PELEMBE, LOUIS AUGUSTO MUTOMENE

PEARL MILLET MALTING: FACTORS AFFECTING PRODUCT QUALITY

Ph.D. (Food Science) U.P. 2001

PEARL MILLET MALTING: FACTORS AFFECTING PRODUCT QUALITY

by

LOUIS AUGUSTO MUTOMENE PELEMBE

Submitted in partial fulfillment of the requirements for the degree

Doctor of Philosophy (Food Science)

Department of Food Science

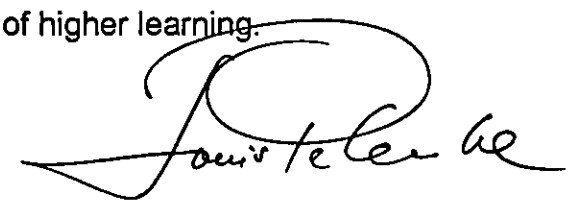
Faculty of Natural and Agricultural Sciences

University of Pretoria

Pretoria

November 2001

I declare that the thesis herewith submitted for the Ph.D. (Food Science) degree at the University of Pretoria, has not been previously submitted by me for a degree at any other university or institution of higher learning.



DEDICATION

I dedicate this Ph.D. thesis to my friend for life ***Lucília "Lucy"*** for her love and encouragement.

I also dedicate this thesis to my late father, ***Augusto Mutomene Pelembe "Wata Funucula/Vovo Mbito"***, (20/09/1933 - 26/02/1997). I wish you were here to share this.

"The road of success is always under construction"

- Ghodonyana Waka Sibiya (04/03/1964 ---)

ACKNOWLEDGEMENTS

The author wishes to express his sincere gratitude and appreciation to the following persons and institutions for their contribution to the successful completion of this study:

Prof. John RN Taylor, University of Pretoria, my supervisor and mentor, for his constant motivation, invaluable guidance, expert advice and personal encouragement throughout my studies. Thanks for your patience and kindness. Dr. Janice Dewar, CSIR, my co-supervisor, for her technical support, constructive criticism and invaluable advice particularly during the last part of this project.

The Universidade Eduardo Mondlane (UEM), Maputo, for allowing me to continue my post-graduate studies. Special thanks to Prof. Dr. Eng. Gabriel L. Amos, Dean of the Faculty of Engineering, for his support. The Capacity Building Project at UEM for awarding me its bursary for my Ph.D. studies.

Prof. A. B. Obilana, Dr. E. Monyo and Ms. Effie Chinhema of SADC/ICRISAT, Matopos Research Station, Bulawayo, Zimbabwe, for the donation of pearl millet of variety SDMV 89004 used in this research.

CSIR – Bio/Chemtek (Food Division) for providing the malting facilities. Ms. Carin Carstens of CSIR – Bio/Chemtek (Division of Food, Biological and Chemical Technologies) for her suggestions and inputs during the malting stage.

Mr. Allan N. Hall of the Laboratory for Microscopy and Microanalysis, University of Pretoria, for his assistance with microscope analysis.

The entire staff and post-graduate students here at the Department of Food Science, University of Pretoria for being helpful in many different ways.

A special thanks to all sensory panelists for their patience and perseverance. My heartfelt gratitude to Miss Paballo Gloria Mokoena for her assistance in sensory preparation.

My wonderful wife, Lucília, and my beautiful children, Jennifer Moyass "Moya-de-papá" and Melanie Lucy "Dona Pukurucha/Vovó Gaveya", who gave me the encouragement I so often needed and give me so much to live for.

My admirable mother, Violeta, who gave me the motivation and support, needed to get the most out of life, for her continuous support. This thesis may be the result of years of study on my part but it is the culmination of a lifetime of love and care on hers.

My brothers, Calvin (Camilo), Buta, Aduto, Marito "Sr. Pelembe" and friends for all the support they offered during the course of this project.

Finally, to all not mentioned here but without their support could not possible to finish this project. I owe you a special thank you. Khanimambo[1]!

[1]Khanimambo /kaneemam:boo/n - thank you in southern Mozambican languages.

ABSTRACT

PEARL MILLET MALTING: FACTORS AFFECTING PRODUCT QUALITY

by

Louis Augusto MUTOMENE PELEMBE

Supervisor: Prof. John RN Taylor
Co-Supervisor: Dr. Janice Dewar
Department: Food Science
Degree: Ph.D. (Food Science)

Pearl millet (*Pennisetum glaucum* (L.) R. Br.) is a drought tolerant cereal crop grown primarily as a food grain in southern Africa. In this southern Africa region, the grain is traditionally processed either by germination or fermentation prior to consumption. Malting involves the limited germination of cereal grain in moist air under controlled conditions.

Malts were prepared by malting two varieties of pearl millet, SDMV 89004 and SDMV 91018. The grain was steeped for 8 h with a cycle of 2 h wet and 2 h dry (air rest) and germinated at four temperatures, 20 °, 25 °, 30 ° and 35 °C over 5 days. The malts were then dried at 50 °C for 24 h.

Modification of starch granules and protein bodies in pearl millet grain structure due to germination was found to start at the germ-floury endosperm interface and move in the direction of the peripheral endosperm. Aleurone layer, cell wall and vitreous endosperm were not greatly involved in modification process.

Ungerminated pearl millet grains do not exhibit Diastatic Power (DP), α- or β-amylase activity. DP, α- and β-amylase activity increase as germination time and temperature increases. DP, total and soluble β-amylase activity increase with germination time and watering treatment probably because high moisture promotes high metabolic activity.

Free amino nitrogen (FAN) increases as the germination time, temperature and watering treatment increases. This may be related to the fact that high temperature and moisture promote the growth of roots and shoots, which are a good source of malt FAN.

