousands more in related industries beside generating a netsaving of ₦187,552,000 on barley importation and ₦494 million Naira on co-operation and excise taxes by the year 2004 (Srugis and Hope, 1987).

However, the Nigerian Government and most Nigerian entrepreneurs have not shown much interest in this proposal inspite of the attractive projected profits. According to Sugris and Hope, (1987) the reluctance of Nigerian entrepreneurs to invest in the malting industry might be attributed to the highly scientific and technically complex and capital intensive nature of the industry, the absolute requirement for highly skilled manpower, the continuing use of raw grain and the illegal importation of barley malt as well as the uncertainty about Governments future intention, the assurance that barley malt has been banned for good notwithstanding.

2.10 BREWHOUSE PROCESS

Beer is a fermented hopped malt beverage. The brewing process involves milling, mashing, wort treatment fermentation and lagering (Pomeranz, 1975.)

2.10.1 MILLING

This is a process of critical importance in brewing. It's main objective is to reduce the malt or unmalted grain to a particle size suitable for rapid enzymic digestion and hot water extract during mashing. Too small particle size results in wort run off impairment while too large particle size results in extract yield reduction (Palmer and Pathgate 1976; Briggs, et al., 1989). A particle size range of 300-1,000m is considered ideal for lager beer production (Chukwura, 1989). Therefore correct roller mill setting must be used to ensure that the particle size falls largely within this range during the milling operation. This can be achieved by estimating the extract yield of the grist at varying roller mill setting (ie, trial milling) and selecting the setting which gives grist of intermediate extract yield

(Chukwura, 1989). Sorghum grist from setting 2 of the muhler-miag roller mill has been shown to produce grist of excellent extract yield (Igyor, 1989). According to Briggs et al., (1981) brewers grist size in the range 3000-1,600 um is practicable with the use of only the roll stand coupled with appropriate adjustment of the roll gap. Alternatively, the malt may be milled coarsely or finely and two separate mashes carried out. The difference in the extract yield of these mashes is an indication of the degree of modification. Modification is considered satisfactory for values less than $5^{\circ}L/Kg$ for barley malt (Palmer and Pathgate, 1986). However, it is essential to keep the quantity of fines (flours) to a minimum to prevent the formation of substances which cause excessive dough in the mash (Joseph, 1977).

Unlike barley milling, millet and sorghum are precrushed by a hammer mill before feeding into a roller mill because of the relatively smaller sizes of their kernels. Failure to do so results in grist with poor extract yield since small kernels pass through the feed and crushing rollers of the mill without being crushed (Chukwura, 1989). The efficiency of the milling operation is greatly influenced by the uniformity of grain sizes. Since most breweries in Nigeria obtain their sorghum directly from the open market, mixed varieties are often obtained with a concomitant lack of uniformity of grain size. It is therefore mandatory to adjust the roller mill setting for different batches of sorghum (Chukwura, 1989).

Wet-milling is generally preferred for barley malt or grain meant for lager beer production. This helps to preserve the husk, which facilitates wort separation when the lauter turn is used for mash filtration (Arri, 1989). It also increases the extract potential of the malt and the possibility of duct explosion during milling (Palmer and Pathgate, 1976). However, sorghum and millet are huskless and cannot be successfully handled in a lauter turn. The mash filter has been shown to give a better filtration for sorghum mash (Arri, 1989). Besides, the product obtained by wet-milling of millet is less pure than those of other cereals due to the difficulty of degermination of the small kernel's (Freeman and Bocan, 1973). The efficiency of the milling operation can be ascertained by various quality control tests. These include fibre, Ash and fat determination (Briggs et al., 1981).

2.10.2 MASHING

The main objective of mashing is the enzymatic conversion of malt or solid adjunct to fermentable extract suitable for yeast growth and beer production (Palmer and Pathgate, 1976; Briggs et al., 1981). It involves the mixture of a definite proportion of malt, grains and adjunct (grists) in water followed by heating and cooling cycles. The ratio of grist to liquor is very important since mash concentration influences wort fermentability. This decrease's with increasing extract loss at extreme concentrations (above 60%). However a thick mash also results in greater enzyme protection leading to wort richer in total aminor-nitrogen (Kobalch et al., 1950 Omagbemi 1983). The grist liquor ratio varies with mashing technique employed. Aisen, (1989) mashed sorghum by infusion technique using a grist liquor ratio of 3:1.

The type of water used in mashing is very important. The brewing water should be clean, fresh, palatable, free from iron and preferable of a moderate degree of permanent or sulphate hardness. Generally acidic liquor is used, hence temporary hardness which gives undesirable alkaline pH must be removed from the mashing liquor (Palmer and pathgate, 1976). The mash pH also affects wort composition. Increased acidity leads to higher soluble nitrogen in the wort due to greater protein degradation by proteolytic enzymes, resulting in beer less prone to non-biological haze (Diagboya, et al, 1983).

The ionic composition of the brewing water is also very important. Ions such as calcium, bicarbonate, and carbonate have profound effect on the brewing process. These ions precipate the soluble phosphates thereby reducing wort pH from about 6.0 to about 5.4. The pH of water which contains excess calcium ions but no carbonate is closer to the pH optimum for ß-amylase and several proteolytic enzymes. This results in greater wort fermentability, total soluble nitrogen (TSN) and free amino-acid and less polyphenol and silica extraction during sparging. It reduces the mash pH from about 5.4 to 5.2, encourages protein precipitation (as hot breaks), reduces beer bitterness by reducing hop utilization and also precipitates oxalates thereby reducing its potential for haze formation and beer gushing problem. It also causes yeast flocculation and improves foaming of beer by calcium soap formation with lipids (Briggs, et al., 1981; Palmer and Pathgate, 1976).

The carbonate ions, on the other hand, raises wort pH and therefore deccreases wort fermentability and total soluble nitrogen. Thus it counteracts the action of calcium ions as described above. Magnesium ion imparts a marked sour to bitter flavour and astringency to beer at concentration above 5mg/litre and produces a laxative effect at high concentrations. It is also an essential co-factor for certain yeast, enzymes. The sodium ion (as sodium chloride) gives a desirable palate fulness to beer but sodium gives unpleasant taste. Potassium inhibits certain yeast enzymes and gives beer a laxative effect at concentrations above 10mg/L). Iron at concentration above 1 mg/L weakens yeast activity, results in slimes and rusty deposits and promotes protein tannin haze formation; it is however essential in trace amounts. Copper is mutagenic to yeast cells and also promotes tannin-haze formation while nitrate ion is reduced to toxic nitrite ion by wort bacteia (enterobacterial species). Though toxic at high concentration, zinc is stimulatory to yeast growth when added at 0.1-0.2mg/L (Palmer and Pathgate, 1976).

Therefore the brewing water is generally demineralized or treated carefully with sodium phosphates and with chelating agents such as ethylenediamine tetraacetic acid (EDTA) to ensure that the level of these ions fall within acceptable limits. Brewers generally ensure a supply of water (or liquor) of constant ionic composition by relying on their own bore-hole rather than obtaining water from local supplies (Palmer and Pathgate, 1976).

There are two basic types of mashing techniques, although various modifications exist. In infusion mashing a mixture of the grits (or grits) and liquor is heated gradually to temperatures just below boiling point in a mash tub and kept at this temperature for a definite duration. In the upward infusion process, the malt is doughed in with water at 38-50°C and kept at this temperature for one hour to permit sufficient proteolytic activity. Cooked adjunct at boiling temperature are then added to raise the temperature to 65 to 70°C and kept at this temperature for a few minutes to permit starch saccharification (Briggs et al., 1981). Jayatissa et al, (1980) desccribed a modified downward infusion technique for sorghum intended for ale beer production. They gelatinized a mixture of sorghum flour and distilled water (1:4) by boiling and then added diastase after cooling and maintained the temperature at 45°C for 30 minutes and at 70°C for 3 hours. Decoction mashing on the other hand is characterized by part-withdrawal of the mash which is boiled and returned to the main mash (ie,

Decantation mashing). In the two stage decoction mashing another one third of the mash is removed, boiled and returned to the main mash to raise the temperature to a higher value. Thus decoction mashing could be one or multiple-staged depending on how many times a portion of the mash is withdrawn, hoiled and returned (Briggs, et al., 1981).

One of the problems associated with sorghum mashing is poor saccharification due to the high gelatinization temperature of sorghum starch (65-75°C). This implies that even if there are sufficient enzymes in sorghum malt, they are inactivated in the process of gelatinization (Arri, 1989). The same is probably true for millet mashing given its high gelatinization temperature mentioned earlier. Thus sorghum beer is characterized by it's opaqueness due to the presence of starch and dextrin (Glenie et al., 1986). Although complete saccharification of sorghum mash can be achieved with the aid of exogenous enzymes, this is not economical, especially with the poor handling and *storage of enzymes under tropical conditions.*

The problem of poor protein solubilization during mashing has also been reported. This can be corrected by the use of exogenous protease and the inclusion of protein-rich materials in sorghum mash. The watery taste of sorghum beer or low body fulness has also been attributed to low protein, residual sugars and other materials after mashing (Arri, 1989).

Studies show that the type of mashing technique directly affects the mash temperature, texture, pH, viscosity, wort filterability, thickness and the general composition of the resulting wort. Thus infusion wort generally contain higher levels of limit dextrin, hemicellulose and glucans than decoction wort (Diagboya et al., 1983). They also showed that decoction wort is less succeptible to non-biological haze formation due to increased rate of tannin lipid and phytate degradation since these react with proteins thereby imparting colour to the wort (Narziss, 1981). The improved foam retention and decreased wort stability of infusion wort has also been attributed to increased rate of air absorbtion during infusion mashing. Also infusion mashing is more energy and labour intensive than decoction mashing resulting in higher production cost. (Hug, et al., 1979).

Mash pH affects wort composition significantly; at a pH of 5.8, 60-61% of the phytate and sugars present are hydrolysed while up to 90% are hydrolysed at a pH of 5.3 (Diagboya, et al., 1983). Also part of the insoluble protein is hydrolysed (solubilized) to simpler molecules such as

albuminoides, peptones, and polypeptides which account for the flavour, tactile impression (mouthfeel) and foam potential of the beer. The conversion of peptones and polypeptides to amino-acids by peptidases during mashing is also very important from the point of view of yeast nutrition (Briggs, et al., 1981).

Since the conversion enzymes have different pH and temperature maxima, the choice of operational pH during mashing possess a technical problem. ß-glucanase which degrades the ß-glucans of the cell wall has an optimum temperature of 40°C and is inactivated at 60°C. Inadequate conversion of ß-glucans often results in slow run off and causes beer filtration problem. The -and ß-amylase have pH and temperature optima of (5.6 - 5.8), (50 - 5.5) at (65-67)°C and (52-62)°C respectively. The limit dextrinase (debranching enzyme) has a temperature and pH of 40°C while the peptidases are most active at pH and temperature range of 45-50°C and 4.2-5.3. The hemicelluloses are most effective at temperature and pH range of 40-45°C and 4.2-5.3. Thus a compromise pH of 5.2 to 5.7 is generally adopted in most breweries (Joseph, 1977). Mashing temperature on the other hand is varied in stages (depending on the mashing technique) to allow the different enzymes to act at their temperature optima.

Various additives are employed during mashing. Calcium sulphate is used to precipitate the sulphate ions which are responsible for the bitter taste of certain beers. It also enhances the heat stability of -amylase. Sodium chloride and Hcl are also added to act as buffer and to give palate fulness. Formaldehyde is added to the mash water to prevent chillhaze formation in lager beer and sugar to increase alcohol content. Exogenous enzymes such as -, ß-amylase, ß-gluccanase and protease are added to improve saccharification, wort filtration and yeast activity and also to reduce trub formation (Briggs et al., 1981).

2.10.3 WORT PRODUCTION

This includes wort filtration, boiling and cooling. Following mashing, sweet wort is separated from solids of the mash, by straining or filtration. Traditionally the lauter-tun is used for wort filtration. The vessel is designed to separate the husk from the wort. However, for huskless grains such as sorghum and millet, the mash filter is more suitable (Chukwura, 1989).

The filtered wort is normally boiled for one to two hours to arrest enzyme activity, to ensure wort sterilization and to bring about complete ionic changes which lowers the pH from about 5.4 to 5.2; to bring about the coagulation of proteins and phenols, for colour development from sugar caramelization, melanodin formation and polyphenol oxidation. Other effects of wort boiling includes the loss of volatile materials, the isomerization of the bitter acids and volatile materials in hop and to ensure wort bittering (Palmer and Pathgate, 1976). The extent of protein coagulation during wort boiling is influenced by the combined effects of temperature, pH, multivalent ions and polyphenol levels. The wort colour also increase's with pH, temperature and boiling duration (Palmer and Pathgate, 1976).

A number of additives are usually employed during wort production. These include the use of potassium metabisuphite to ensure wort brilliance; Hydrochloric acid to lower wort pH, hop for wort bettering and flavour enhancement. The brewing value of hop is found in its resins (The alpha acids of humulones and the bitter acids of lupulones) and essential oils which flavours the beer. Hop also contains tannins, monosaccharides, pectins, amino-acids, proteins, lipids, wax, cellulose lignin etc (Palmer, 1989). Ascorbic acid is used to reduce unwanted reducing action of reductones which combine with the -SH groups of proteins and are responsible for beer discolouration, turbidity and bad taste (Knor, 1965).

Quality control tests normally carried out on wort includes the determination of free amino-nitrogen (FAN), total soluble and permanent soluble nitrogen (TSN) and (PSN), total reducing sugars, wort gravity (original and apparent gravity) real extract, colour, pH, titratable acidity, filtration rate and yeast fermentability) (briggs et al., 1981).

The total nitrogen content of an unboiled wort (from harley) varies from 60-100 mg/looml and the alpha-amino nitrogen content from 160-200 mg per litre. The TSN/TN ratio (ie, Kobalch index) gives an indication of malt and nitrogen modification. Barley malt which gives values below 35 are said to be poorly modified while values above 42 indicate over modification. Malts which give values between 38 and 42 are considered well modified. An amino-nitrogen level of 130-250 mg/ml is considered adequate for yeast nutrition and the wort pH is expected to lie between 5-6.0 before yeast pitching (Palmer and Pathgate, 1976). Hot and cold water extract of about 290^{0}L/Kg and 17%, a

wort colour of about 2.5°C, a total soluble nitrogen and yeast fermentability of 1.6% and 0.75% have been reported for a high quality barley wort (Joseph, 1977). The original and specific gravity, and bitterness of barley malt wort varies from (10.7-18°P), (I.040-L.080) and (20-40 B.U) respectively (Joseph, 1977).

2.10.4 WORT FERMENTATION

The fermentation process involve's yeast selection and pitching, wort aeration, primary and secondary fermentation of lagering. Most or the worlds beer is produced by bottom fermentation although top fermenting yeasts are commonly used in Britain and some parts of the United States (Kirk Othmer).

The yeast species S.Cerevisae or Saccharomyces Calbergensis are generally preferred. S. cerevisae are top fermenters and are most active at temperature range 60-70°F (15.6-21.1°C). S. Calbergansis are bottom fermenters and are most active around 90°F (32°C). Thus it has the advantage that most infecctous micro-organisms that contaminate the cooled wort have less chance of causing spoilage (Briggs et al., 1981). Most breweries usually employ yeast from middle skimming of the previous fermentation. This has the advantage that it is less contaminated and also flocculates easily. Since yeast viability and purity is of utmost importance in brewing, it is usually subjected to many quality assurance tests. These include yeast sampling, dead cell count, test for consistency of yeast pitching etc (Joseph, 1977).

The cooled wort (7-8°C) is fermented in specially designed, sterile tanks (fermenters). These vary in design, although they serve the primary function of providing a controlled environment for yeast growth. The main feature of a fermenter is that it can be sceptically operated for a number of days. It is constructed in such a way as to allow minimum evaporation and is provided with facilities for agitation, aeration, sampling and effective temperature control. It must also have a smooth surface and be constructed from a suitable material such as glass or stainless steel which can withstand repeated steam distillation (Knusden, 1977).

Yeast pitching is carried out in starting or settling tanks immediately after wort cooling to avoid microbial contamination. A pitching rate close to a million cells per ml. of wort suspension is normally employed at the start of fermentation depending on the wort specific gravity, temperature profile and the desired fermentation rate. Low pitching rate generally results in aromatic beer (Palmer and Pathgate, 1976).

Wort aeration is necessary for the synthesis of unsatured fatty acid and sterols by yeast cells for their internal membranes. Therefore, the addition of these substances reduce the need for dissolved oxygen since oxygen combines with polyphenols in wort to give a dark wort (Briggs et al., 1981). Temperature control during fermentation is also very critical due to the exothermic nature of the process (160K.Cal/Kg of extract). The temperature is generally kept high after the initial fermentation when most of the fermentable extracts have been converted to in order to reduce the vicinal diketone (VDK) flavour components particularly in larger beer which lacks the heavy flavour unpleasant that could mar the impleasant diacetyl flavour formed at the initial stage (Joseph, 1977).

The green beer obtained after the end of primary fermentation (7-14 days) is stored at low temperature (-1 to 4^{o}C) to permit secondary fermentation (lagering) to take place. A two yeast system comprising flocculent and non-flocculent yeast is generally employed for the secondary fermentation if the extract was not completely fermented during the primary fermentation. The course of fermentation is generally measured in terms of wort attenuation. This is the decline in wort specific gravity over the fermentation period. The degree of the fermentation (fermentability) is the difference between the extract in the original wort and the real extract in sample (obtained after dealcoholization of the fermented wrot) divided by the original extract multiplied by 100. However the maximum achievable attenuation for a given batch is expressed as final degree of attenuation (Briggs, et al., 1981; Palmer and Pathgate, 1976).

Factors which bring about abnormal fermentation includes abnormal wort composition, wort infection, abnormal wort aeration, poor temperature control, weak yeast, wrong pitching technique etc. Abnormal fermentation is characterized by late start of fermentation, development of strange odour, foaming and others (Palmer and Pathgate, 1976).