Malt extract increases with germination time and watering treatment. This increase in hot water extract is an indication of the progress of modification of the malt during the germination process. The increase in malting loss with germination time, temperature and watering treatment observed is related to the high respiratory activity during germination.

A germination temperature of 25-30 °C and germination time of 3-5 days, medium watering treatment are optimum for pearl millet. These conditions result in high DP, α- and β-amylase activity, good FAN and moderate malting loss. The levels of DP, FAN, α-amylase activity and malting loss of pearl millet malts, which are similar to sorghum malts, represent an excellent potential for utilisation of pearl millet malt for sorghum beer brewing purposes. Additionally, pearl millet malt could be a better alternative than sorghum for lager beer brewing due to the fact that it has higher β-amylase activity.

Phytic acid decreases during malting, probably due to phytase activity. Soluble proteins and the Nitrogen Solubility Index increase due to partial hydrolysis of storage proteins by endogenous proteases. This is complimented by an increase in *in vitro* protein digestibility of pearl millet malts. A reduction in the viscosity of flours made from pearl millet malts, which is due to increased α-amylase activity, may contribute to the use of this malt to improve the energy and nutrient density of porridges for young children.

Germination significantly reduces the mousy odour, characteristic of ground pearl millet meals when stored. This is probably due to the growth of lactic acid bacteria which decrease the pH in the grain affecting the water soluble phenolics which leached out. These phenolics are believed to be responsible for the mousy odour of the stored pearl millets meals.

Pearl millet malt represents an excellent potential for utilisation of pearl millet for sorghum beer and it appears that it can be used in lager beer brewing. The improved nutritional and functional properties of pearl millet malt are an indication that the malting process, a low-

cost processing technology, usable at both rural and industrial level, can be successfully applied to prepare nutritious and functional food products.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER 1

INTRODUCTION

Pearl millet (*Pennisetum glaucum* (L.) R. Br.), known as "mexoeira" (Portuguese), "hanzelo" (Rhonga), "whahuva" (Shangaan) and "mhala" (Xitswa) in Mozambique, as "babala", "manna" (Afrikaans), "nyalothi" (southern Sotho), "inyouti" (Ndebeli), "mhuga", "mhungu" (Shangaan), "unyaluthi", "unyawothi" (Zulu), "leötsa" (northern Sotho) and "lebelebele" (Tswana) in South Africa, and as "mapfunde" (Shona) in Zimbabwe is a drought tolerant cereal crop grown primarily as a food grain in Southern Africa. Data from the FAO (1998) refers that although Mozambique produces only 11% of the total millet production of the Southern Africa, the utilisation of pearl millet for human consumption in this country is about 90% of the respective production, which is the highest in the region.

One of the main constraints in utilization of pearl millet in the industry is the small size of the grain. It also has a large germ which gives rise to the rapid development of fatty acids in whole pearl millet meal, mainly due to the action of lipase (Lai & Varriano-Marston, 1980a) which causes bitterness and makes meal unacceptable within 5 to 8 days after milling (Kaced, Hoseney & Varriano-Marston, 1984). Ground pearl millet also generates a mousy odour when stored for any length of time. Reddy, Faubion & Hoseney (1986) found that this odour is not associated with oxidative rancidity of kernel lipids, but with enzymatic deterioration, as the odour generation requires relatively high moisture levels in the grits.

The utilisation of millets is also limited due to the presence of various anti-nutrients, poor digestibility of proteins and carbohydrates, and low palatability (Sharma & Kapoor, 1997). Various processing technologies are, however, able to affect positively the physicochemical composition of food grains, in

order to improve their nutritional value. Such primary processing technologies include fermentation and malting. Malting is a primary processing technology which describes the process of soaking or steeping the dry grains in water until they are saturated, followed by germination under controlled conditions for a specific period (reviewed by Chavan & Kadam, 1989). Malting is relatively easy and can be carried out without sophisticated equipment.

The process of malting (sprouting) is commonly used for the production of traditional foods. Malting improves the vitamin content of pearl millet and lowers levels of lipids, phytate, and oxalate (Opoku, Ohenhen & Ejiofor, 1981). The concentration of free sugars, amino nitrogen, B vitamins, and ascorbic acid are increased as well (Hamad & Fields, 1979), due to the partial loss of soluble carbohydrates by respiration. The partial protein digestion via intrinsic grain enzymes or extrinsic (microorganisms) proteolytic enzymes improves protein quality and digestibility (Serna-Saldivar, McDonough & Rooney, 1990).

1.1 STATEMENT OF THE PROBLEM

With the continued increase in the Mozambican and southern African population great emphasis has been placed throughout the region on increasing the production of plant foods, improving their nutritional quality, and developing simple and economical methods for their storage and processing. Therefore, malting technology appears to be one of the low-cost technologies that could be used for nutritional improvement of cereal grains such as pearl millet.

In Mozambique and other parts of southern Africa pearl millet is often traditionally processed by sprouting prior to consumption. Sprouting is used to make weaning foods for infants. Malted pearl millet is also brewed to make "uphutsu" and other low alcohol beverages, consumed specially during

traditional ceremonies and other occasions. These low-alcoholic beverages are largely consumed not only during traditional ceremonies, like Masses and weddings "lobola", but also by the low-income groups, which are increasing due to financial-economical reasons.

For commercial brewing, much of the malt used in Mozambique and other SADC (southern African Development Community) countries is from barley. Since barley cultivation in the SADC region is not generally economically feasible; the barley malt used is mostly imported from overseas which makes it very expensive. Therefore, pearl millet could be an alternative for increasing malt availability for both traditional and industrial use at low cost in the SADC countries.