The finished beer is subjected to diverse quality assurance tests. These include the determination of foam collapse rate, colour, bitterness refractive index, pH, protein, reducing sugars, gravity and alcohol content. Most Nigerian beers have an alcohol content of between 3-8% and a pH between 3.9 - 4.4 (Joseph, 1977).

2.10.5 SENSORY EVALUATION

Finished beer is normally subjected to sensory evaluation based on the organoleptic assessment of attributes such foam stability, colour, bitterness, odour, sweetness, Brilliance, odour, sparkling and sourness. The quality of a given beer is considered as its relative excellence based on the sensory estimates of these attributes. Thus a good quality beer is expected to be slightly sour and bitter. Its foam stability should be good. It should be brilliant, sparkling. Must not be sweet and should possess a characteristic bodyfullness and flavour (Joseph, 1977).

Four basic types of organoleptic assessment techniques used for food items are the preference, the acceptance, the discriminatory and the descriptive test tests (Ihekoronye and Ngoddy, 1985). These include the single stimulus method, the paired comparison, the triangle, multiple comparison and the ranking tests. Because organoleptic tests are subjective, techniques which minimize errors are usually built into each test. For example, the use of two controls, one of which is revealed to the panellist, helps to identify experienced testers since these are able to match the unknown sample among the various samples tested (Elizabeth, 1977).

Four basic scoring systems are used in the sensory evaluation of foods, viz, the nominal, ordinal, interval and ratio scale (Amerine, et al., 1965). The nominal technique is generally used for attributes such as colour which cannot be described precisely. For example, the colour of cider has been scored on the basis of the following descriptive terms: Green, Straw, Pink, Yellow, Gold, Amber, Brown. The panellist is merely requested to tick the box adjacent to the appropriate colour for the sample provided (William, 1978). 1979). The ranking technique uses numbers to indicate magnitude or a line scale to measure the magnitude of a given attribute. The ends of the scale are fixed using appropriate descriptions which denote the complete absence or presence of a given attribute. Thus the odour intensity of cider has been scored on a number scale ranging from zero

(no odour) to 5 (very strong odour); Also the bitterness of a cider has been scored on a scale based on descriptive terms such as much less bitter, slightly less, same slightly or none bitter. This involves at least two samples one of which is a known control. The panellist merely ticks the relevant descriptive term (William, et al., 1979). The conventional hedonic scale usually contain between 9 to 10 points. For example, (l-very bad or, not acceptable). 2(bad), 4(indeferent), 5(fair), 6(pleasant), 7(Good), 8(very good), 9(Excellent or most acceptable). It has been suggested that the hedonic information is much less amendable to interval scoring than objective data and that fewer sub-divisions should be used for negative attributes since people are much less critical when judging unacceptable products (William, et al; 1979). Also a statistically valid sample size is necessary to obtain reliable hedonic information since people differ considerably with respect to the organoleptic acceptability of a given product. This variation arises partly due to the difficulty in obtaining a uniform descriptive terms for sensory perceptions and also because people are apt to compare sample to an internal standard based on what they like.

The type of scale used has a considerable influence on the result obtained because the state of mind of the panellist influences his/her judgement and this is in turn influenced by the adjectives and phrases used to determine the scale. Thus the line scales 'No roughness to extremely rough', 'no smoothness to extremely smooth', 'extremely smooth to extremely rough' produce information capable of diverse interpretation when applied to the same attribute.

The interpretation of data from any scoring technique is based on statistical analysis. Thus the significance of rank data may be tested by comparing the rank sum for each sample with rank total tables or by comparing the distribution with Fisher Z distribution table (Wright, 1976); Data obtained from scales involving underlying continum are usually subjected to analysis of variance couple with F-tables to determine the significance of variance. Analytical techniques such as the canonical variate analysis are considered most appropriate for multivariate systems which involve many attributes. (Kendal, 1962). According to Wright (1976), tables of t and f theoretically apply only when the raw scores, in an experiment are randomly and independently sampled from population distributions that are Gaussian in shape. When these assumptions are not valid, Non-parametric statistics such as the Mann whitney-U-test (Rank sum test) or the Chi-square (X^2) technique may be

used. Although the Mann whitney "U" test is about 95% power efficient, its use is limited by frequent recurrence of ties in the raw scores. Fortunately, this limitation does not apply to the Chi-square techniques. It can be used to test whether any difference between two or more populations arose by chance within a specified confidence limits and degree of freedom (Wright, 1976).

Chapter 3

3. MATERIALS/REAGENTS

3.1 MATERIALS/REAGENTS

A local variety of pearl millet (LVPM) was purchased from a farm at Ungwar Kanawar Kaduna. The chemicals and equipment used were obtained from the Department of Biochemistry Ahmadu Bello University, Zaria and the Food and Drug Administration, Kaduna.

3.2 SAMPLE PRETREATMENT

3.2.1 CLEANING:

The bulk of millet grains (as purchased from the farm) were washed serveral times with tap water to separate the stalks, sand and other foreign particles. It was then sundried for 3 days. The untreated portion was reserved for the determination of foreign kernels and percentage floats.

3.2.2 MILLING:

An aliquot of the sample was taken randomly and milled using a hammer mill. Various sieves were tried and the grain was finally milled to a suitable particle size (355-1000 Um) as determined by screen analysis described below. The sieve which gave this particle size range was employed for all the subsequent milling operations in this project.

3.2.3 PARTICLE SIZE ANALYSIS:

The empty sieves of the screen analyzer were separately weighed. 100g of flour was mixed thoroughly and placed on the top most sieve and covered. All the sieves were fastened together, and the shaker was switched on for 15 minutes.

At the end of this time, the sieves and the retained flour were weighed and the weight of flour retained in each sieve was calculated by difference and expressed as a fraction of the total weight. Calculation showed that 93.21% of the flour has particle sizes in the range 355-1000 Um.

3.3 MALTING PROFILES OF MILLET GRAIN

3.3.1 DETERMINATION OF PERCENTAGE FLOAT

The method of the American Society of Brewing Chemists (ASBC) 1958 was used (Anon, 1958).

One hundred millet kernels were randomly selected and mixed with water in a test-tube. The floating kernels were scooped off, counted and expressed as percentage floats. The procedure was carried out in triplicates.

3.3.2 DETERMINATION OF PERCENTAGE OF FOREIGN MATTER AND BROKEN KERNELS

The A.S.B.C method (Anon, 1958) was used. Triplicate samples of the millet grains (30g each) were spread on a wide piece of white cardboard paper on a laboratory bench. All the non-millet grains and broken kernels were carefully picked out, weighed and expressed as a percentage of the original weight.

3.3.3 DETERMINATION OF GERMINATION ENERGY

The A.S.B.C method (Anon, 1958) was used for this determination. Triplicate samples of millet grains (100 kernels each) from which foreign matters and broken kernels have been removed, were placed in test-tubes and covered with distilled water at 27°C (room temperature). filling the tube nearly to the lip. The floating kernels were forced down the water surface from time to time and the water was replaced with fresh distilled water after 24 hours. The whole steeping lasted 48 hours.

The steeped grains were washed with distilled water and spread in a single layer on a moistened filter paper which was folded such that one half of it was pressed down firmly on the layer of grain.

The moistened filter papers were left in a desiccator (containing some water to maintain the relative humidity near saturation) to germinate for 72 hours. The grains with roots or sprouts were counted and the number expressed as a percentage of the total grains evaluated.

3.3.4 DETERMINATION OF GERMINATION CAPACITY

This is a continuation of the procedure described above (3.3.3). Kernels that showed no evidence of growth after the 72 hours period of the germination energy test were replaced between the damp filter paper and returned to the desiccator for an additional 48 hours. The grains with roots and sprouts were counted and this was added to the number obtained for the germination energy. The resultant grains was expressed as a percentage of total grains evaluated.

3.3.5 DETERMINATION OF ONE THOUSAND KERNEL WEIGHT

The method of the association of official analytical chemists (A.O.A.C) 1980, was used. An aliquot of 500g of millet grains (from which broken kernels and foreign matter have been separated), was taken. This was quartered into smaller portions until not more than 20g remained in opposite quarters. The number of millet grains in each quarter was counted. The one thousand kernel weight (in grams) was subsequently calculated using the formular $G = \frac{W \times 1000 \times DM}{N \times 100}$

Where, W = Total weight of millet taken

N = Total number of grains counted

DM = Per cent dry weight of the sample.

3.3.6 MOULD DETECTION

The A.O.A.C. method (1980) was used. An aliquot of millet grains were spread out on a white cardboard sheet on a laboratory bench and each grain was visually examined for the presence or absence of mould. The result was reported as, "none," "traces" etc.

3.4 PHYSICAL AND BIOCHEMICAL ANALYSIS OF MILLET GRAIN

3.4.1 MOISTURE DETERMINATION

The method of the American Society of Brewing Chemists (A.S.B.C) 1958, was used. An aliquot of milled grain (3.00g) was placed in a weighed moisture dish. The dish and its content was then heated in a preheated oven for 3 hours at 105°C. The dish was removed from the oven, cooled down in a desiccator, and finally weighed to the nearest 0.001g. The percentage of moisture in the millet sample was calculated from the formula: $M = \frac{W_1 n - W_2}{W_1} \times \frac{100}{1}$

Where, M = Moisture content (%)

W_1 = Weight of sample before drying

W_2 = Weight of sample after drying.

3.4.2 PROTEIN DETERMINATION

The micro-Kjedahl method of analysis (as described by Pearson 1976) was used. An aliquot of 0.15g of the millet flour, 0.9g of Kjedahl digestion tablet and 2ml of mitogen free concentrated sulphuric acid were dispensed into a micro-Kjedahl digestion flask. The contents were digested for 2 hours on the electric heater in a fume cupboard until a clear liquid was obtained. Subsequently 2ml of distilled water and 15ml. of 10N NaOH were added to the clear digest and transferred into a Markham's distillation apparatus.

10ml of 2% boric acid solution was pipetted into a conical flask and 2 drops of mixed indicator (Bremocresol green and methyl red) were added. The digest was then steam-distilled for 10 minutes into the boric acid solution.

The light - green solution obtained after steam distillation was titrated against 0.02NHC1 to a light-red end-point.

The percentage nitrogen $(\%N)_2$ was calculated thus:

$$\%N_2 = \frac{\text{Volume of Hcl x Normality (0.02) x0.14007 x 100}}{\text{Weight of sample}}$$

3.4.3 FAT DETERMINATION

The A.O.A.C method (1980) was used for this determination. Ten grams of millet flour was accurately weithed out into an extraction thimble and covered with cotton wool. Petroleum ether was poured into a weighed round-bottomed flask up to two-third of its volume, and the soxlet extraction apparatus set up.

The flask was heated for 2½ hours on an electric heater, after which the petroleum ether was distilled and the flask (plus fat residue) was dried to constant.
Weight in an oven at 105°C.

$$\%Fat = \frac{W_2 - W_1}{1} \times \frac{100}{1}$$

Where, W_2 = Weight of flask + fat residue

W_1 = Weight of empty flask

$$\%Fat\ (Dry\ weight) = \frac{\%Fat\ (as\ is) \times 100}{100 - M}$$

3.4.4 STARCH DETERMINATION

(a) ACID HYDROLYSIS:

The A.O.A.C method (1980) was used for this determination. Ten grams of defattened millet flour (from fat determination already described) was mixed to a fine consistency with 25ml of ether in a mortar. The coarse materials were allowed to settle while the supernatant was decanted into an llcm filter paper (of sufficiently fine texture to retain the crude starch). The process was repeated until no more coarse particle remained. The fat-free residue on the filter paper was washed twice with a jet of cold water to remove any trace of reducing sugar.
The resultant insoluble residue together with 200ml distilled water and 20ml Hcl were transferred into a 250ml flask and heated under reflux for 2½ hours. The cooled mixture was neutralized with a few drops of NaoH, made up to the mark with distilled water, filtered, and stored for dextrose determination. The weight of starch is given by the weight of dextrose multiplied by 0.925.

(b) **DEXTROSE DETERMINATION:**

This was determined by the Lane and Eynons method as described by Pearson, 1976.
A 2% glucose solution was titrated against 20ml mixed Fehling's solutions under a constant state of ebullition in a narrow-necked flask until the blue colour was nearly discharged. 3 drops of methylene-blue indicator were added to the mixture and the titration was continued until the indicator was completely decolorized.
A second set of titration was carried out using the same procedure in which the 2% glucose solution was replaced with the hydrolysate (from millet flour). The weight of dextrose was calculated from the formula:

$$\text{Dextrose (mg/100ml)} = \frac{F \times 100}{C}$$

Where, F = Lane and Eynons factor as obtained from the table (Appendix 4)

C = Titre (for hydrolysate).

3.4.5 TOTAL FREE AMINO ACID:

This was determine by the A.O.A.C (1980) method. 20g aliquot of millet flour was mixed with some distilled water in beakers and the resultant mashes were sequenced through a piece of cloth into centrifuge tubes and centrifuged at 5000 r.pm for 10 minutes.

The supernatant obtained (170 ml) was deproteinized with trichloroacetic acid (1.0g/20 ml). The precipitated proteins were removed by centrifugation at 5000 rpm for 15 minutes and the supernatant obtained was assayed for total free amino acid as follows: Iml; samples were prepared in a series of test-tubes containing 0.00 to 4.00 mg/ml. D.L Glutamine. Triplicate samples containing Iml of the deproteinized millet extracts were also prepared in three other test-tubes. 4ml ninhydrin was added to each test-tube and the solution was left for 10 minutes in a water bath at 100°C. The absorbance of the blue colour developed was read at 420nm on an sp6 spectrometer. The total free-amino-acid concentration was obtained from the standard curve of D.L glutamine absorbance (Appendix 1).

3.4.6 DETERMINATION OF POLYPHENOL

(A) EXTRACTION:

The method of Martins, 1973 was used for the extraction. Aliquots of 5.00g of the millet flour were blended with 37.5ml of chilled diethyl ether. The resultant mash was filtered by a vacuum filter and the filtrate was reserved. The residue was further extracted with 50 ml of ethanol and filtered as above.

The combined filtrates were extracted with an equal volume of diethylether in a separatory funnel. Fresh distilled water was gradually added from a flush bottle until phase separation occurred. The aqueous layer was run off into a beaker and re-extracted with half its volume of diethyl either in a separatory funnel, and then run off into a beaker for polyphenol determination.

(b) DETERMINATION OF POLYPHENOL:

This was determined by the method of Singleton, 1973. Iml samples were prepared in series of test-tubes containing 0-100 Ug catechd per ml. Triplicate samples containing Iml (each) of millet extract were also prepared in three other test-tubes. 5ml Ciocalteaus reagent (50%) and Iml sodium carbonate were added to each test-

tube and the solutions were heated for 5 minutes in a water bath at 50°C. The absorbance of the colour developed was read off at 600 nm on an Sp-6 spectrometer using a 1cm glass cuvette.

A calibration curve of absorbance versus catechol (phenol) concentration was then plotted (Appendix 2) and the concentration of phenol in the test solution was read off from the curve.

3.4.7 ESTIMATION OF TANNIN CONTENT

(a) PREPARATION OF VANILLIN - Hcl REAGENT

One gramme of vanillin was dissolved in 100ml of methanol to give an effective concentration of 1% vanillin in methanol. 22.2ml of conc. hydrochloric acid was added to 77.8ml of methanol to give 100 ml of the solution. Equal volumes of both solutions were mixed to give a freshly prepared vanillin-Hcl reagent.

(b) DETERMINATION

To 2.00g of millet flour was added to 10 ml of methanol in a conical flask and kept on the shaker for 1 hour at room temperature. The mixture was centrifuged and the clear supernatant obtained transferred into two test-tubes. 5ml of vanillin-Hcl reagent was added to each test-tube at 1 minute interval and then incubated at 30°C for 20 minutes. The absorbance of the colour developed was measured on an SP-6 spectrophotometer at 500 nm and the tannin concentration in the sample was read from the standard curve.

CONSTRUCTION OF STANDARD CURVE

From a stock solution (containing 100 mg of catechin dissolved in 50mls of methanol) 1ml samples were prepared in a series of test tubes containing 1-10 mg per ml. 5ml vanillin - Hcl reagent was added to each test tube and the solution was left for 20 minutes. The absorbance was then measured on an SP-6 Spectrophotometer at 500 nm and a plot of catechin concentration (mg/ml) versus absorbance (A 500) was made (Appendix 3).

3.4.8 PROTEOLYTIC ACTIVITY:

The method of the American Association of Cereal Chemists (Anon, 1962) was used. Duplicate portions (10g each) of millet flour and 50 ml, 0.1m acetate buffer (pH 7.4) were mixed together in 100 ml centrifuge tubes. The tubes were placed in a water bath at 45°C and stirred at 5 minutes intervals.

After one hour, 10 ml trichloroacetic acid (TCA) solution (30%) was added to one mash and three hours later, another 10 ml TCA solution was added to the second mash. Each mash was centrifuged and filtered. 3 ml of concentrated sulphuric acid was added to 20 ml of each filtrate in a digestion flask and the mixture was everporated to dryness. Subsequently the non-protein nitrogen in each flask was determined by micro-Kjedahl analysis as described in 3.4.2.

The proteolytic activity was calculated from the increase in non-protein nitrogen from one to three hours of digestion and reported as mg.nitrogen per 100g millet (or millet malt).

3.5 EFFECT OF DURATION OF STEEPING ON GERMINATION ENERGY

The germination energy was determined at varying steeping duration (3,6,9,12,15,18,21,24,47,30,33,36,39,42,45 and 48 hours respectively) following the A.S.B.C. method, 1958, as described in 3.3.3.

The steeping water was changed at 24 hours for steeping durations exceeding 24 hours.