1.2 OBJECTIVES

The objectives of this project were:

- To develop and optimise pearl millet malting conditions suitable for the southern African food industries, with special emphasis on weaning foods, other starchy foods, traditional and conventional brewing.

- To determine the pearl millet grain structure and modification occurring during and due to malting.

- To determine the effects of malting on the physico-chemical, nutritional and functional properties of pearl millet.

- To determine the effect of malting on anti-nutrients in pearl millet, i.e. phytate and polyphenols.

- To determine the effects of malting on the off-odour which appears in pearl millet when ground.

CHAPTER 2

LITERATURE REVIEW

2.1 MORPHOLOGY OF PEARL MILLET

The structure of the mature pearl millet caryopsis is very similar to that of sorghum caryopsis but with several differences (reviewed by Serna-Saldivar & Rooney, 1995). In comparison with sorghum, pearl millet is a smaller grain with a proportionally larger germ and consequently a smaller endosperm (Abdelrahman, Hoseney & Varriano-Marston, 1984). Pearl millet grain is reported to consists of 73.9-76.2% endosperm, 15.5-17.4% germ, and 7.2-10.6% pericarp (Table 1) (Abdelrahman, Hoseney & Varriano-Marston, 1984).

The pericarp of pearl consists of the epicarp, mesocarp, and endocarp and is of variable thickness. Cultivars with thick pericarps do not have starch granules in the mesocarp (reviewed by Serna-Saldivar & Rooney, 1995).

The sub-aleurone endosperm has a very dense protein matrix with only small starch granules in the first one or two cell layers (reviewed by Serna-Saldivar & Rooney, 1995). The corneous (horny or vitreous) area contains large, uniformly sized polygonal starch granules embedded in a protein matrix with small numbers of protein bodies. The respective average sizes of starch granules and protein bodies are 6.4-7.6 and 0.6-0.7 μm (reviewed by Serna-Saldivar & Rooney, 1995).

As stated, the pearl millet germ is proportionally larger than most other cereals. It contains an embryo and scutellum. Pearl millet scutellar epidermal or epithelial cells are very similar to those of sorghum (Zeleznack & Varriano-Marston, 1982). A dark pigmented material (black layer) is deposited

in the basal adgerminal (chalazal cells) surface of the grain during seed development (Fussell & Dwarte, 1980).

2.2 CHEMICAL COMPOSITION OF PEARL MILLET

Compared to sorghum and finger millet, pearl millet is notable for its relatively higher protein and oil levels, which are due to the large proportion of germ to endosperm (Tables 1 and 2) (reviewed by Serna-Saldivar & Rooney, 1995).

The starch content of pearl millet varies from 56 to 65% and the amylose content of the starch ranges from 17 to 29% (McDonough & Rooney, 1985). Table 3 summarises the starch properties of pearl and finger millets. Pearl millet starches have a higher amylose content and lower gelatinisation temperature than finger millet. The initial and end-point temperatures of starch gelatinisation are 59-63 °C and 68-70 °C, respectively (McDonough & Rooney, 1985). Pearl millet appears to require a higher temperature to initiate pasting or to develop viscosity than other cereal starches (Abd Allah, Mahmoud, El-Kalyoubi & Abou Arab, 1987). The water holding capacity of its starch is higher than that of sorghum but lower than that of maize. The swelling power and solubility are also higher than that of other starches (Abd Allah, Mahmoud, El-Kalyoubi & Abou Arab, 1987).

TABLE 1- Chemical composition of pearl millet and its anatomical parts

Component	Whole Grain (%)	Endosperm (%)	Germ (%)	Pericarp (%)
Whole kernel	100	75.1	16.5	8.4
Range		73.9-76.2	15.5-17.4	7.2-10.6
Protein	13.3	10.9	24.5	17.1
Percentage of total protein	100	59.5	31.2	9.4
Fat	6.3	0.5	32.2	5.0
Percentage of total fat	100	6.2	87.8	5.9
Ash	1.7	0.3	7.2	3.2
Percentage of total ash	100	13.9	72.2	13.9

Source: Abdelraham, Hoseney & Varriano-Marston (1984)

TABLE 2- Nutrient composition (proximate composition) of sorghum and millets

Cereal	Protein (%)	Fat (%)	Crude Fibre (%)	Ash (%)	NFE (%)	Starch (%)
Sorghum	11.0	3.2	2.7	1.8	81.3	70.8
Range	7.3-5.6	0.5-5.2	1.2-6.6	1.1-4.5	68.1-89.9	55.6-75.2
Pearl millet	14.5	5.1	2.0	2.0	76.4	71.6
Range	8.6-19.4	1.5-6.8	1.4-7.3	1.6-3.6	62.9-86.9	63.1-78.5
Finger millet	8.0	1.5	3.0	3.0	84.5	59.0
Range	6.9-10.9	1.0-4.6	2.0-6.8	2.3-3.9	73.8-88.7	ND

Source: Reviewed by Serna-Saldivar & Rooney (1995); NFE- Nitrogen-free extract; ND- Not determined.

TABLE 3- Properties of millet starches

Property	Pearl millet	Finger millet
Gelatinisation temperature (°C)	61-69	65-76
Amylose relative (%)	17.0-21.5	15-16
Amylopectin relative (%)	78.9-83.0	84-85
Starch		
Granule size (μm)	4.0-12.0	10-16
Form	Polygonal, round	Polygonal, round
Hilum	Large	Centric

Source: Reviewed by Serna-Saldivar & Rooney (1995).