3.6 PRODUCTION OF MILLET MALT AT VARYING DURATION OF STEEPING

Millet malt was produced at varying duration of steeping according to the modified procedure of Aisen and Muts, 1987. Allquots of 200g millet (from which foreign matters and broken kernels have been separated) were steeped in distilled water (1 part of grain to 2 parts of water) for various durations, 6,12,24,27,33,39 and 45 hours respectively. After steeping the grains were drained of water and heaped up in a wooden box for 24 hours to build up enough heat (ie, casting). Germination was achieved by spreading out the grains in a wooden box at a constant grain bed thickness of 2 cm. The germinating grains were sprinkled with water at six-hourly intervals and turned at the same time to ensure even distribution of moisture and to avoid matting of the roots. The malting was carried out at room temperature (28°C) and germination was terminated after 5 days (the 1 day casting period inclusive).

The germinated grains were kilned in an oven at 50°C for 24 hours after which the temperature was raised to 55°C for another 25 hours.

The roots were finally removed from the kilned grains by friction (abrasion).

3.7 ANALYSIS OF MILLET MALT

3.7.1 MOISTURE:

The moisture content of the various millet malts corresponding to 6,12,18,24,27,33, and 39 hours respectively) were determined as previously described (3.4).

3.7.2 DETERMINATION OF MALTING LOSS

This was determined according to the standard procedure of the Institute of Breweries (I.O.B) Anon, 1977. At the end of malting, the whole of the kilned grains were weighed after the removal of rootlets. The malting loss was estimated as the difference between the weight of the malt and that of the original grains (ie, dry weight), expressed as a percentage of the weight of grains malted.

3.7.3 DETERMINATION OF HOT WATER EXTRACT

The method of the Institute of Breweries (Anon, 1977) was used.

50.2g (+0.5) an aliquots of millet malt flour were weighed into tared stainless steel barkers, which had been preheated in the water bath. 350 ml of water ($68^{O}C$)was added to the malt flour (in each beaker) with stirring, and the water bath temperature was maintained at $65^{O}C$.

The mash was stirred after 30 minutes and was removed from the water bath after 1 hour, and quickly cooled down to $20^{O}C$. The cooled mash was made up to 515 ml with distilled water, and the temperature was adjusted to $20^{O}C$. The mash was then filtered into a beaker using a wide stem funnel and whatman No.1 filter paper. The first 50 ml. filtrate was returned to the funnel and the volume of the clear liquid (ie, the hot water extract) collected in the first 30 minutes was noted for the calculation of wort - filtration time. The hot water extract was estimated from its specific gravity.

SPECIFIC GRAVITY DETERMINATION

The method of the Institute of Breweries (Anon, 1977) was used for this determination. An oven-dried $25cm^3$ specific gravity bottle (with lid) was weighed empty, filled to the brim with distilled water (at $20^{O}C$), covered with lid, mopped with a filter paper and weighed again.

The bottle was emptied, rinsed with some of the hot-water extract, filled to the brim with same (at $20^{O}C$) covered with lid, mopped with a filter paper and weighed again.

The specific gravity (S.G) of the hot-water extrracct was calculated thus:

$$S.G = \frac{\text{Weight of S.G bottle filled with hot-water extract minus weight of empty S.G. bottle}}{\text{Weight of S.G bottle filled with distilled water minus weight of empty S.G. bottle (ie., the water equivalent).}}$$

The extract 'as is' (E) is expressed in litre degree per kilogram (Lit. deg. Kg^{-1}), is calculated thus:

$$E = G \times 10,13$$

Where, G is the excess degree of gravity of the filtrate, taking water at 20°C as 1000 is given by the formular:

$$G = 1000 (S.G.^{-1}) + C.F.$$

CF = Correction factor

= 0.236 x Every 1°C above 15.5°C.

$$\text{Extract (dry basis)} = \frac{\text{Extract 'as is'} \times 100}{100 - m}$$

Where, M = moisture content of the malt.

3.7.4 DETERMINATION OF COLD WATER EXTRACT (CWE)

Ten grams of millet malt flour was digested with 200ml of distilled water (containing 12 ml of 0.1N NH_3) for 3 hours at room temperature. The mixture was stirred at 30 minutes interval. It was then filtered through whatman No.1 filter paper and the specific gravity of the cold water extract was determined at a temperature of 15.15°C following the procedure described in 3.7.3

The cold water extract (CWE) was calculated thus:

Thus:

$$CWE = \frac{(SG - 1)\ 1000 \times 20}{3.86}$$

Where, G = The excess degree of gravity taking water at 15°C as 1000

G = 1000 (SG - 1)

SG = Specific gravity of extract.

N.B:

All the weighings in SG determination were carried out to 4 decimal places.

3.7.5 DETERMINATION OF DIASTATIC POWER

This was determined by the A.S.B.C. method (1958) as modified by Novellie, 1959. 25g of millet malt flour was mixed with 500 ml of distilled water at 30°C for 2½ hours, with constant stirring at 30 minutes intervals.

The mixture was then filtered through whatman No.1 filter paper in a glass funnel. 10 ml of the filtrate was pipetted into 200 ml, 2% soluble starch. 20 ml of 0.5N NaoH was added to the mixture 30 minutes later to stop the reaction and the volume was made up to 250 ml with distilled water.

The digested starch was then titrated (from the burette) against 5 ml of mixed Fehlings solution (in a narrow-necked flask, to avoid air ingress) under a constant state of ebullition until the blue colour of the mixed fehlings solution had disappeared. Three drops of 1% methylene blue solution (indicator) was added, and the titration was continued until the blue colour had been decolorized and a brick-red colour obtained.

3.8 DETERMINATION OF ALPHA AND BETA AMYLASE ACTIVITIES AT VARYING DAYS OF GERMINATION

(a) PREPARATION OF CRUDE EXTRACTS OF ALPHA AND BETA AMYLASES

The method of Botes, et al., 1967 was used. 10g of the milled millet grain and malts (produced by steeping the grain in water for 24 hours and germinating the grain for 1,2,3,4, and 5 days respectively), was extracted overnight at 2°C with 25 ml of distilled water. The suspension obtained was centrifuged to remove the coarse particles and then filtered using a filter aid (celite analytical filter aid). The clear extract was 40% saturated with ammonium sulphate (by adding appropriate amount of $(NH_4)_2SO_4$ to each extract based on the volume). The alpha-amylase precipitated was allowed to settle overnight and collected by centrifugation, dialysed against 0.05 acetate buffer (pH 4.6) and freeze-dried. The supernatant was reserved for beta-amylase extraction.

ESTIMATION OF ALPHA-AMYLASE ACTIVITY (NOVELLIE 1967)

Each precipitate was re-dissolved in an appropriate volume of buffer solution required to bring the concentration down to 25% saturation. 0.1ml each of the dilute alpha-amylase extract was then mixed (in triplicates) with 2ml of 2% gelatinized starch in acetate buffer (pH 4.6) and allowed to stand for ten minutes after which the reaction was stopped by the addition of 1ml of 1% dinitrosalycyclic acid. The absorbance of the red solution developed was read off at 540 nm on SP-6 spectrometer. The α-amylase activity is expressed in saccharifying units, defined as the amount of enzyme which would librate 1 μ-mole of reducing sugar (calculated as maltose) per minute.

ESTIMATION OF BETA-AMYLASE ACTIVITY

The supernatant obtained after precipitation of Alpha-amylase was saturated to 70% with ammonium sulphate and the precipitate obtained was allowed to settle overnight and collected by centrifugation. Each precipitate was then re-dissolved in an appropriate volume of buffer solution required to bring the solution down to 25% saturation. 0.1ml each of the solution was then used to hydrolyse the gelatinized starch and the colour developed with dinitrosalicyclic acid was measured as described in the last paragraph for Alpha-amylase.

(b) CONSTRUCTION OF STANDARD CURVE FOR AND β-AMYLASE CALIBRATION CURVE

1ml each of 1% dinitrosalycylic acid was mixed in different test tubes (in triplicates) with 1ml $CaCl_2$ and 1ml each of varying concentrations of standard maltose solutions (0.02, 0.04, 0.06, 0.07 and 0.10 1mg/ml). Each test tube was then made up to 4ml with an appropriate volume of 0.5m Acetate buffer solution (pH 4.6). A blank solution was prepared containing a mixture of 1ml. 1% dinitrosalyclic acid (0.5M), 1ml $CaCl_2$, 1ml distilled water and 1ml. 0.5m acetate buffer solution. The spectrometer was set to zero at 540 nm with the blank solution and the absorbance reading of the standard solutions were read off and recorded. A plot of absorbance versus maltose concentration was then made Appendix 4.

3.9 EFFECT OF GERMINATION DURATION (DAYS) ON MALT QUALITY

Four malt samples were produced by steeping 200g of millet grain in 0.1% aqueous solution of calcium hydroxide $Ca(OH)_2$ for 24 hours at varying germination duration (1, 32, 5 and 7 days respectively). Each sample was kilned at 50°C for 24 hours and at 55°C for another 24 hours. The same set of analysis as in 4.4 above were carried out on the malt samples using raw grain (zero germination) as control.

3.10 EFFECT OF ADDITIVES ON MALT QUALITY

Five malt samples were produced following the procedure described in 3.3. However, 24 hours steeping duration was used and the steeping media consisted of 0.1% aqueous solution each of ammonia, calcium hydroxide, formaldehyde, hydrogen peroxide and distilled water (control). The procedure was carried out in triplicates.

The following analysis were then carried out on each malt sample following the procedures described in the preceeding pages:

(i)	Moisture per cent	(vi)	Malting loss
(ii)	Hot water extract	(vii)	Crude protein
(iii)	Cold water extract	(viii)	Fat %
(iv)	Filtration time	(ix)	Starch %
(v)	Diastatic power	(x)	Ash %

The results obtained were statistically analysed using the analysis of variance (Ihekoronye and Ngoddy, 1985) technique.

3.11 PRODUCTION OF LARGER BEER FROM MILLET MALT (MODIFIED) METHODS OF AISEN AND MUTS 1987 MICHALE, 1989 AND AISEN, 1989

(i) MALTING:

Millet malt was produced following the procedure earlier described. (The steeping duration used was 24 hours and the steeping medium was 0.1% $Ca(COH)_2$.

1.25Kg millet malt grist was weighed out into a 5 litres beaker and kept in a water bath at 50°C to a temperate; The beaker was made up to the mark with hot distilled water to bring the initial temperature of the mash (ie, mash-in temperature) to 50°C. Sufficient NaCl/Hcl buffer solution was added to adjust the mash pH to 5.4 and both protease (130 PU/mg malt grist) and ß-glucanase (0.25g/500g malt grist) were added. The mash was maintained at 50°C for 30 minutes.

The supernatant (1.5L) was decanted and the main mash was brought to boil for 10 minutes on a bunsen flame and then cooled down to 50°C in a bowl of cold water. The mash pH was re-adjusted to 5.4 by addition of a few drops of NaCl/Hcl buffer solution and exogenous -amylase (0.3 Kg^2/ton of malt grist) was added together with the supernatant. Calcium sulphate (2.04g), Formaldehyde (a few drops) sulphate (0.62g) and sodium coloride (7.5g/5%) were added and the temperature was gradually raised to 65°C and maintained at this temperature for 30 minutes. The mash was cooled down to 50°C and the pH adjusted to 5.7 with Nacl/Hcl buffer. ß-Amylase (0.2g/500g malt grist) was added and the temperature was gradually raised to 75°C (Mash off).

The cooled mash was first strained in a clean jute bag, sparged with distilled water and then filtered through a graded analytical filter paper (Whatman No.1) in the presence of celite filter aid.

The clear wort obtained was boiled in an aluminum pot for 90 minutes. Hop (2.5g/litre) and sugar (22g/5 litres) were added 30 minutes to the end of boiling. Ascorbic acid (0.003g/L) was finally added to the cooled wort.

Brewers yeast Saccharamyces Calbergensis Uvarum (25g litre) was mixed vigorously with some of the cooled wort for 3 hours in a sterile petridish and transferred immediately into a sterile 10 litre glass jar (fermentation tank). The tank was thoroughly agitated to allow aeration, covered and transferred into a refrigerator at between 8 and 10°C and fermented for one week. The green beer so produced was filtered through a whatman N0.1 filter paper to remove the bottom flocculated yeast and cold trub, and then stored at 0°C for two weeks (ie, lagering at 0-4°C.).

The mature beer was finally filtered through Kieselghur (diatomaceous earth) at 6°C and manually carbonated and bottled.

3.12 SENSORY EVALUATION OF MILLET LAGER BEER

(i) SELECTION OF TEST PANEL:

Three coded samples were presented to each of the twenty test panellists. These are sample 412 (known control), sample 246 (millet lager beer) and 252 (unknown control). Each panellist was requested to score the beer attributes, foam, colour, odour, bitterness, sourness, bodyfulness sparkling and brilliance following the instructions in appendix 5.

The test panel was carried out in a well air-conditioned room at 10.00 am in the morning. At the end of the first test, 10 people who were able to identify the two control (star lager beer) were selected for the test panel.

(ii) ACTUAL TEST:

The selected panellists (10) were presented with two samples coded 328 (control) and 419 (millet lager beer) under the same condition (as described above) and requested to score the same beer attributes as in appendix 5.

The results obtained were statistically analysed using the chi-square technique. (Appendix 8).

Chapter 4

RESULTS AND DISCUSSION

4.1 *QUALITY ASSESSMENT OF UNGERMINATED MILLET*

The biochemical and brewing profiles of pearl millet are presented in Table 1. The one thousand kernel weight of the millet sample ranged from 3.4 to 10.10 with a mean value of 7.83. This is consistent with the findings of several workers. (Adrian, et al., 1957, Jambunathan as reported by Hulse, 1980).

The low germination energy and capacity of the millet sample (66.7 and 69.3) relative to barley (95 and 96)% observed in this study suggests a poor readiness of the grain for malting. Available evidence (Briggs, et al., 1981) however suggests that grains with low germination energy and capacity may malt readily on a large scale, indicating that these tests provide a rather pessimistic estimate of a grains readiness for germination. The low percentage float (3.7%) and percentage broken kernel (0.37%) all tend to suggest that the millet sample contains a high proportion of mature grains, and its utilization in brewing may be more economical, especially since broken kernel is much lower compared to the U.S. standard of 4-15% broken kernel required for grade 1-5 sorghum.

The low cold water extract of the sample (2.32%) suggests low enzyme activity in the ungerminated grain since the cold water extract measures the level of preformed soluble matter in the grain. However, the hot-water extract was relatively high (60.36 Lit. Deg. Kg^{-1}) indicating higher enzymic activity under the test condition.

The diastatic power of ungerminated millet was found to be 4.52°Lintner. This is much lower than that of barley. However, Schwartz, (1956) reported diastatic power between 0-7.8°Lintner for sorghum malt suggesting that the levels of diastatic power in such sorghum variety is comparable to this variety of millet. Other workers (Novellie 1962; 1977; Aisen, 1989 have since reported higher diastatic power for sorghum. This might be attributed to an improvement in sorghum breed as well as malting technology. It is noteworthy, however, that many parameters influence the diastatic power of a grain. For example, the diastatic power of sorghum increases with storage duration (Novellie, 1967).

The level of ß-amylase in the millet grain (55Ug maltose/Ml/Min) was higher than that of -amylase (45 Ug/ml/min). This is consistent with the findings of previous workers (Majmudar et al., 1980; Novellie et al., 1967). The low levels of tannin (2.48%) and polyphenol (1.07 Ug/100g) evident in present study is consistent with previous reports (Hulse, 1981; Tripton, 1974). These low levels are positive malting qualities since beer brewed from such malt is less disposed to chill haze formation from protein-polyphenol interaction.

Generally, the proximate analysis of pearl millet gave comparative nutrient levels with that of sorghum and barley (Table 1). The starch level in millet (66.5%) was slightly higher than barley (58-60) but lower than sorghum (70-& 2%). However, the malting significance of starch lies not only in the absolute level, but also in the starch gelatinization temperature the amylopectin/amylose ratio and starch solubility (Novellie 1960). Besides the carbohydrate profile of starch hydrolysis varies with different starch sources and this is more indicative of the level of fermentable extract that could be generated from a given starch. Since starch is bound to protein bodies the efficacy of starch degradation also depends on the proteolytic activity of the grain and malt. Analysis showed a level of proteolytic activity in the dormant grain (184.5mgN/100g.)

This is higher than the level reported for sorghum and barley (U.S.D.A., 1968). Although slightly lower than the level reported by several workers, proximate analysis gave a fat level of 4.5%. This is still significantly higher than the level reported for sorghum (2.8 - 3.5%) and barley (2-3%). Thus the fat level is in excess of 3.5% required for a good malting grain (Malomo, 1989). Although the calcium soap of fatty acid, the hydrolysis product of fat, is important for the foam stability of beer it has been demonstrated that very high level of fatty acid in the wort could dispose the beer to haze formation due to the formation of polyphenol-fattyacid complex (Briggs et al, 1981). This probably explains why a very high fat level may be regarded as a negative quality index.

The protein level in the pearl millet (11.06%) compares favourable with that of barley (8-13%) and sorghum (9.5-12.5%) (Nwasike et al, 1982; Aisen, 1989; Pomeranz, 1977). Thus the suitability of wort from pearl millet malt for yeast nutrition is assured, since this depends on the total soluble nitrogen and free amino nitrogen level.

The low tannin (2.48%) and polyphenol level (1.07%) is consistent with the findings of many workers (Chandra et al, 1977). However, higher levels have been reported for certain millet varieties. Thus the problem of beer haze associated with protein-polyphenol complex might be expected to be at its minimum in larger beer brewed from this millet malt. This was confirmed by the favourable organoleptic score on brillance and colour of the lager beer produced from this millet variety. Furthermore, the low polyphenol level suggests that the α-and ß-amylase of this millet variety would be relatively water-soluble, and this implies an enhanced malting potential.

TABLE 1: **DATA ON PEARL MILLET GRAINS (PROXIMATE, PHYSICAL AND BIOCHEMICAL PROPERTIES)**

	MILLET
Moisture (%)	10.03
Ash (%)	1.87
Fat (%)	4.25
Starch (%)	66.27
Crude Protein (%)	11.06
Reducing Sugars (%)	0.00
One thousand kernel weight (g)	7.83
Germination Energy (%)	66.7
Germination Capacity (%)	69.3
Percentage foreign matter (%)	0.67
Percentage float (%)	3.67
Cold water Extract (%)	2.32
Hot water Extract (Lit. Deg. Kg^{-1})	60.36
Alpha Amylase Activity (Ug Maltose/ml/Min)	45.00
Beta-Amylase Activity (Ug Maltose/ml/Min)	55.00
Diastatic Power (^{o}L)	4.52
Proteolytic Activity (Mgn/100g)	184.55
Phenol (Ug/100g	1.07
Tannin (%)	2.48

4.2 GERMINATION STUDY/MALT QUALITY (MILLET AS AN ALTERNATIVE FOR BREWING)

Figure 1. shows the plot of enzyme activity as a function of germination duration (days). ß-amylase activity rose from 55 µg maltose/ml/min in dormant grain, reaching a peak value of 248 µg/ml/min on the third of germination and subsequently fell gradually. ∝-amylase on the other hand rose from 45 µg maltose/ml/min throughout the seven days of study, giving a maximum level of 252 µg/ml/min on the seventh day. The proteolytic activity (Fig 2) on the other hand rose from 184.5 mgN/100g in the dormant grain, and reached a peak activity of 572.7 mgN/100g on 4th day. There after, the level gradually declined. The change in enzyme activity during millet germination is consistent with the findings of several workers.

Novellie, 1959, carried out a similar study and demonstrated a progressive rise in -amylase activity through a ten-days germination period. Swaminathan and Chandrasechara, 1953, reported a similar rise in proteolytic activity of H-22 millet variety. Adams, 1976. (As cited by Hulse, et al., 1980) also reported a similar change in the proteolytic activity of millet during germination.

It is not know whether these changes in enzyme activity is directly connected with the germination process, since germination has been shown to preceed rise in enzyme activity (Mayer and Poljakoff, 1963). However, the levels of the nutrient fell during germination (Figure 3). The starch level fell from 66.23 to 58.10; the protein from 11.06-10.08% and this probably resulted from the increased enzyme activity during germination. The brewing significance of these enzyme profiles is that they serve as a basis for establishing the appropriate time to stop the germination process during malting, since the primary aim of malting is to develop the maximum levels of modification enzymes whilst at the same time holding the nutrient loses (malting losses) to a minimum. Therefore it makes practical malting sense to stop the germination at the peak of enzyme activity which lies between the third and fifth day of germination for both the proteolytic enzymes and ß-amylase.

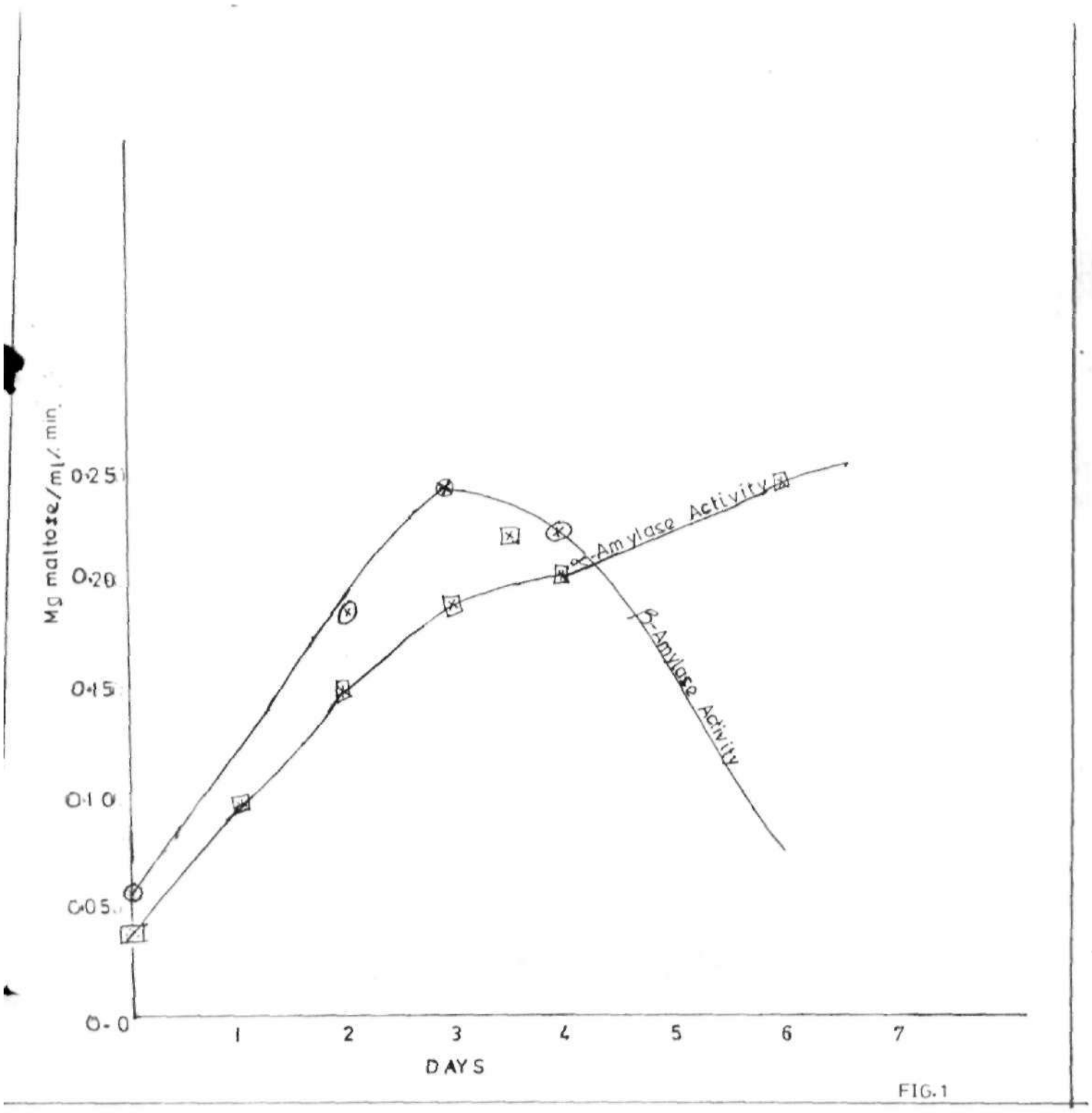

Mg maltose/ml/ min.
0·25
0·20
0·15
0·10
0·05
0·0
1
2
3
4
5
6
7
DAYS
α-Amylase Activity
β-Amylase Activity

FIG. 1

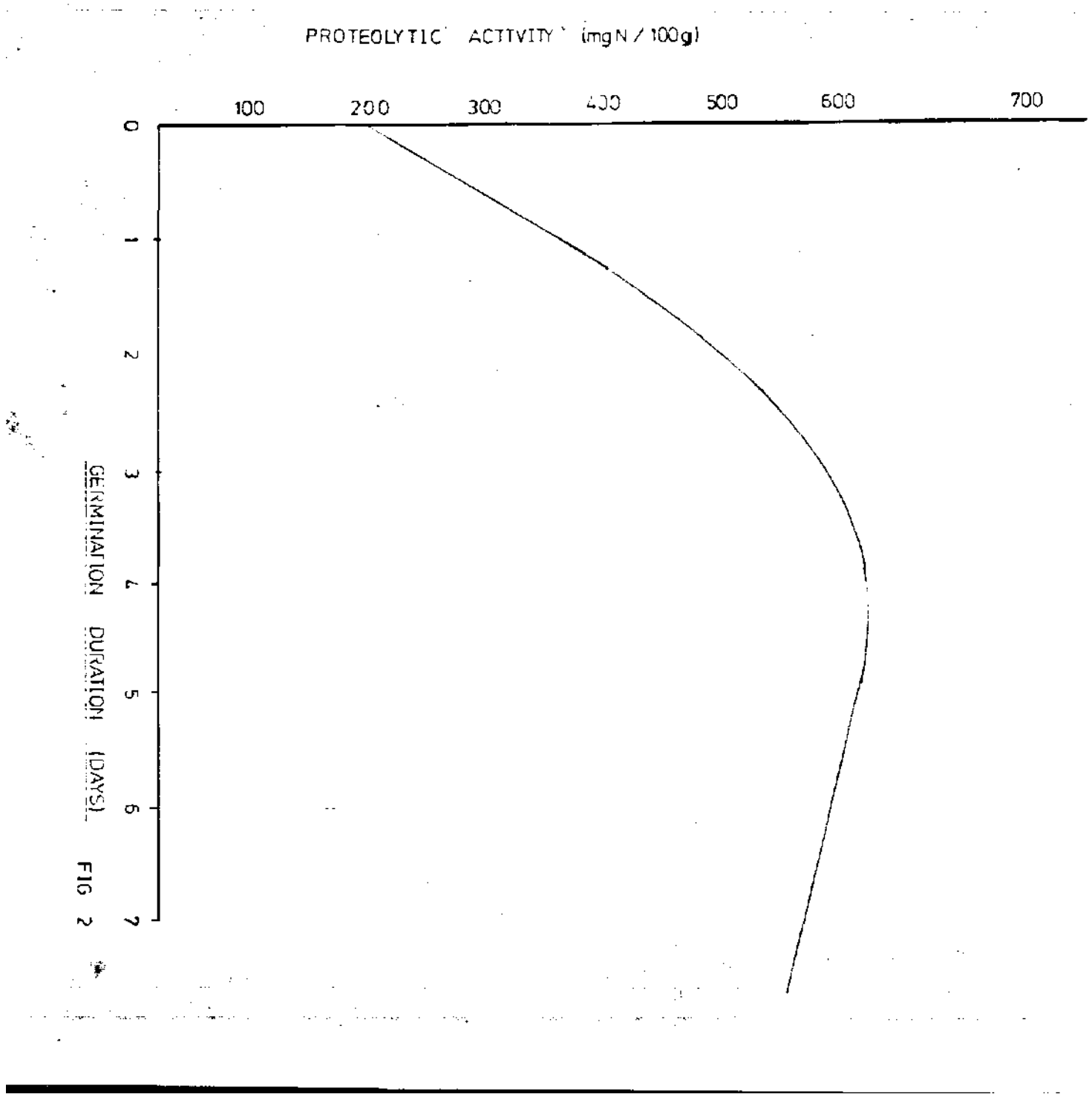

PROTEOLYTIC ACTIVITY (mgN / 100g)
100
200
300
400
500
600
700
0
1
2
3
4
5
6
7
GERMINATION DURATION (DAYS)

FIG 2

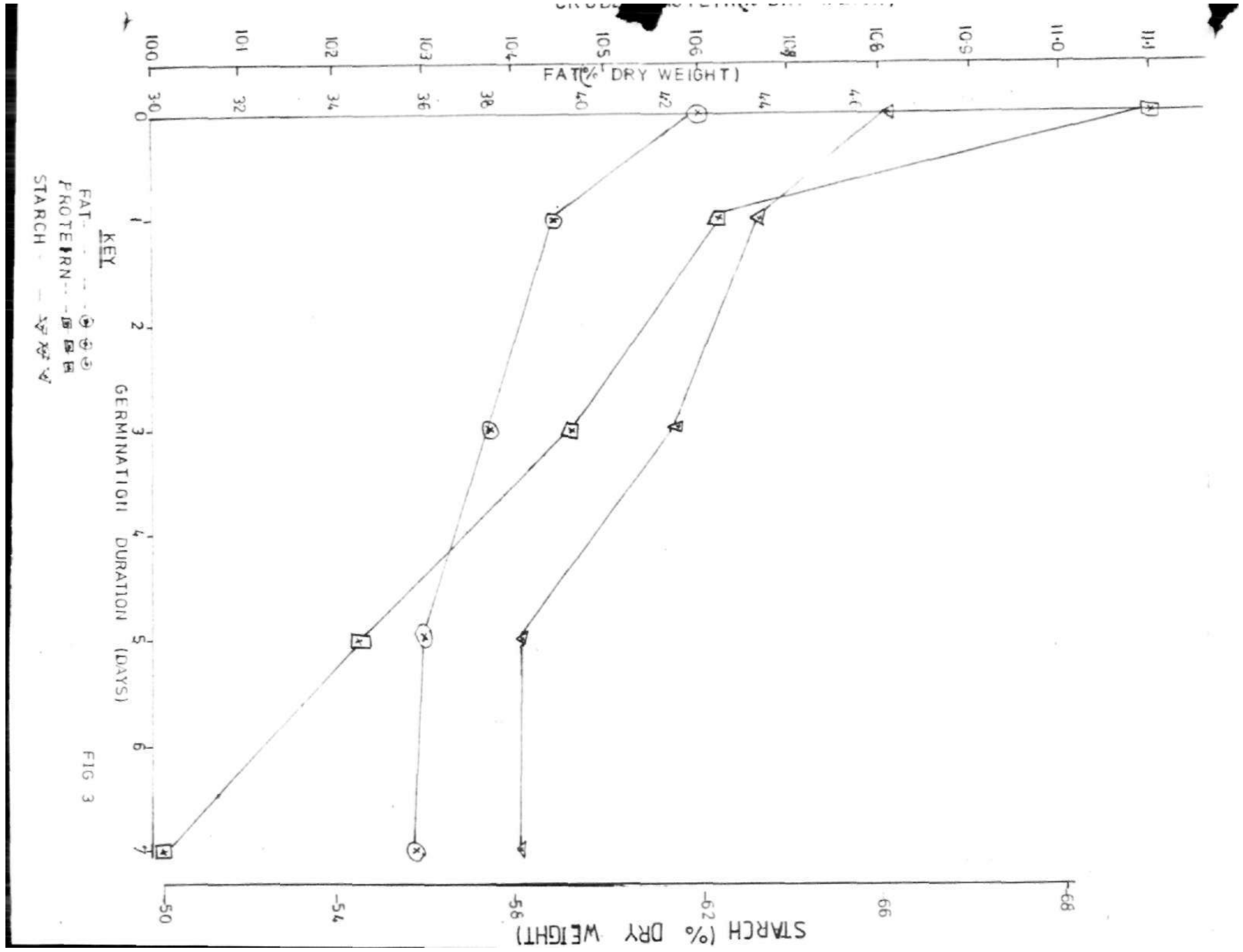

STARCH (% DRY WEIGHT)
99
99
-62
-56
-54
-50
FAT (% DRY WEIGHT)
11·1
11·0
10·9
10·8
10·6
100
101
102
103
104
105
106
30
32
34
36
38
40
42
44
46
0
1
2
3
4
5
6
7
GERMINATION DURATION (DAYS)
KEY
FAT
PROTEIN
STARCH

FIG 3

Figure 4 shows clearly the effect of steeping duration on the moisture level in the grain. The moisture level rose sharply during the first six hours of steeping. Thereafter the rise was gradual, reaching a peak of between 35-45% between the 18th and 30th hour; since a low moisture in grain leads to a reduction in the diastatic power (Novellie, 1962) the choice of an appropriate steeping duration is critical during malting. The moisture profile (ie, plot of moisture level versus duration of steeping) is characteristic for a given grain variety and therefore serves as useful guide in determining the appropriate steeping schedule to be adopted for each batch of grain during malting on an industrial scale (Briggs, et al., 1981). This is very important given the Nigerian malting experience where the supply of raw grains for most of the industries is directly from the open-market (Chukwura, 1989).

The germination energy of the millet increases with steeping (figure 5) and fell thereafter. This suggests that maximum water uptake is not the same as ultimum water requirement for grain germination. Figure 6 shows that malting loss is highest around the 15th hour suggesting that high germination energy is associated with a high malting loss. Thus a steeping duration of between 24 to 27 hours appears more suitable for this variety since it produces malt with higher diastatic power and moderate malting loss. Besides the highest cold water extract was obtained for steeping duration between 24 and 27 hours (Figure 7). Although a peak hot water extract was obtained after 6 hours of steeping, this is also associated with a high malting loss and low diastatic power. Besides a moderately high hot water extract was obtained between the 21st and 24th hour of steeping (figure 8).