The protein content of pearl millet varies widely; values ranging from 6 to 23% have been reported, with an average of from 10 to 13% (Lásztity, 1984). Concerning protein distribution between the different morphological parts of the kernel, the same rules are valid as in other cereal grains. The storage proteins (prolamin and glutelin) predominate (over 60% of the total protein). The albumin fraction averages 15% and the globulin 9% (Lásztity, 1984). The differences in amino acid composition between different morphological parts of the kernel are similar to other cereal grains (Lásztity, 1984). The amino acid composition of the proteins of pearl and finger millet is given in the Table 4. Among the most common tropical cereal crops, pearl millet is known to contain a higher protein content (Table 2) and better amino acid balance (Table 4) than sorghum (reviewed by Serna-Saldivar & Rooney, 1995). The higher ratio of germ to endosperm is responsible for the higher protein (Table 2), albumin, and globulin contents and improved amino acid composition. As in other cereals, the albumins and globulins are rich in lysine and tryptophan, whereas the prolamins are low in these essential amino acids (Table 4). According to Lásztity (1984), pearl millet prolamin apparently differs

markedly from that of the other cereals, being unusually high in tryptophan, although like that of other cereals pearl millet prolamin is rich in glutamic acid and proline and deficient in lysine (Table 5).

Cereal grains are rich source of dietary fibre. Most of the dietary fibre of millets is insoluble. Therefore the pearl millet fibre may decrease transit time and prevent gastrointestinal problems (reviewed by Serna-Saldivar & Rooney, 1995). The soluble dietary fibre is low which probably does not reduce blood cholesterol and arteriosclerosis as oat fibre does (reviewed by Serna-Saldivar & Rooney, 1995).

Osagie & Kates (1984) reported that the lipid content of pearl millet was 7.2% and consisted of 85% neutral lipids, 12% phospholipids, and 3% glycolipids. Neutral lipids contained approximately 85% triglycerides and small amounts of mono- and diglycerides, sterols, and free fatty acids.

Pearl millet is reported as being the cereal grain that most rapidly develops off-odours and flavours after milling (reviewed by Serna-Saldivar & Rooney, 1995). The reasons for the rapid deterioration are as follows: 1) high lipid content (Kaced, Hoseney & Varriano-Marston, 1984); 2) higher amounts of unsaturated fatty acids than other cereals; 3) insufficient naturally occurring anti-oxidants, and 4) high enzymatic-hydrolytic activity (reviewed by Serna-Saldivar & Rooney, 1995).

The off-odour precursor in pearl millet was found to be methanol soluble and had characteristics similar to apigenin, the aglycone of the major C-glycosylflavone present in pearl millet (Reddy, Faubion & Hoseney, 1986). However, Seitz, Wright, Waniska & Rooney (1993) reported that the mousy odour from raw pearl millet was due to 2-acetyl-1-pyrroline.

Millets are important sources of minerals and vitamins. The pericarp, aleurone layer, and germ are rich sources; therefore, in refined millet products part of these important nutrients will be lost (reviewed by Serna-

Saldivar & Rooney, 1995). Malting and fermentation, both primary processing technologies, are known to significantly increase phosphorus availability due to increased phytase activity (reviewed by Serna-Saldivar & Rooney, 1995).

TABLE 4- Amino acid composition of the proteins of millets (g/16 g N)

Amino Acid	Pearl Millet	Finger Millet
Essential		
Phenylalanine	4.4-5.6	4.4-8.4
Histidine	1.8-2.6	1.5-4.0
Isoleucine	3.6-5.9	3.8-8.5
Leucine	8.0-25.1	9.2-16.2
Lysine	1.7-6.5	2.6-5.5
Methionine	1.5-2.9	1.3-4.3
Threonine	1.2-4.8	3.5-5.8
Tryptophan	1.1-2.8	1.0-1.7
Valine	4.8-7.0	5.8-10.4
Non-essential		
Aspartic acid	4.9-10.3	6.5-10.0
Glutamic acid	12.3-25.4	20.3-37.8
Alanine	7.5-10.5	5.9-8.9
Arginine	3.2-8.1	3.8-8.2
Cystine	0.7-2.8	0.7-2.9
Glycine	2.8-5.8	3.6-5.9
Proline	5.9-14.2	4.2-10.1
Serine	3.7-5.6	5.1-8.7
Tyrosine	1.7-4.8	2.0-5.6
Amino acid score (%)	60.6	68.0

Source: Reviewed by Serna-Saldivar & Rooney (1995).

Millets are good sources of B vitamins, except for vitamin B-12 (Gazzaz, Rasco, Dong & Borhan, 1989). Dried, matured kernels do not contain vitamin C. The B vitamins are concentrated in the aleurone layer and germ (reviewed by Serna-Saldivar & Rooney, 1995). The alkali treatment used to produce tortillas, maize-based flat breads, which are traditional to Latin America, or *tô*, a food gel, improves bioavailability because the glycosidic bond that renders niacin unavailable is alkali labile. Malting increases the amount of B vitamins and their availability (reviewed by Serna-Saldivar & Rooney, 1995).

TABLE 5- Amino acid composition of the storage proteins of pearl millet (g /16g N)

Amino Acid	Prolamin	Glutelin
Lysine	1.66	2.14
Histidine	2.14	1.64
Arginine	3.04	4.92
Aspartic acid	7.48	6.36
Glutamic acid	22.24	21.68
Serine	6.12	5.35
Threonine	3.46	2.92
Cystine	1.42	1.34
Methionine	1.02	1.34
Phenylalanine	3.82	4.94
Glycine	1.23	2.74
Alanine	9.41	10.42
Valine	3.24	4.12
Proline	10.23	9.42
Tyrosine	4.62	4.44
Isoleucine	3.32	3.83
Leucine	12.06	10.18
Tryptophan	2.84	0.69

Source: Lásztity (1984)

Fat soluble vitamins are mainly located in the germ of pearl millet. It is a good source of tocopherols (vitamin E); but only kernels with yellow endosperm contain some provitamin A activity (reviewed by Serna-Saldivar & Rooney, 1995).