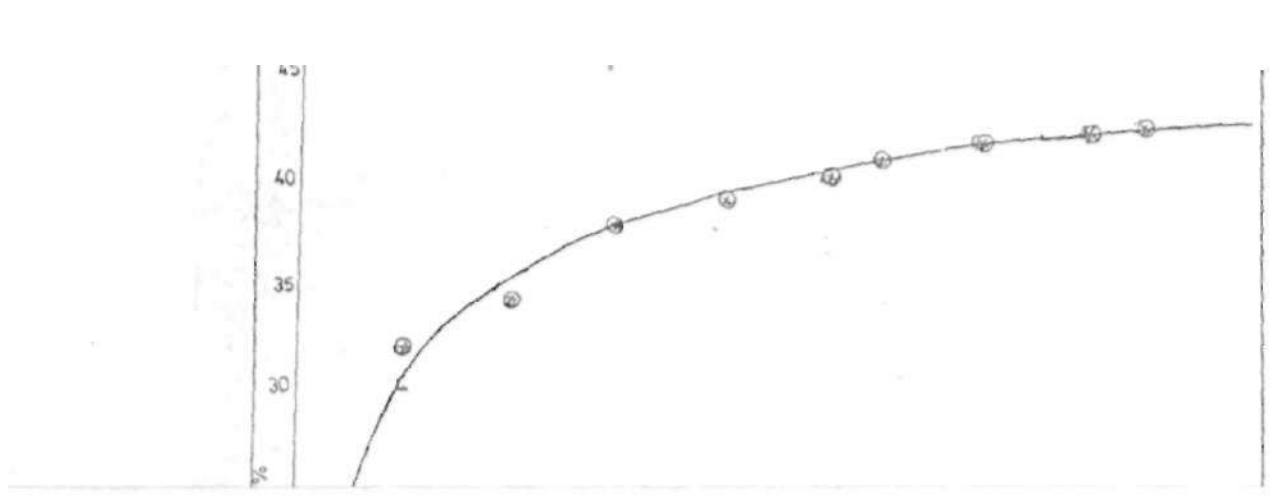


FIGURE IV: MOISTURE PERCENT VERSUS STEEPING TIME (HRS)

FIGURE V: STEEPING DURATION VERSUS GERMINATION ENERGY (%)

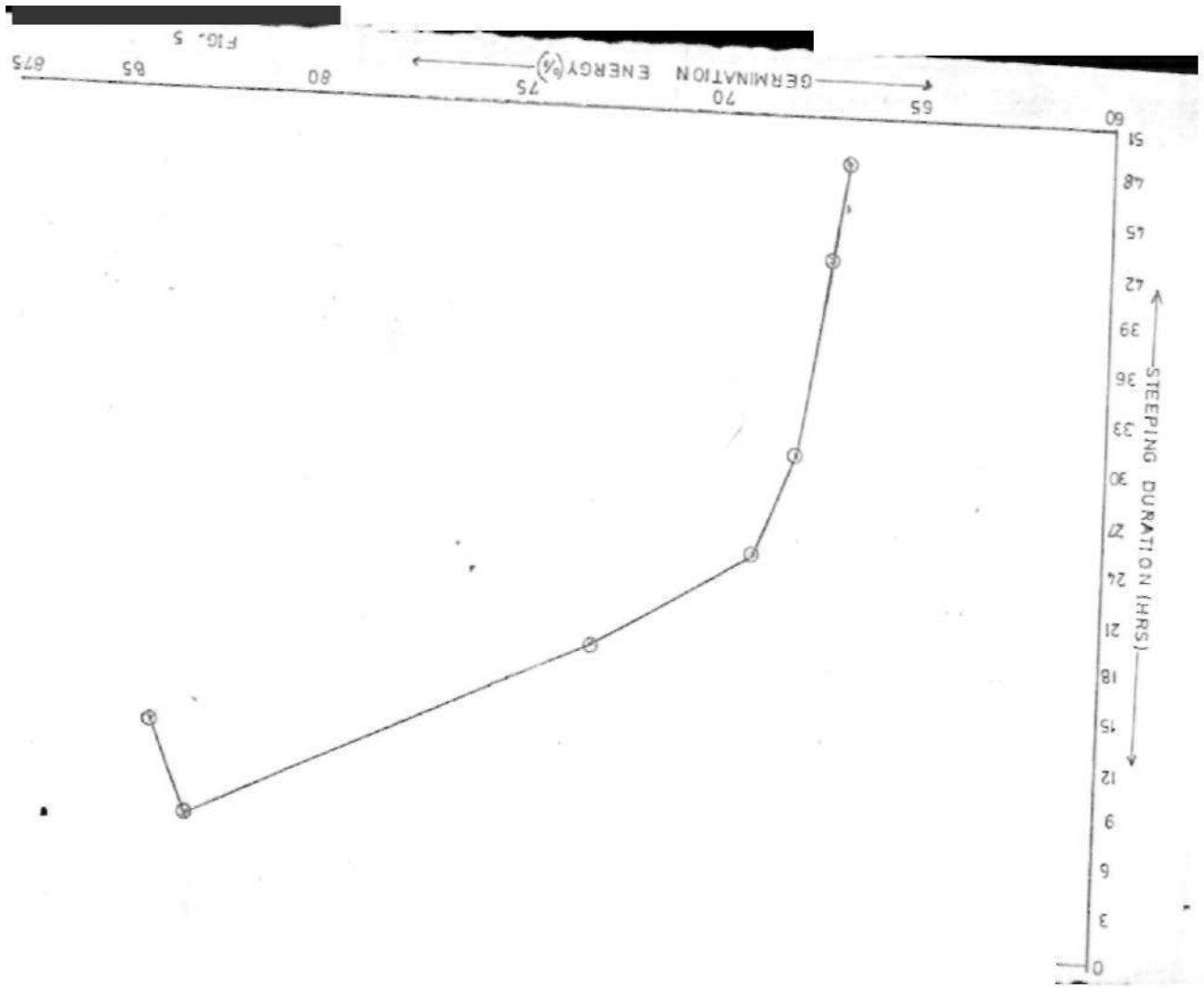
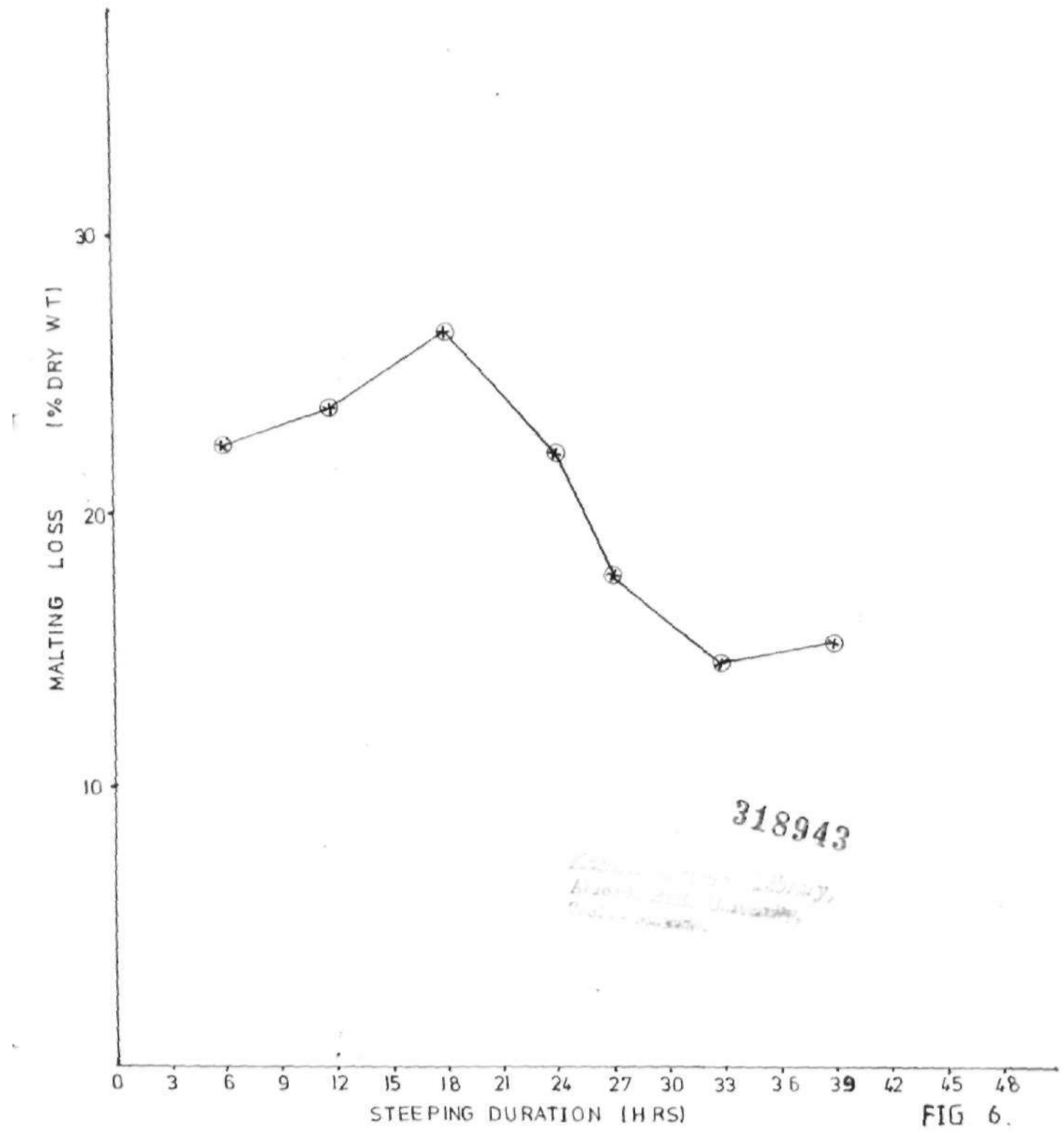

GERMINATION ENERGY (%)
60
65
70
75
80
85
90
STEEPING DURATION (HRS)
0
3
6
9
12
15
18
21
24
27
30
33
36
39
42
45
48
51

FIG. 5


MALTING LOSS (% DRY WT)
30
20
10
0
3
6
9
12
15
18
21
24
27
30
33
36
39
42
45
48
STEEPING DURATION (HRS)

318943

FIG 6.

FIGURE VI: MALTING LOSS VERSUS STEEPING DURATION

FIGURE VII: STEEPING DURATION VS COLD WATER EXTRACT

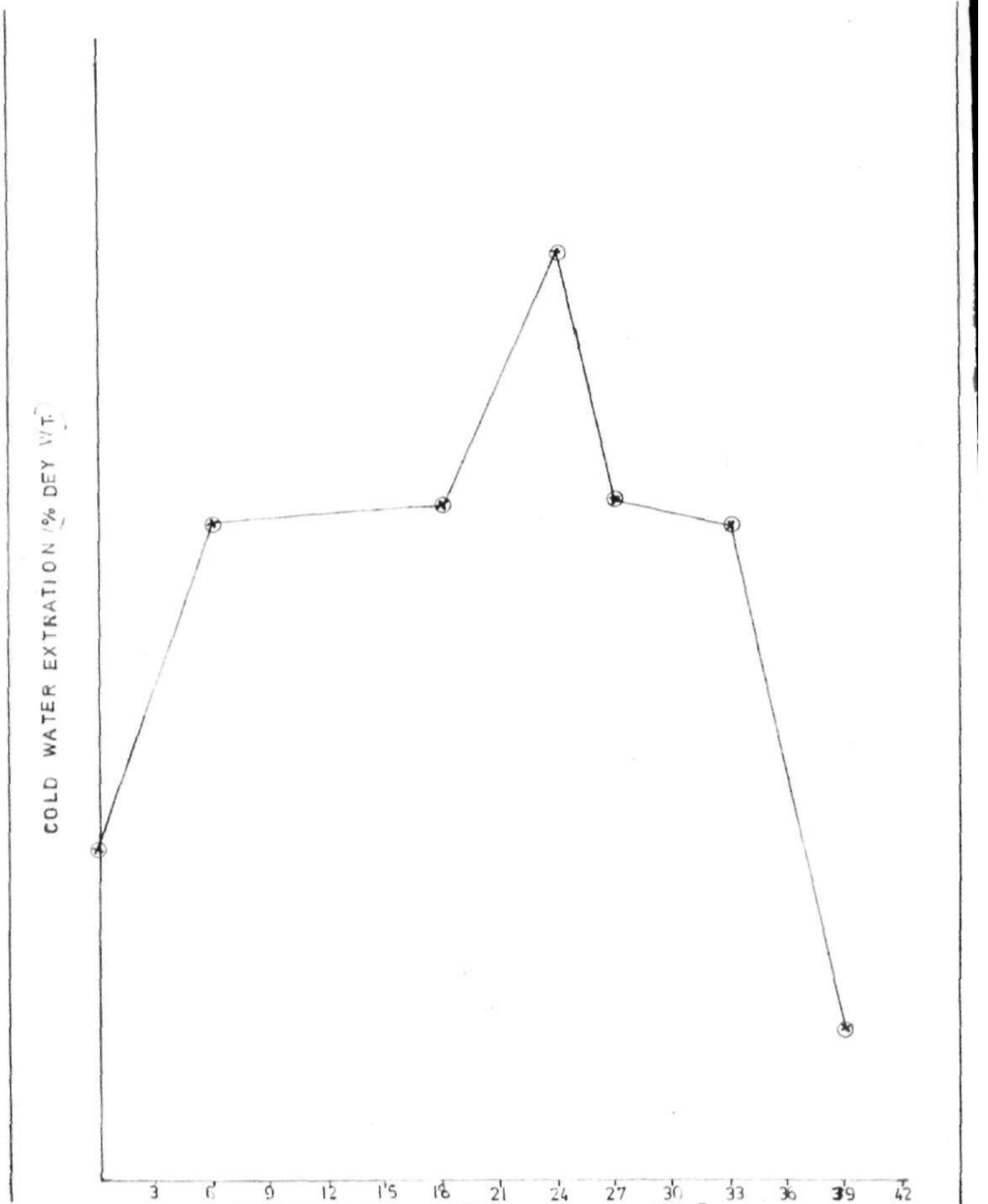
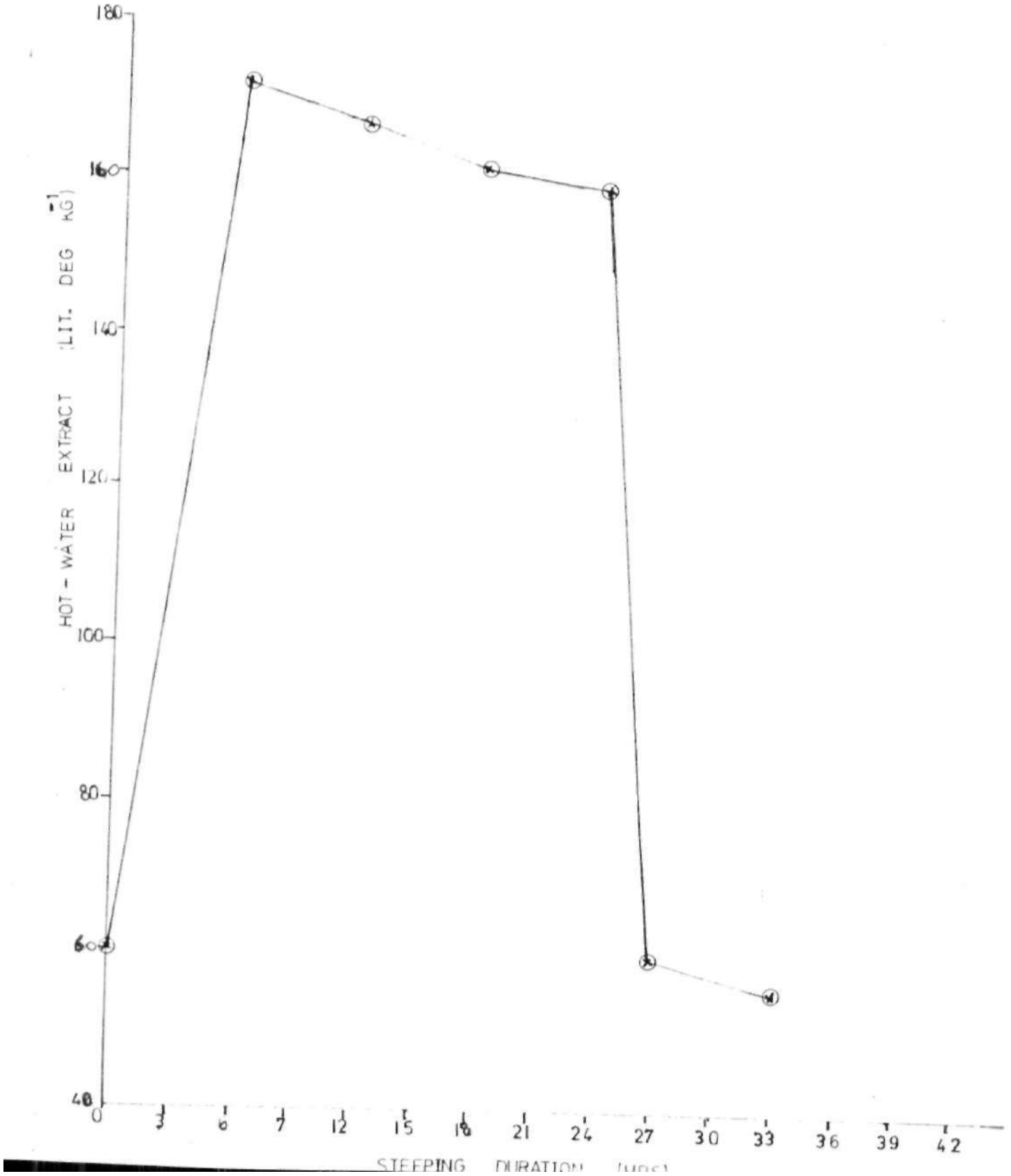

COLD WATER EXTRATION (% DEY WT.)
3
6
9
12
15
18
21
24
27
30
33
36
39
42
STEEPING DURATION (HRS)

FIG. 7

FIGURE VIII: HOT WATER EXTRACT VERSUS STEEPING DURATION


HOT - WATER EXTRACT (LIT. DEG KG^{-1})
180
160
140
120
100
80
60
40
0
3
6
7
12
15
18
21
24
27
30
33
36
39
42
SLEEPING DURATION (HRS)

The wort filtration rate fell during the first 15 hours to steeping (figure 9). This is indicative of reducing level of ß-glucanase, (Chukwura, 1989) probably due to leaching of this enzyme (present in the ungerminated grain) into the steeping liquor. From the 15th hour, the wort filtration rate increased, reaching a peak between the 14th and 27th hour and thereafter declined. This suggests that ß-glucanase biosynthesis in the germinating grain is at its maximum when millet is steeped for this duration. The fall in wort filtration rate over the first 15 hours of steeping might also be attributed to the high amylopectin content of millet starch (Twagirmukiza, 1983) and the increasing level of low molecular weight dextrin due to the partial hydrolysis of starch by dextrifying amylase (α-amylase) which has been shown to increase at a faster rate during germination than the saccarifying amylase (Maymudar, et al., 1980). Since the combined action of these two enzymes (diastase) is required for the complete conversion of starch to maltose and dextrose, longer steeping duration would be required for their complete convertion and this tallies with Novellie's (1962) findings that the diastatic power of sorghum grain increased with prolonged steeping.