Non-germinated cereals indigenous to the tropical and subtropical areas of the world such as pearl millet and sorghum have no more than traces of either of the starch degrading enzymes α- and β-amylase (Novellie & De Schaepdrijver, 1986). Good amylase activity of grain is desirable to obtain solubilisation of starch and its subsequent conversion to maltose (Jain & Date, 1975). Germination leads to the production of both amylases with α-amylase predominating. The diastatic activity of pearl millet is between 1.18-5.65 times higher than that of barley (Jain & Date, 1975). Sheorain & Wagle (1973) reported that β-amylase activity in pearl millet reaches a maximum value after 30 h of germination whereas at 72 h it is almost equal to that at zero time. Later, Pal, Wagle & Sheorain (1976) reported that barley malt has higher amount of maltose than pearl millet and that both pearl millet and barley malts have comparable amylolytic as well as proteolytic activities. These authors did not find significant differences in enzyme activities of the two malts. Sheorain & Wagle (1973) reported that α-amylase activity is eight to fifteen times greater in pearl millet malt than in wheat malt. Millet amylase shows higher amylolytic action on wheat starch than on millet starch (Klopfenstein & Hoseney, 1995). High amylase activity in millet flour probably is responsible for its improving effect in wheat flour breads (Klopfenstein & Hoseney, 1995).

2.2.1 Nutritive Value of Pearl Millet

The nutritional properties of pearl millet have received more attention than those of the other common millets, because it is the largest-seeded, most widely grown type (Hoseney, Andrews & Clark, 1987).

Protein digestibility and lysine content of pearl millet are higher than those of sorghum and comparable to those of maize (Serna-Saldivar, McDonough & Rooney, 1990). In contrast to sorghum, pepsin digestibilities of pearl millet and maize do not decrease as much upon cooking (Serna-Saldivar, McDonough & Rooney, 1990).

Although lysine is the most limiting amino acid in pearl millet protein, quite a range of concentrations has been reported, with values at the higher end of the range (3.6 g/100 g protein) as high as that for opaque-2 (high lysine) maize (Badi, Hoseney & Casady, 1976). Except for its lysine deficiency, pearl millet has well-balanced protein, with higher concentration of threonine and lower (but adequate) leucine than sorghum protein (Klopfenstein & Hoseney, 1995). Tryptophan levels are generally higher in pearl millet than in other cereals (Chung & Pomeranz, 1985).

Other important nutritional aspect of pearl millet is the fact that this grain cereal is rich in polyunsaturated fatty acids (PUFAs), which are believed to lower blood cholesterol levels (Potter & Hotchkiss, 1995).

2.2.2 Antinutrients in Pearl Millet

Like other cereals, pearl millet is reported to contain considerable amounts of phytic acid, representing more than 70 % of the total phosphorus in the grain (Chauhan, Suneja & Bhat, 1986). The antinutrients, mostly polyphenols and tannins, present in pearl millet are concentrated in the bran (reviewed by Klopfenstein & Hoseney, 1995).). Hulse, Laing & Pearson (1980) reported phytic acid levels from 208 to 246 mg in finger millet and from 170 to 470 mg in proso millet.

In general, the phytic acid is destroyed during pearl millet grain germination (Sebolev, 1962 according to Klopfenstein & Hoseney, 1995) when the enzyme phytase is synthesized and activated. After 48 h germination with devegetation of the malt, the percentage of phosphorus as phytate phosphorus in pearl millet decreased from 38 to 20% (Malleshi & Desikachar, 1986a). However, the reduction in phytic acid found by these authors may have been due to the fact that some of the phytic acid was removed with the roots and shoots during the devegetation process.

Pearl millet also contains oxalic acid (Opoku, Ohenhen & Ejiofor, 1981), which forms an insoluble complex with calcium, thereby reducing biological availability of the minerals (Whitney, Cataldo & Rolfes, 1987). Malting decreases the levels of oxalate from 0.520 to 0.068% (Opoku, Ohenhen & Ejiofor, 1981).

Epidemiological studies carried out in Sudan and reported by Klopfenstein, Hoseney & Leipold (1983) have suggested that pearl millet might be at least partly responsible for the higher goitre incidence in that country. It was then suggested that in pearl millet, the goitrogen is thioamide and/or other compounds derived from flavonoids such as C-glucosylflavones, vitexin, glucosylvitexin, and glucosylorientin (Birzer & Klopfenstein, 1988). Goitrogen is mainly found in the bran (Klopfenstein, Hoseney & Leipold, 1983b) and it is

believed to produce goitre due to the inhibition of the normal conversion of thyroxine (T4) to triiodothyronine (T3) (reviewed by Serna-Saldivar & Rooney, 1995). Hence, the elimination of the possibility of occurrence of mousy odour should be an advantage in the reduction of goitre incidence in the countries where pearl millet is a staple food.

Pearl millet contains two trypsin inhibitors with molecular weights around 11,000 Da (Chandrasekhar & Pattabiraman, 1981). These authors reported that the inhibitors are resistant to pepsin and α-chymotrypsin but are partially inactivated by pronase treatment. They also found both inhibitors fairly heat-stable and stable also to exposure to a wide pH range of 1-9.

2.3 MALTING OF CEREALS

With increasing dependence upon cereal grains to provide both the energy and protein requirements of humans in developing countries, the need for raising the overall nutritional status of cereal grains has become increasingly important, and much effort has been made to improve the amount and quality of cereal proteins by using processing techniques such as malting and fermentation. Malting is a technique that can effect positively the physicochemical composition of cereals by improving their nutritional value (reviewed by Chavan & Kadam, 1989).