It would appear, therefore, that 24 hours is the compromise steeping duration for this millet variety since it promotes enhanced wort filtration and adequate grain modification without an excessively high malting loss. This corresponds to a moisture level of 38-40% (figure 4). This is slightly lower than the ultimum moisture levels of 42-48% and 40-60% required for maximum modification intensity of barley and sorghum respectively (Briggs, et al., 1981 Ikediobi, 1989).

The duration of germination had a significant effect on malt quality. The hot and could water extract, the diastatic power and malting loss all rise with inccreased germination duration (figure 10) reaching a peak between the 4th and 6th day. This follows the same profile as the ß-amylase and proteolytic activity. A five days germination period thus seem adequate for ultimum grain modification. Expectedly, increased germination was accompanied by nutrient loss (Figure 3) and this probably accounts for the fall in extract yield after the 6th day of germination (Figure 10). Thus the hot and cold water extract profile (Figure 7 and 8) could be explained in terms of the changing enzyme level suggesting that protein and starch hydrolyses increase the extract yield up to a point and that enzyme and nutrient leaching set in after a prolonged steeping to cause their lowering.

FIGURE IX: WORT FILTRATION TIME VS STEEPING DURATION

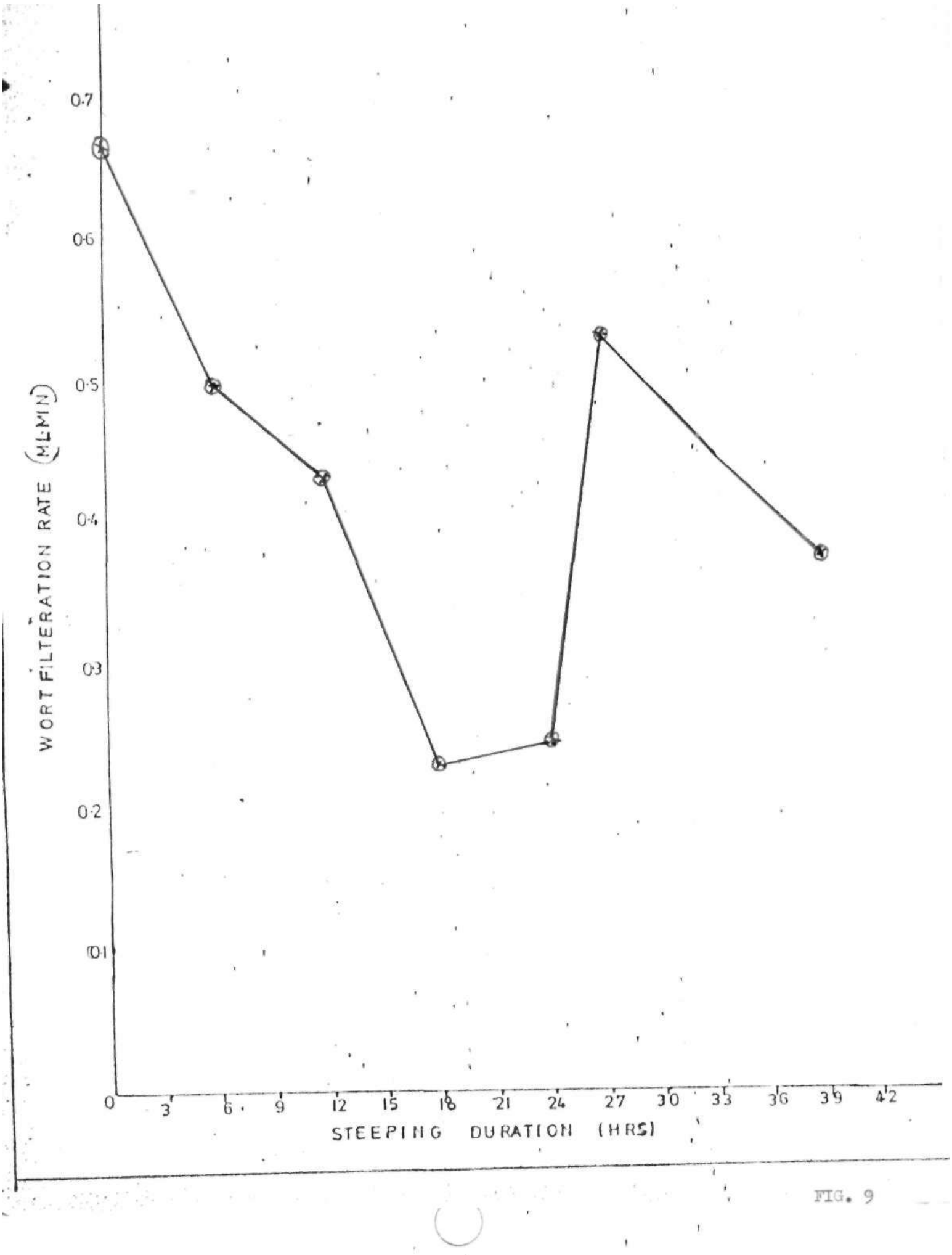

WORT FILTERATION RATE (ML/MIN)
0·7
0·6
0·5
0·4
0·3
0·2
0·1
0
3
6
9
12
15
18
21
24
27
30
33
36
39
42
STEEPING DURATION (HRS)

FIG. 9

FIGURE X: PATTERN OF MALT CHARACTERISTICS OF A LOCAL MILLET VARIETY

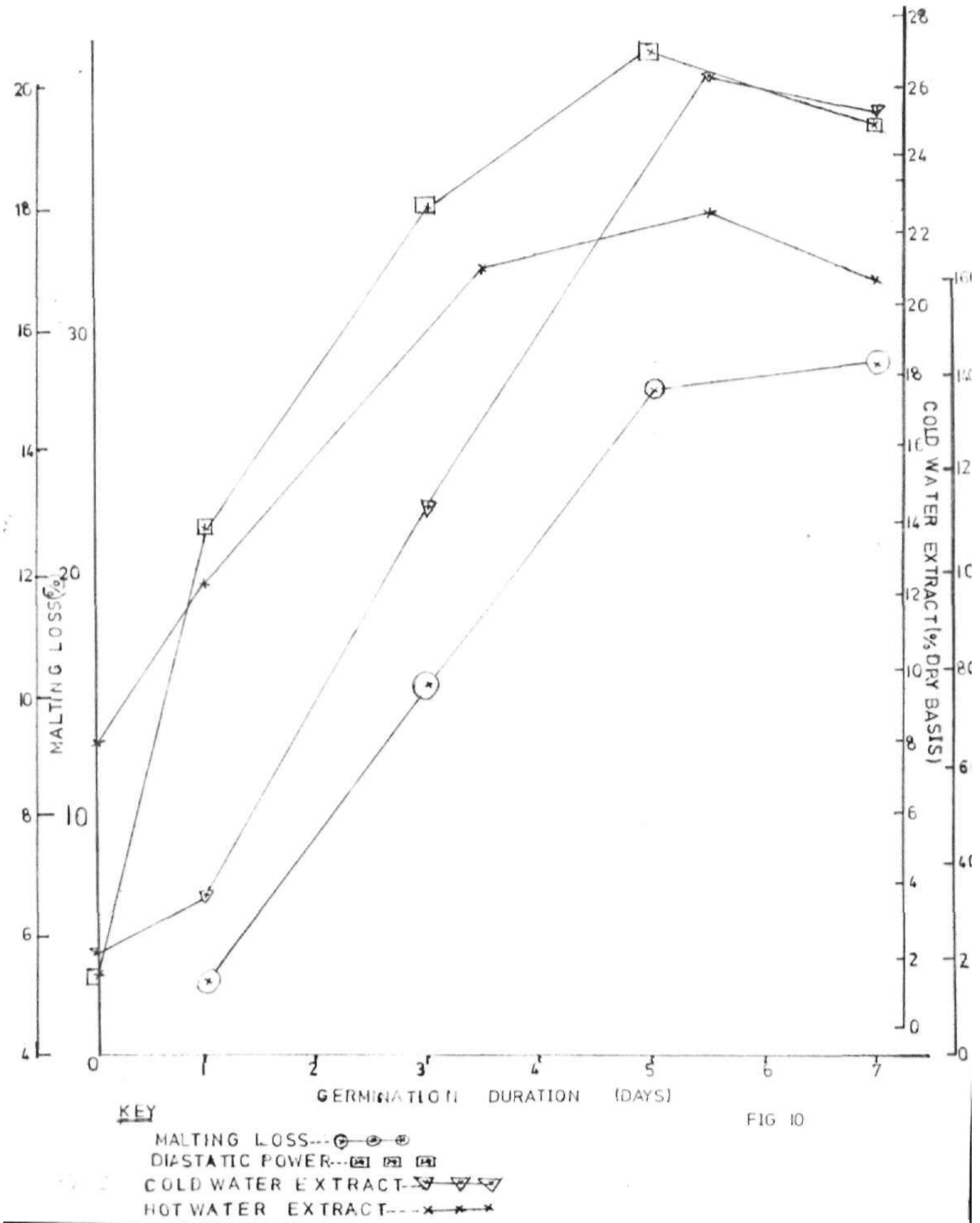

MALTING LOSS(%)
COLD WATER EXTRACT (% DRY BASIS)
GERMINATION DURATION (DAYS)
KEY
MALTING LOSS
DIASTATIC POWER
COLD WATER EXTRACT
HOT WATER EXTRACT

FIG 10

Calcium hydroxide (0.1%) had a significantly positive effect (P>05) on the diastatic power and cold water extract and a significantly negative effects on the malting loss and wort-filtration rate relative to the control (ie, malt from distilled water-steeped millet). However no significant difference in the hot water extract was found (P < 05).

While formaldehyde (0.1%) produced a significantly positive effect on the cold water extract, no significant difference was found with regards to the malting loss, diastatic power and hot water extract. On the other hand, ammonia (0.1%) produced significantly positive effects on the malting loss, diastatic power and wort-filtration rate but a significantly negative effects on the cold and hot - water extracts; while hydrogen-peroxide (0.1%) produced a significantly positive effect on the cold water extract, no significant effect on the malting loss and hot water extract, it had negatively significant effects on the diastatic power and wort-filtration rate. These effects are vividly demonstrated in the bar chart diagrams (figure II).

Thus calcium hydroxide appear to be the most suitable of all the additives studied, given that its effect on diastatic power, the best parameter for assessing malt quality (Novellie, 1967) is significantly higher than those of the other additives (P > 05).

The enhancement of enzyme activity by calcium hydroxide is probably due to its ability to enhance the leaching of phenolic materials (Briggs et al., 1980) thereby reducing enzyme entrapment by these compounds. Beside, calcium ion enhances amylase activity. (Hough, 1976). The reduction in malting loss by formaldehyde has been attributed to its ability to slow down the germination process (Briggs et al., 1981).

Chi-square analysis of the result of sensory evaluation of the millet lager beer (Table 3) indicate a generally high level of acceptance. However commercial star lager beer (control) gave higher acceptability scores (χ^2 = 1.54, 0.24, 0.26, 2.24, 0.9, 1.21, 2.17 and 0.69 for foam, colour, odour, bitterness, body-fulness, Brilliance, sparkling and sourness respectively against a critical value of 3.325). The following attributes of the millet lager beer were accepted: Colour, odour, bitterness, bodyfulness, brilliance and sourness (χ^2 = 0.83, 0.29, 2.35, 1.12, 1.18 and 0.6 respectively), while the foam stability and sparkling were not accepted (χ^2 = 4.044 and 5.16 respectively against a critical value of 3.325).

TABLE 2: RESULTS OF CHI-SQUARE ANALYSIS

	General Acceptability	Foam 2 =	*Colour* 2 =	*Odour* 2 =	*Bitter*ness 2 =	Body Full Ness	Brilli ance 2 =	*Spar*ling 2 =	*Sour*ness 2 =
Star lager Beer (Control)	1.54	2.17	0.24	0.26	2.24	0.95	1.21	2.17	0.69
Millet lager beer	1.80	4.04	0.83	0.29	2.35	1.12	1.18	5.16	0.6

N.B: THE CRITICAL CHI-SQUARE (χ^2) VALUE AT 95% CONFIDENCE LIMIT AND NINE DEGREES OF FREEDOM = 3.325.

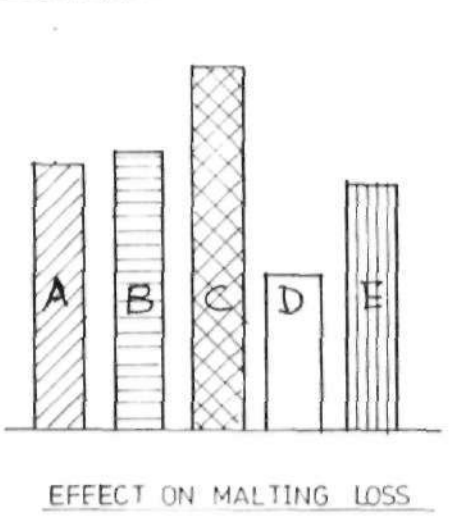
The high acceptability of the colour of millet lager beer is consistent with the low level of phenolic compounds in the millet, since no colour syrup was used. It could also be attributed to the mashing technique employed. According to Narzis (1981) decoction wort is less susceptible to non-biological haze formation due to the increased rate of tannin, lipid and phytate degradation. These molecules react with proteins to impart colour to the wort. However the colour acceptability score was lower for the millet lager beer than that of the control. This was probably due to an increased oxygen uptake during wort-filtration. Other factors which could have influenced the disparity in colour includes the variation in contact time of wort with grain, composition of the brewing water and variation of wort boiling time (De Clerk, 1959). The high acceptability score for odour suggests a low level of diacetyl in the beer, and indicates an efficient temperature control during fermentation. It also reflects the asepsis of the brewing process and yeast purity (De Clerk, 1958).

The poor foam stability of the millet lager beer could be adduced to many causes. First, the manual carbonation process employed (due to the small scale of production) made it technically impossible to carbonate the beer up to the required pressure, since there was no filter counterpresure as is normally the case when beer is mechanically filled into bottle and carbonated. This meant a lowered carbondioxide pressure. It has been demonstrated that a deviation even as small as half a pound in pressure (which represent 0.05 volumes of carbondioxide) is significant (De Clerk, 1958).

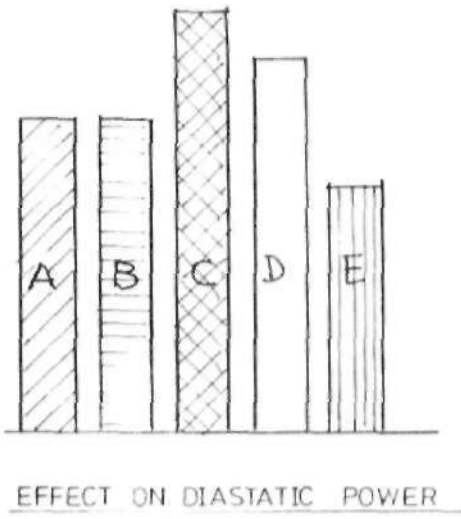
The high acceptance score for bodyfulness of the millet lager beer implies efficient proteolytic activity during mashing since the solubilization of proteins to simpler molecules such as albuminoids, peptones and polypeptides has been shown to contribute to the tatile impression of the beer (Briggs et al., 1981).

The disparity in bitterness score of the millet lager beer and star lager could be attributed to several factors. These include the condition and quantity of hops used, the method of addition, pH etc. The serious wortfilteration problem encountered probably contributed significantly to the lower bitterness score of the millet lager beer since much more of the bitter principles in the wort was probably retained in the foam during filtration (Briggs, et al., 1981).

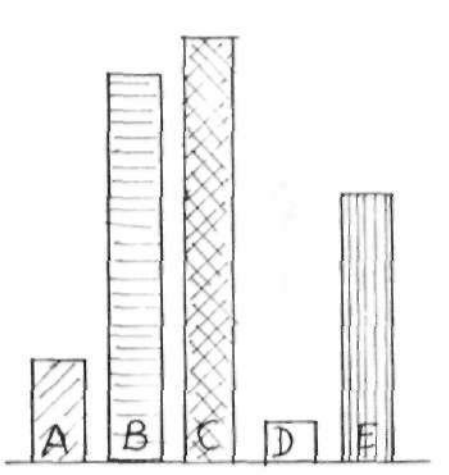
FIGURE XI: EFFECT OF ADDITIVE (IN STEEPING LIQUOR) ON MALT XTERISTICS



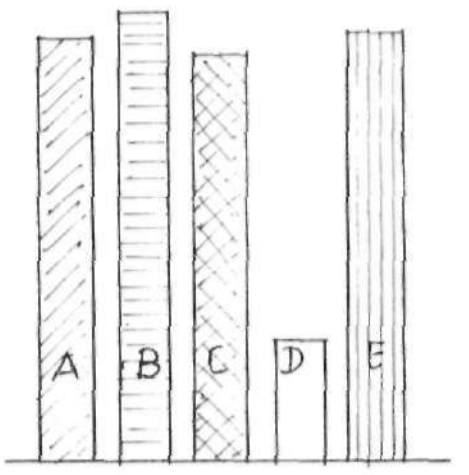
EFFECT ON MALTING LOSS



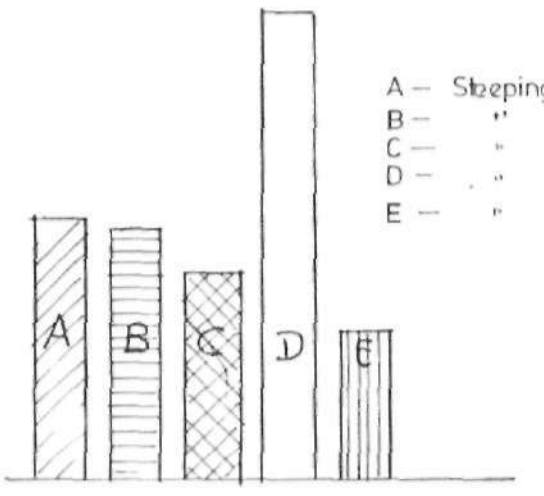
EFFECT ON DIASTATIC POWER



EFFECT ON COLD WATER EXTRACT



EFFECT ON HOT WATER EXTRACT



EFFECT ON WORT FILTRATION RATE

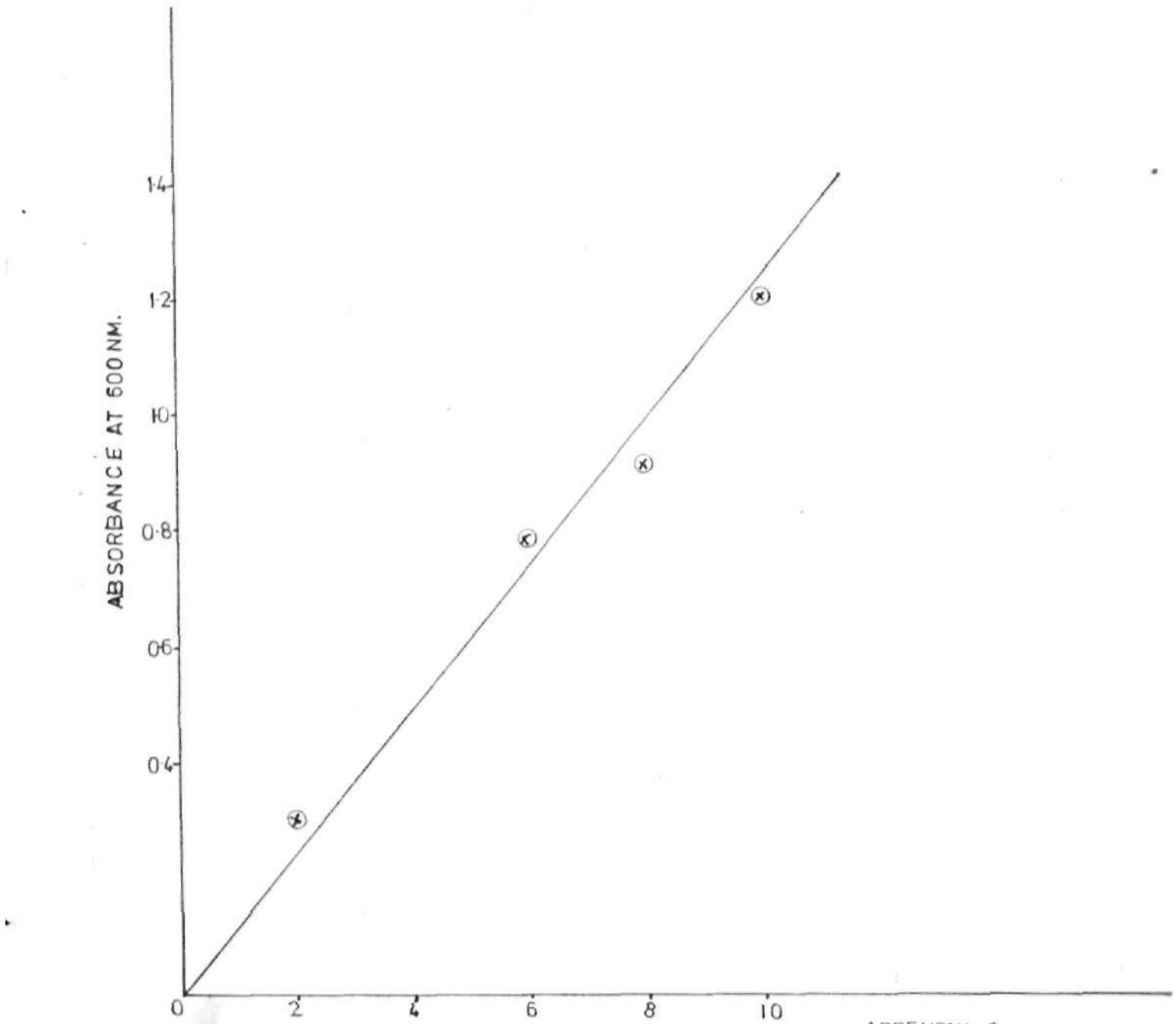
KEY

A — Steeping Liquor is DISTILLED WATER
B — " " " 01% FORMAIDEHYDE
C — " " " " CALCIUM HYDROXIDE
D — " " " " AMMONIA
E — " " " " HYDROGEN PEROXIDE

FIG 11

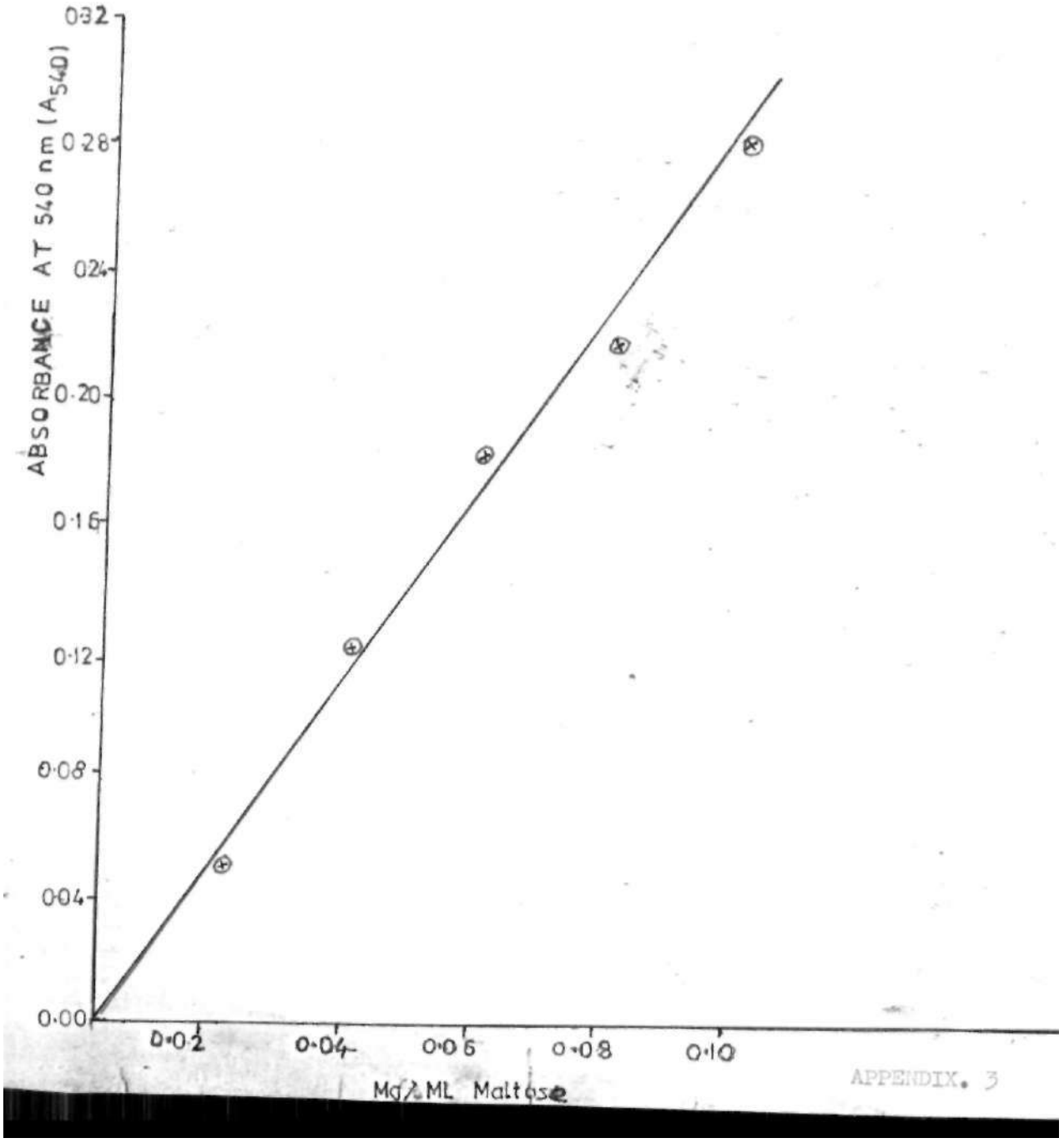
APPENDIX 1: STANDARD CURVE FOR FREE AMINO ACID ESTIMATION

APPENDIX 2: STANDARD CURVE FOR POLYPHENOLS ESTIMATION



APPENDIX. 2

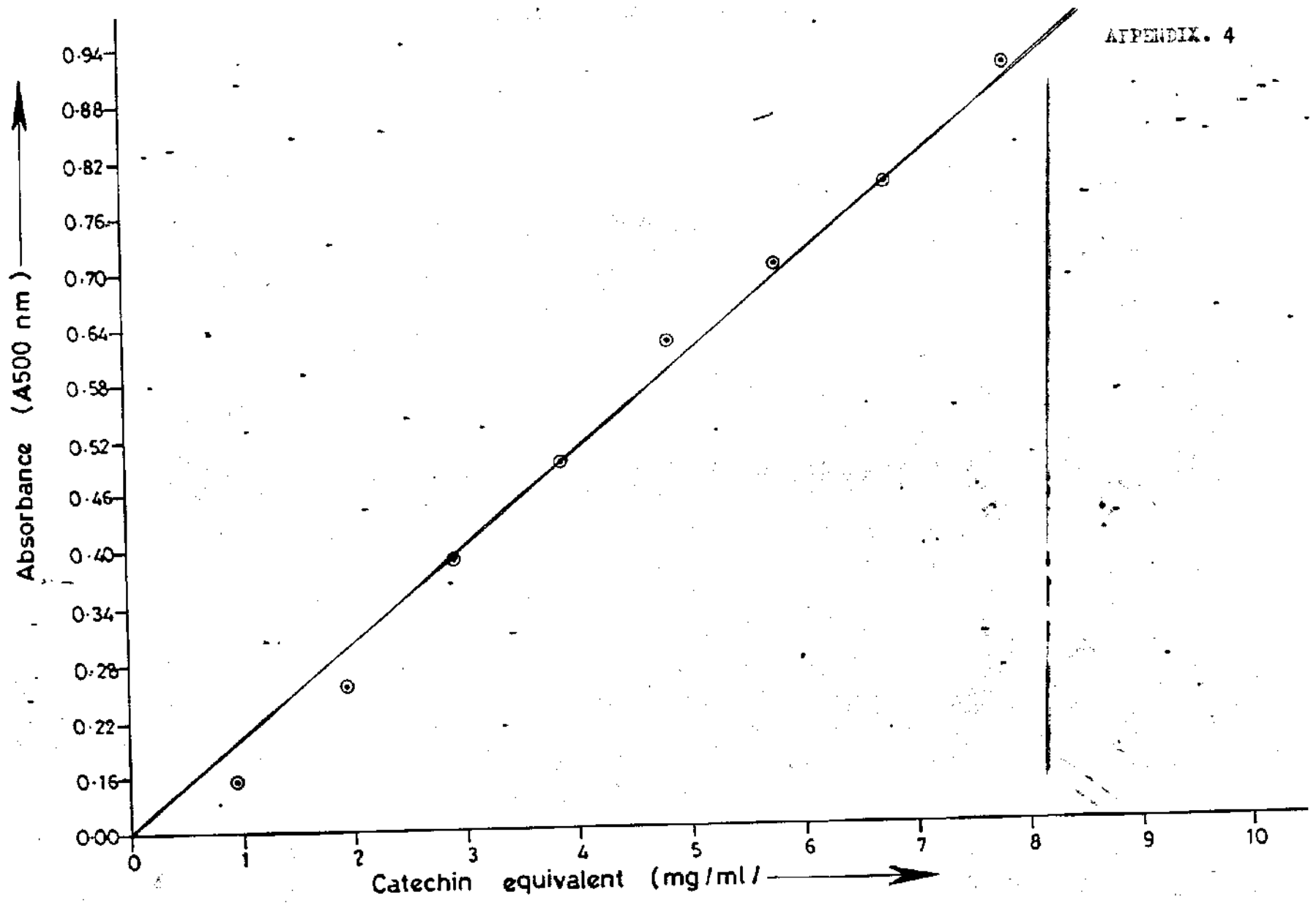
APPENDIX 3: STANDARD CURVE FOR MALTOSE


ABSORBANCE AT 540 nm (A_{540})
0·32
0·28
0·24
0·20
0·16
0·12
0·08
0·04
0·00
0·02
0·04
0·06
0·08
0·10
Mg/ML Maltose

APPENDIX. 3

APPENDIX 4: STANDARD CURVE FOR TANNINS

APPENDIX. 4


Absorbance (A500 nm)
0·94
0·88
0·82
0·76
0·70
0·64
0·58
0·52
0·46
0·40
0·34
0·28
0·22
0·16
0·00
0
1
2
3
4
5
6
7
8
9
10
Catechin equivalent (mg/ml)

APPENDIX 5: EFFECT OF STEEPING DURATION ON MALT QUALITY

	MALT CHARACTERISTICS					
STEEPING DURATION (HRS)	MOISTURE (%)	MALTING LOSS (% DRY WT)	COLD WATER EXTRACT % (DRY)	HOT WATER EXTRACT (DRY) LINTNER DEGREES	DIA-STATIC POWER LI	FILTERA-TION TIME (M1/MIN)
O Hr (Millet grain ie. control)	10.03	-	2.232	60.36	4.56	0.67
6 hrs	6.84	22.35	5.53	171.38	8.42	0.49
12 hrs	9.04	23.74	5.61	166.95	11.06	0.43
18 hrs	7.21	26.47	5.64	161.16	15.4	0.23
24 hrs	8.65	22.40	7.74	159.86	18.65	0.25
27 hrs	4.48	17.35	5.74	120.8	18.69	0.17
33 hrs	3.27	14.61	5.50	120.13	17.13	0.45
39 hrs	5.60	15.74	4.16	118.6	17.04	0.38

APPENDIX 6: EFFECT OF ADDITIVES ON MALT QUALITY

MALT TYPE STEEP MEDIUM ADDITIVE	MALTING LOSS (%)	DIASTATIC POWER (%)	COLD WATER EXTRACT (o_L)	HOT WATER EXTRACT (LIT. DEG KG)	WORT FILTERA-TION RATE (M1/MIN)
Distilled Water (Control)	20.48	15.4	5..61	166.95	0.83
0.1% Formaldehyde	20.98	15.38	22.652	176.924	0.81
0.1% Calcium Hydroxide	28.42	20.6	26.639	158.97	0.68
0.1% Ammonia	11.96	18.1	2.2033	47.159	1.53
0.1% Hydrogen Peroxide	19.37	11.98	15.4	168.20	0.5

APPENDIX 7: RESULT OF PROXIMATE ANALYSIS OF MILLET MALT USING DIFFERENT ADDITIVES AS STEEPING LIQUOR

ADDITIVE	MOISTURE (%)	ASH (%)	FAT % DRY WT	PROTEIN % DRY WT	STARCH % DRY WT	TANNIN %
Distilled water	7.8	1.82	3.47	10.81	54.75	1.88
0.1% Formal Dehyde	7.014	1.81	4.22	9.88	55.69	1.15
Calcium Hydroxide (0.1%)	4.3	1.89	3.91	10.25	59.53	1.24
Ammonia (0.1%)	7.3	1.76	3.47	10.93	63.46	1.76
0.1% Hydrogen Peroxide	4.74	1.74	3.76	11.00	58.25	1.85

APPENDIX 8: EFFECT OF GERMINATION DURATION ON MALT QUALITY

GERMINATION TIME (DAYS)	MALTING LOSS (%)	DIASTATIC POWER (%) (^{o}L)	COLD WATER (% DRY BASIS)	HOT WATER EXTRACT (LIT. DEG. KG^{-1}) DRY BASIS
0 (ie. Millet Grain Control)	-	4.56	2.088	60.37
1	3.17	12.85	3.75	90.84
3	13.53	18.02	14.6	162.90
5	28.42	20.6	26.4	168.97
7	28.45	19.6	24.91	160.26

APPENDIX 9: PROXIMATE ANALYSIS OF MILLET MALT AT VARYING GERMINATION DURATION

GERMINATION DURATION (DAYS)	MOISTURE %	ASH %	FAT %	PROTEIN %	STARCH %
0 Millet Grain ie, control	10.03	1.87	4.25	11.06	66.27
1	10.62	1.88	3.95	10.62	63.4
3	8.9	1.87	3.80	10.46	61.60
5	7.3	1.87	3.6	10.25	58.22
7	7.08	1.88	3.58	10.08	58.10

APPENDIX 10: ORGANOLEPTIC ASSESSMENT OF LAGER BEER

INSTRUCTIONS

Assess the following beer samples glasses (1 to 4) (foam stability, colour, odour, bitterness, body fulness, brilliance, sparkling, sweetness and sourness), and enter the appropriate score for each parameter on the questionnaire provided according to the table shown below:

PARAMETER	GRADE	SCORE
Foam	Excellent	1
	Good	3
	Fairly Good	2
	Fair	1
	Poor	0
Colour	Golden Yellow	4
	Gold	3
	Amber	2
	Brown	1
	Colourless	0
Odour	Very Pleasant	4
	Moderately pleasant	3
	Pleasant	2
	Slightly Pleasant	1
	Unpleasant	0
Bitterness	Moderately Bitter	4
	Slightly Bitter	3
	Very Bitter	2
	Extremely Bitter	1
	Not Bitter	0
Body fulness	Not Watery	4
	Moderately Watery	3
	Watery	2
	Very Watery	1
	Extremely Watery	0

APPENDIX 11: RESULT OF THE SENSORY EVALUATION OF LAGER BEER (CONTROL ie. STAR LAGER BEER)