2.3.1 Malting Technology

The terminologies, viz., sprouting, malting, and germination, are often used interchangeably in the literature to describe the process of soaking or steeping the dry grains in water until they are saturated followed by germination under controlled conditions for a specific period (Briggs, Hough, Stevens & Young, 1981; reviewed by Chavan & Kadam, 1989). Here the technologies used to malt sorghum will be described primarily, because of their relevance to malting pearl millet.

Malting involves essentially the limited germination of cereal grains in moist air under controlled conditions. The main objective of the malting process is to mobilize the grain's endogenous hydrolytic enzymes, particularly the amylases for the breakdown of starch into fermentable sugars (Taylor & Dewar, 2001).

According to Briggs, Hough, Stevens & Young (1981) and Briggs (1998) it is useful to consider malting as consisting of three stages:

1) Steeping or soaking of the grain;
2) Germination, i.e. seedling growth and
3) Drying.

The metabolic processes of germination are initiated during steeping by immersing the grain in water and allowing it to imbibe a suitable amount of water. During the phase of germination, the moist grain is allowed to grow in a humid atmosphere under controlled conditions. When the degradation of the endosperm, which naturally sustains the development of the growing embryo during germination, has progressed to only a limit extent, both the degradation and the growth of the germ is terminated to produce a shelf-stable product, by the process of drying (Briggs, Hough, Stevens & Young, 1981).

The malting process is initiated by steeping. The main objectives of steeping are to hydrate (enzymes need water to be active) the dry, resting grain sufficiently to initiate the metabolic process of germination and to clean the grain by washing, removing the dust and light grains (floaters) (Briggs, Hough, Stevens & Young, 1981; Briggs, 1998). The process of steeping is carried out until the water content of the cereal grain rises to the desired level. The temperature of the water in which the grain is steeped is crucial on the rate of water absorption of the cereal grain (Dewar, Joustra & Taylor, 1993).

In the case of sorghum, in South Africa for instance, commercial malting is generally performed using one of two processes (*viz* Floor Malting and Pneumatic Malting) (Dewar, Joustra & Taylor, 1993; Dewar, 1997; reviewed by Taylor & Dewar, 2001). In both malting processes the steeping stage is common.

The most common types of steeping vessels are called self-emptying and self-cleaning devices and are made from steel and are cylindrical or rectangular in cross-section; with conical bottoms at an angle of at least 45-50 ° from the horizontal which allow the grain to slide out either by gravity or by using pumps (Briggs, Hough, Stevens & Young, 1981; Briggs, 1998; Dewar, Joustra & Taylor, 1993). In most cases with sorghum, temperature control is not available. However, some of steeping vessels are equipped with a means of warming the steep water to a fixed temperature, i.e. the steep water temperature cannot be changed much once established. These vessels can be filled with water from bellow or above and the grain can be loaded by running it down a chute from an overhead silo.

Several factors influence the rate of water uptake during steeping. The so-called steeliness (vitreousness, corneousness, glass)/mealiness (floury) character of the starchy endosperm appears to be of particular importance (Palmer & Harvey, 1977; reviewed by Palmer, 1989). The temperature of the water at steeping is also relevant, since moisture uptake is more rapid at elevated temperatures (Macey, 1977; Lewis & Young, 1995; reviewed by

Briggs, 1998). Besides water, the grain requires a supply of oxygen to support respiration. Oxygen access may be inhibited if the grain is submerged in water for prolonged periods. Air rests, a period during steeping where the grain is not immersed in water, serve the added role of removing carbon dioxide and ethanol, which may inhibit germination. Aeration is also achieved by blowing compressed air through the water during the submerged steep periods.

The visual signs of germination (seedling growth) are the elongation of the radicle and emergence and elongation of the acrospire. The appearance of the white "chit", the coleorhiza or root sheath are the first indication of germination, which may occur at the end of steeping or shortly after casting the grain onto the germination bed (Briggs, Hough, Stevens & Young, 1981; Dewar, Joustra & Taylor, 1993; reviewed by Briggs, 1998).

Although the steeping step is common to both floor and pneumatic malting of sorghum, the germination and drying steps are different. Under outdoor floor malting conditions, the grain is malted outdoors in the traditional way in relatively thin layers on a concrete floor. In this process, the control of the conditions of germination appears to be difficult. Consequently the quality of the malt produced tends to be low and inconsistent (Taylor & Dewar, 1992; reviewed by Dewar, 1997). Sorghum malt of high and consistent quality is required when it is used as an ingredient in industrial brewing. Today, most of the sorghum malt used in factory brewing is malted indoors in modern pneumatic industrial installations. In South Africa, the Saladin box type maltings are generally used to carry out pneumatically sorghum malting (Dewar, Joustra & Taylor, 1993). This system has a rectangular chamber on top of a perforated steel false floor on which the sorghum is germinated, below which is a second chamber or plenum. Fans are used to blow air into the lower chamber and then up through the false floor and subsequently through the bed of germinated grain (Dewar, Joustra & Taylor, 1993; Briggs, 1998).

The relative merits of the utilisation of a floor over a pneumatic maltings in developing countries are the fact that pneumatic maltings are very expensive. Furthermore, the equipment, particularly the fans and turners, require regular and fairly sophisticated maintenance. They require electricity, coal, oil or gas. Contrarily, floor maltings do not require expensive construction, sophisticated maintenance and use solar energy (Taylor & Dewar, 2001). However, pneumatic maltings have a merit over floor malting in producing better quality malt (Taylor & Dewar, 2001). Probably, floor malting is most suitable for pearl millet, particularly in view of the small size of the grain, which is known to fall through the perforated false floor.

After a germination stage sufficient to achieve even modification, the "green malt" is dried to arrest germination and stabilize the malt by lowering moisture levels, typically to less than 10% (Bamforth & Barclay, 1993). During the drying process the growth of the green malt, which was promoted by the biochemical and physical changes, is stopped and the seedling dies by the flow of hot air (Dewar, Joustra & Taylor, 1993; reviewed by Briggs, 1998).

In the process of drying, undesired raw flavours are removed with high drying temperatures (kilning) and pleasant "malty" notes are introduced (Bamforth & Barclay, 1993). The process of kilning is also responsible for the development of malt colour. To ensure survival of enzymes, the drying process must be carefully regulated since the enzymes are crucial in the brewery or distillery to hydrolyse the malt starch into fermentable sugars (Bamforth & Barclay, 1993).

In floor malting, the malt is generally dried by exposing it directly to the sun. The malt is spread into a thin layer and is turned intermittently. Some floor malting plants use mechanical drying to dry the malt. The mechanical drying process involves a flow of warm dry air from a furnace which is passed through the malt. This process is also mostly used in pneumatic maltings. Some maltings use the same germination box to dry the malt, whilst others dry the malt in a separate drying chamber (Dewar, Joustra & Taylor, 1993;

Briggs, 1998). The advantage of the former system is that it "sterlises" the germination box.

As stated, in southern Africa, very little pearl millet malting is carried out at a commercial level. In Mozambique, pearl millet malting is only carried out at the household level as for floor malting. The grains soaked in a clay pot (called *hotso* in Rhonga and Shangaan) and only removed after chitting has occurred. The steeped grains are then germinated by spreading out on a sack bag, which is placed on the floor and covered with an other sack bag. The germinating grains are periodically watered. After 3 to 5 days of germination the cover bag is removed and the green malt is usually placed on the roof, for direct exposure to the sun, to dry. These malts are ground by pounding and mostly used to prepare both opaque beers and soft porridges. When used in the production of traditional beers, they are used with rice grits as the starchy adjunct, which differs from sorghum beer preparation, where maize is used as starchy adjunct.

2.3.2 Malting Science

As described, malting is similar irrespective of the foodstuff for which the malt is intended. The steeping of the grain in water to achieve a moisture level sufficient to activate metabolism in embryonic and aleurone tissues, leading in turn to the development of hydrolytic enzymes initiates the process of malting (reviewed by Bamforth & Barclay, 1993). Moisture uptake into the starchy endosperm is also critical before the food reserves of that tissue can be mobilized through the action of the enzymes. The enzymes migrate through the starchy endosperm, progressing from the embryo end of the kernel to distal end (Bamforth & Barclay, 1993). In barley, the mobilization phase is generally referred to as "modification", the cell walls and protein matrix of the starchy endosperm are degraded, exposing the starch granules and rendering

the grain friable and readily milled (Briggs, Hough, Stevens & Young, 1981; Bamforth & Barclay, 1993; Briggs, 1998).

At the beginning of steeping, the embryo and husk (in the case of barley) absorb water far more rapidly than does the starchy endosperm. It seems that water uptake is regulated by the embryo, although the mechanism by which it does this is still unkown (Briggs, Hough, Stevens & Young, 1981; Bamforth & Barclay, 1993; Briggs, 1998).

To produce homogeneous malt, it is necessary to achieve an even moisture content across the grain bed. Hence, the steeping operation is the most critical stage in the malting process. Steeping conditions must take in to consideration the nature of the grain, i.e. cultivar, grain size, protein content and physiological conditions.

Mealy endosperms, characteristics of good malting grains, have a relatively open structure containing many cracks, and the starch granules are relatively loosely packed in the protein matrix. Water diffuses more readily through such an open structure than it does through a steely endosperm, which has tight protein-starch packing (Bamforth & Barclay, 1993). Thin kernels absorb water more rapidly than do larger ones.

Of all the plant hormones, or plant growth regulators, gibberellins, appear to be the most important in controlling, i.e. stimulating, of germination (Mayer & Poljakoff-Mayber, 1989; Fincher & Stone, 1993).

Gibberellins, produced by germinated barley embryos, induce the synthesis of α-amylase, which leads to the breakdown of the storage reserves of the endosperm and the transport of the substrates of liberation of energy to the metabolising embryo (Palmer, 1989; Fincher & Stone, 1993).

In barley, the germinating embryo produces gibberellins. Because these plant hormones stimulate the aleurone layer to produce and release the enzymes of modification including α-amylase, this is of crucial importance for the process of malting (Lewis & Young, 1995). In fact, this is probably the only desirable function of the embryo. Gibberellic acid can be applied during steeping to bolster the natural hormone and to accelerate modification (Lewis & Young, 1995). The embryonic axis synthesizes the gibberellins possibly from pre-formed precursors including *ent*-kaurene in the scutellum, and are released to stimulate the aleurone layer to synthesize enzymes (Lewis & Young, 1995). This formation and release of gibberellins takes place during steeping, i.e. in the first 2 days of embryo growth and the significant proximal-distal flow of water can carry the hormones with it through the endosperm to the aleurone (Palmer, 1989; Lewis & Young, 1995).

The gibberellins travel through the aleurone from the proximal to distal end in barley. In response to gibberellins the aleurone layer releases the bulk of the enzymes of modification with the scutellum making an early and useful contribution (Lewis & Young, 1995). Once the aleurone layer receives the chemical message delivered from the embryo in the form of the gibberellin hormones it responds with a massive increase of enzyme (protein) synthesis at the expense of the reserve substances (Lewis & Young, 1995; reviewed by Briggs, 1998). Alpha-amylase, endo-β-glucanases and proteases are formed. These enzymes play significant roles in endosperm degradation.