Panellist	Form	Colour	Odour	Bitter-ness	Body-ful-ness	**Brilli-ance**	Spark ling	Sour Ness	Total Score
1	5	5	5	4	5	5	5	2	36
2	5	5	4	4	4	2	4	5	33
3	5	5	3	4	5	3	3	3	34
4	4	4	4	5	5	5	5	4	36
5	5	4	4	4	4	4	5	4	34
6	5	5	4	4	4	4	5	5	36
7	4	3	4	3	3	3	4	4	28
8	5	3	5	2	5	2	5	5	32
9	5	5	5	4	3	5	3	4	34
10	2	5	3	5	4	5	5	5	35
TOTAL	46	44	41	39	42	39	45	42	338 338

APPENDIX 12: RESULT OF THE SENSORY EVALUATION OF MILLET LAGER BEER

Panellist	Form	Colour	Odour	Bitter-ness	Body-ful-ness	**Brilli-ance**	Spark ling	Sour Ness	Total Score
1	5	3	4	4	4	5	3	4	31
2	1	4	5	4	4	3	2	5	25
3	1	4	3	3	3	4	1	5	22
4	3	4	5	5	5	5	5	5	37
5	2	5	4	3	3	3	2	5	25
6	1	5	4	4	4	3	1	5	27
7	1	4	4	2	2	4	4	3	23
8	1	4	4	4	4	5	1	5	28
9	2	4	4	4	4	3	2	4	26
10	3	5	3	4	4	5	4	5	33
TOTAL	20	42	40	27	37	25	46	46	277 277

APPENDIX 13: CHI-SQUARE ANALYSIS OF THE RESULTS OF SENSORY EVALUATION

Panellist	Total Score For Star Lager (ie Observed)	Total Score For Millet Lager (ie Observed	Marginal Total	Expected (e) Star Lager (ie Control)	Expected (e) Millet Lager	Chi-Square (X^2) Star Lager	Ch-Square (X^2) Millet Lager
1	36	21	67	35.66	29.23	0.0032	0.11
2	33	25	58	30.87	23.99	0.147	0.043
3	34	22	56	29.81	24.43	0.59	0.24
4	36	37	73	38.86	31.84	0.21	0.84
5	34	25	59	31.40	25.74	0.22	0.02
6	36	27	63	33.53	27.48	0.18	0.008
7	28	23	51	27.15	22.25	0.23	0.25
8	32	28	60	31.94	26.17	0.0001	0.13
9	34	26	60	31.94	26.17	0.13	0.001
10	35	33	68	36.19	29.66	0.04	0.38
Marginal TOTAL	338	227	635 (ie Grand Total			1.54	1.80

SUGGESTIONS FOR FURTHER WORK

1. There is need for more work on varietal difference in the malting quality of millet.
2. Once identified the best malting varieties should be subjected to further breeding studies to improve their malting qualities.
3. The millet mash filtration pattern should be studied at the different levels, of exogenous ß-gluconase. It may be necessary to ascertain the effect of ß-glucanase from different sources on millet mash.
4. Hydrolyses profile of millet starch from different varieties should be studied as a function of malt fermentability.
5. There is need for fermentation studies involving different varieties of yeast to establish the most suitable yeast for millet fermentation.
6. There is need for further studies on the various conditions which affect the millet malting (ie. steeping, germination and kilning). These includes temperature, moisture/relative humidity, additives, time, grain maturity etc.)
7. An economic feasibility study on the use of millet malt for lager beer production should be carried out.
8. There is need for a detailed biochemical analysis of millet lager beer produced under standard brewing conditions.
9. A mashing study of millet malt using different mashing techniques is also very important.

Chapter 5

REFERENCES

1. **Adrian and Sayerse (1957)**
   Composition of Senegal Millet and Sorghum. British Journal of Nutrition, 11, 99 - 105.
2. **Aisien, A.O. and Muts, G,.C.J., (1987)**
   Miccro-Scale Malting and Brewing studies of some Sorghum Varieties Inst. Brewing; (93): 328-331.
3. **Aisien A.O. (1988)**
   Sorghum: A suitable source for brewing? Brewing and Distilling International, 18(3): 20-22
4. **Aisien A.O. (1989)**
   Utilization of Sorghum in Lager beer in Nigeria. Paper for the Symposium "Current status and potential for Industrial uses of Sorghum." ICRISAT-WASIP-IAR Workshop 4-6th December, 1989, Kano.
5. **Amerine, M.A. Pangborn, R.M. and Roesslier E.B. (1965):**
   Principles of Sensory Evaluation of food. Acedemic Press, New York.
6. **Anon (1958, 1977).**
   Recommended Methods of analysis of the American Society for Brewing Chemists (A.S.B.C).
7. **Anon (1962)**
   Official Methods of Analysis of the American Association of Cereal Chemists (AACC) Cereal Laboratory Methods 7th Edition Rainbow C and Floats G.E.S. (Ed).
8. **Anon (1977)**
   Recommended Methods of Analysis of the Institute of Brewing (IOB).
9. **Anon (1980)**
   Recommended Methods of Analysis of the Association of Official Analytical Chemists (A.O.A.C).
10. **Anon (1986)**
    Recommended Methods of Analysis of the American Society of Brewing Chemists (ASBC) 7th Edition.
11. **Arri B.K. (1989)**
    Problems Associated with the use of Sorghum for lager beer production: Paper presented on the current status and potential of industrial uses of Sorghum in Nigeria. ICRISAT-WASIP and IAR, 4-6th December, 1989 Kano - Nigeria.
12. **Asenime (1985)**
    Report on Malting Barley Production at the Symposium on "The Nigerian Brewing

Industry, Production, Processing and Utilization of the Local Raw Materials held at Hamburg Germany, November, 1985.

13. **Bishop L.R. (1930)**
J. Inst. Brewing 36, 336 Factors affecting the yield and quality of malt extract from rag, (Eleusine Coracona). As cited by Huylse et al 1980 in Sorghum and the Millets.

14. **Botes D.P., Jourbert F.J. and Novellie L (1967a)**
Kaffircorn malting and brewing studies XVII. Purification and Properties of Sorghum Malt - Amylase. J. Sci Fd. Agric. 18, 409-415

15. **Brrigs, D.E., Hough J.S. Stevens R and Young**
T.W. (1981) Malting and Brewing Science: Malt and Sweet Wort. 2nd Edition.

16. **B.S.C. (Beer Sectorial Group, 1986**
The Brewing Industry: A Major Contributor to Nigeria's development. Beer Sectorial group of the Manufacturers Association of Nigeria.

17. **Burns R.E (1971)**
Methods of Estimation of Tannins In grain Sorghum. Agron, Journal 63, 511-512.

18. **Canales A.M. and Sierra J.A. (1976)**
Uses of sorghum. Master Brew Assoc. Amer. (13): 114.

19. **Chandrasechara M.R. and Swaminathan M (1957)**
Enzymes of rag 1 (Eleusine Cocacona) and ragi malt (II) proteases. As cited by Julse et al 1980 in Sorghum and the Millets.

20. **Chukwura E.N. (1988)**
Recent Development in Sorghum utilization in Nigerian Brewing Industry. Paper presented at a 4-day workshop organized by the Eastern Chapter of the Nigerian Institute of Food Science and Technology at the University of Cross River State, Uyo 27th-29th April

21. **Chukwura E.N. (1989)**
Brew house performance of sorghum grain and malt. Problems and prospects International Crop Research ICRISAT-WASIP - A.B.U. Workshop on Industrial Utilization of Sorghum," 4-6th December, Kano.

22. **Chitre R.G; Desai D.B. and Raut V.S. (1955)**
The Nutritive value of pure brewed Strains of Cereals and Pulses. Part I Thiamine, riboflavin and nicotinic acid content. As cited by Hulse et al (1980) in Sorghum and the Millets. Their Composition and Nutritive value.

23. **Daiber K.H. (1975)**
Enzyme inhibition of polyphenols of Sorghum malt J. Sci Fd. Agric (26), 1399-1411.

24. **De Clerk J. (1958)**

'A textbook of brewing', Vol.I (English Edn) P. 144 (London: Chapman and Hall).

25. **Diagboya P. Omagbemi, O Uba, O and Ebu I (1983)**

Effects of different mashing systems on wort composition and beer quality Brewers Association of Nigeria 2 20.

26. **Egharevba P.N. (1979)**

Agronomic Practices for Improved Millet production. Samaru Misc. Paper No. 90 (1979.

27. **Elizabeth L. (1977) Sensory Evaluation.**

In Norman W.D. (Ed) Elements of Food Technology pp. Avi Publishing Company Inc., Westport, Connecticut.

28. **Fereeman J.E. and Bacon B. J. (1973)**

Pearl Millet a potential crop for wet milling. Cereal Science today 18 69.

29. **Glenie et al (1986)**

Brewing with sorghum. Brewing and Distilling International 18: 21-31

30. **Haase G.W. (1985)**

The production of lager beer by means of sorghum malt. Paper presented on the result of research carried out in West Germany. As cited by Sugris D and Hope, J.J. (1987). In the Technology of Economices of Malting in Nigerian.

31. **Hall R.D; Harris G and McMillan, I (1961)**

Effect of mashing on character of beer. As cited by Hudson J.R. (1969). The brewers digest.

32. **Haris J.O. (1971)**

Haris J.O. (1971) Economics of wort production. Modern Brewing Technology edited by Final W.P.K. McMillan Press London and Basingstock 1, 8, 11-12

33. **Harold M (Ed) 1976**

The practiccal brewer. Master Brewers Association of America 2nd Edition Impressions Inc. Madison, Winsconsin.

34. **Holdt, M. Mand Brand, J. (1960**

"Starch content of Kaffir beer brewing materials" J. Sci. Food Agric, 11th August, 1960.

35. **Hough, J.S; Briggs D.E. and Stevens R (1961)**

The Chemistry and Biochemistry of Mashing Malting and Brewing Science. Chapman and Hall 220 - 233.

36. **Hug. H. and Perininger, H.B. (1979)**

Brewing trials using infusion and Decoction mashing methods. Proc. Eur. Conv. Berlin, 335.

37. **Hulse J.H., Lain E. M and Pearson O.E. (1980)**
Sorghum and the Millets: Their composition and Nutritive Valve. A.P. Inc (London Ltd.

38. **Igyor M.A. (1985)**
The effect of malt and wort composition on Flavour Profiles and Alcohols in Sorghum Beer. Paper presented at the Brewers Association of Nigeria (Northern Zone) quarterly zone meeting at Benue Brewery Limited. Makurdi 3rd June, 1989.

39. **Ihekoronye A.I. and Ngoddy P.O. (1985)**
Integrated Food Science and Technology for topics Macmillam Publishers

40. **Ikediobi C.O. and Ukoha A.I. (1987)**
Enzymology Principles and Application. (In Press)

41. **Ikediobi C.O. (1989)**
Industrial Production of Sorghum malt in Nigeria. Paper presented on Industrial Utilization of Sorghum in Nigeria. ICRISAT - WASIP and IAR (ABU) Zaria 4th-6th December, 1989, Kano- Nigeria.

42. **Inderia, R. and Naik, M.S. (1971).**
Nutrient Composition and Protein Quality of some minor millets Indian J. Agric, Sci. 41, 795-797

43. **Jayatissa R.M; Pantirana R.A. and Sivayosunderam**
K. (1980) Journal of the Institute of Brewing 1980, 86, 18

44. **Jean F.C. (1977)**
Organoleptic Properties of Food. In Norman, W.D. (ed). Elements of Food Technology. Avi publishing company Inc. Westport, Cannecticut.

45. **Joseph J.D. (1977)**
Wort production: The practical Brewer In: Harold M.B. (ed) Master Brewers Association of America 2nd edition Impressions, Inc. Madison, Winsconsin. pp 62-121

46. **Kahan; F. (1973)**
Food Technology Champaign, 27, 61

47. **Kendal M. (1962)**
Rank Correlation Method 3rd Edition, Griffin Press, London.

48. **Kirk Othmer, Encyclopedia of Chemical Technology**
3rd Edition. 3 692-735

49. **Knor, 1965**
The importance of ascorbic acid inbrewing. F. Hoffman-La Roche & Co. Ltd, Basle, Switzer land.

50. **Knudsen, F. B. (1977)**

Fermentation principles and practice M.B.A.A. 2nd Edn. Broderick H. M (Ed)

51. **Kobalch and Schifarth Braneric Wissens Chaffliche**

1950. 3-25. As cited by Hudson J.R. Effect of mashing on the Character of Beer. Brewers digest 98.

52. **Malomo O. (1989)**

Sorghum in lager beer production: Progress and prospects. Paper presented at the International crop Research Institute for Semi-arid tropics (ICRISAT) on Industrial Utilization of Sorghum 4-6th December, Kano-Nigeria

53. **Mayer A.M. and Poljakoff - Mayber (1963)**

The Germination of Seeds. Pergamon Press, Vol.3 Oxford London, N/York Parts 1963

54. **Maxson E. D and Rooney L. W. (1972a)**

Evaluation of Methods for Tannin Analysis in Sorghum Grain Cereal Chem. 49, 719-729

55. **Michael L. (1959)**

Kaffircorn malting and brewing studies. The extraction and nature of the insoluble amylase of kaffircorn malts. J. Sci. Fd. Agric. 5(II): 457-463

56. **Narzis (1981)**

Infusion and decoction processes comparison on small and commercial scale J. Inst. Brew. 121, 768-770.

57. **Novellie L. (1959)**

Kaffircorn malting and brewing studies iii. Determination of amylases in kaffircorn malts. J.Sci. Fd. Agric 10, 441-449.

58. **Novellie L (1962a)**

Kaffircorn malting and brewing studies. ix. Effect of malting conditions on the diastatic power of Kaffircorn malt. J. Sci. Fd. Agric 13, 115-120.

59. **Novellie L. (1962b),**

Kaffircon malting and brewing studies. xii. Effect of malting conditions on malting losses and total amylase activity. J. Sci. Fd. Agric 13, 121-123

60. **Novellie L. (1962c)**

Kaffircorn malting and brewing studies. xiii. Variation of diastatic power with variety, season, maturity and age of grain. J. Sci. Fd. Agric. 13, 124-126.

61. **Novellie L (1977)**

Beverages from sorghum and millets. In proceedings, symposium, sorghum and millets for human food pp 73-77. International association for cereal chemistry, symposium, Institute, London.

62. **Nwasike C. C and Aba D.A. (1986)**

Situation of Sorghum and Millet Production in Nigeria. Avenue for improvement presented at National Accelerated Food Workshop, Feb. 1989, Ibadan.

63. **Obigbemi L.B. (1987)**

Keynote address at the workshop on Cereal Utilization Organized by Institute of Food Science and Technology at Kaduna Polytechnic, Kaduna 25th-26th November, 1989.

64. **Ogundiwin J.O. and Tehinse J.F. (1981)**

Fermentation tests on malted brew. Dist. Inter II.

65. **Ogundiwin J.O. (1989)**

Industrial Utilization of Sorghum in Nigeria ICRISAT-SASIP-IAR (ABU) Workshop. 3rd-6th December, 1989.

66. **Olaniyi O (1984) Symposium:**

8th Annual Conference of the Nigerian Institute of Food Sci and Technology November 18-22, 1984.

67. **Omagbemi C (1983)**

Effects of different mashing systems on wort composition and beer quality Brewers Association of Nigeria Benin City, 2, 20.

68. **Palmer G. H. (1989)**

Journal of the Institute of Brewing 75, 536.

69. **Palmer G.H. and Pathgate G N. (1976)**

Advances in Cereal Sciences and Technology (ed. Pomeranz, Y). American Assoc of Cereal Chem.

70. **Parkrashkina, L.S. and Zherebson, N.A. (1983)**

St Paul. Minnesota. Brewing and Bistilling International Journal 13(6) 33-34

71. **Pomeranz Y (1975)**

Industrial uses of barley. Symposium proceeding, 58th annual meeting of AACC November, 4-8th annual meeting of AACC November, 4-8th, 1975, pp. 380.

72. **Purseglove J.W. (1972)**

"Tropical Crops." Monocotyledon-1.

73. **Rachie, K.O. and Majmudaz J.V (1980).**

Pearl millet. Infusion and deconction processes comparison on small and commercial scale J. Inst. Brew. 121, 768-770.

74. **Ramachandra G; Vivupaksha T. K and Shadaksharans - Wamy M, (1977):**

Relationship between tannin levels and invitro protein digestibility in finger millet (eleusine coracona Gaertn) J. Agric Fd. Chem 25, 1101-1104

75. **Subramanian, V and Jambunathan, R (1989).**

'Industrial Products from Sorghum' paper presented at the symposium "Current status and potential for industrial uses of sorghum in Nigeria" ICRISAT-WASIP Workshop, Kano 4-6th Dec, 1989.

76. **Sugris D and Hope J.J. (1987)**

The Tecchnology and Economics of Malting in Nigeria. Paper Presented at the National Seminar on Brewing Sciencce and Technology organized by OTAGO NIGERIA LTD., NSUKKA: 31st August, to 18th September, 1987.

77. **Twagirmukiza E. (1983)**

Use of sorghum and millet in Rwanda pp 25-35. As cited by Subramanian V and Jambunathan R. Industrial products from sorghum. Symposium, "Current status and potential industrial uses ofsorghum" ICRISAT-WASIP-IAR workshop 4-6th December, 1989.

78. **Vitayathil P.J. and Murthy G.S. (1972)**

New reaction of O. Benzoquinone at the thioester group of methionine. Nature, New biology, 236, 101

79. **Vuataz, 1966/67**

Nestle Research News, 57

80. **Williams A.A. (1978)**

Int. Flavours and Additives, 9, 171.

81. **Williams and Carter and Langron, (1979)**

The fourth wine subject day p 51. Long Ashton (Ed.).

82. **Wright, D.L. (1976).**

Understanding Statistics: An introduction to behavioural Science•Harcourt Brace Jovanorich.