There have been reports that the application of gibberellic acid can promote shoot growth in sorghum (Morgan, Miller & Quinby, 1977; Rood, 1995). However, unlike the situation in barley, there is contradictory evidence as to whether the application of gibberellic acid to germinating sorghum grains increases amylase activity.

For many years authors believed that gibberellic acid does not stimulate amylase activity (Daiber & Novellie, 1968; Aisien & Palmer, 1983; Aisien,

Palmer & Stark, 1993). However, Agu, Okeke, Nwufo, Ude & Onwumelu (1993) and Nzelibe & Nwashike (1995) found that gibberellic acid has a stimulating effect on the diastatic activity of both sorghum and pearl millet. In sorghum, the stimulating effect of gibberellic acid appears to be variety dependent (Nzelibe & Nwashike, 1995).

Currently, sorghum malting practices rely almost entirely on the provision of suitable environmental conditions to initiate germination and promote the development of the essential malt hydrolytic enzymes (Morrall, Boyd, Taylor & Van der Walt, 1986; Dewar, Taylor & Berjak, 1997a).

As stated, in barley malting gibberellic acid can be used to stimulate the modification of the cereal grain during malting. Potassium bromate has been used to reduce the malting loss of pearl millet by reducing the respiratory loss while increasing the modification of the grain (Agu & Okeke, 1991; 1992; Agu & Ezeanalue, 1993).

Endosperm modification in barley and sorghum appears to differ. Nothing is known about pearl millet endosperm modification. In barley, modification is reported to commences at the proximal end of the grain, adjacent to the scutellum and proceeds, roughly parallel to the scutellar epithelium, from the proximal to the distal end of the grain (Gibbons, 1981; Ranki, 1990; reviewed by Fincher & Stone, 1993). It is believed that the major source of hydrolytic enzymes are the enzymes secreted from the scutellum which initially degrade the endosperm adjacent to the scutellum and as germination proceeds, the aleurone layer tissue (Gibbons, 1981). Although there has been great controversy regarding the relative importance of the scutellar epithelial and the aleurone cells in synthesising and secreting the endosperm-degrading enzyme into the endosperm (Palmer, 1989), many researchers accept that the aleurone layer is the tissue that is principally responsible for its synthesis (Ranki, 1990). Biochemical studies indicate that even with some aleurone contamination, the scutellum can account for less than 10 % of the α-amylase

found in the endosperm of barley malt (reviewed by Palmer, 1989). However, the scutellum synthesises, particularly during the early stages of endosperm mobilisation (McFadden, Ahluwalia, Clarke & Fincher, 1988), relatively high levels of another hydrolytic enzyme (*viz* β-(1→3), (1→4)-glucanase) (Stuart, Loi & Fincher, 1986). The latter authors also revealed that the relative contribution of the scutellum and the aleurone to the total hydrolytic activity of enzymes secreted into the starchy endosperm, thus varies according to the particular enzyme and to the time after the initiation of germination.

In sorghum, however, α-amylase is not synthesised in the aleurone layer. The evidence suggests that amylase are synthesised in the scutellum and then diffuse directly to the endosperm (Daiber & Novellie, 1968; Aisien & Palmer, 1983; Aisien, Palmer & Stark, 1983; Glennie, Harris & Liebenberg, 1983; Glennie, 1984). Similar findings, which showed that the degradative enzymes diffuse out from their origin in the scutellum, were found in wheat, rye, oats and maize (Okamoto, Kitano & Akazawa, 1980). Additionally, in sorghum the cell walls, aleurone layer and horny endosperm persist during germination (Glennie, 1984). Although it is known that during pearl millet germination, the α-amylase enzyme preferentially attacks the spherical granules instead of polygonal granules of the grain and that the starch hydrolysis is more vigorous at the centre of the granule than at the periphery (Hoseney, Varriano-Marston & Dendy, 1981); pearl millet grain structure during germination has received very little attention.

In tropical cereals, sorghum and pearl millet, differ from temperate climate cereals (barley) because they have no more than traces of β-amylase. (Novellie & De Schaepdrijver, 1986; Dufour, Mélotte & Srebrnik, 1992; Taylor & Robbins, 1993; reviewed by Zeigler, 1999). Beta-amylase is synthesized during temperate cereal (barley) development but is rendered fully active during germination (MacGregor, Gordon, Meredith & Lacroix, 1972; MacGregor & Lenoir, 1987; reviewed by Palmer, 1989; reviewed by MacGregor, 1996). Beta-amylase plays a crucial role during the mashing

phase of brewing because it is responsible for the degradation of starch and products of α-amylase hydrolysis of starch to maltose, the most abundant fermentable carbohydrate in wort (reviewed by MacGregor, 1996).

2.3.3 Malt Quality

Important quality parameters of malting include: grain germinability, α- and β-amylase activity, free α-amino nitrogen (FAN), malt extract and modification.

A pre-requisite to produce malt of a good and consistent quality is that a high proportion of the grain must germinate (Palmer, 1989; Bamforth & Barclay, 1993). This is measured as Germinative Energy (GE), which is a measure of the percentage of grains which can be expected to germinate if the grain is malted normally at the time of the test (European Brewery Convention, 1987). For sorghum, the recommended GE is 90 % after 72 h of germination (Dewar, Taylor & Joustra, 1995).

The single most important indicator of malt quality for opaque beer brewing is Diastatic Power (DP). DP is a measure of the joint α- and β-amylase activity of the malt. At the biochemical level, the combined action of α- and β-amylases, which develop during malting (Daiber & Novellie, 1968; reviewed by Palmer, 1989; Dufour, Mélotte & Srebrnik, 1992), is responsible for the breakdown of starch to fermentable sugars during the process of malting. Alpha-amylase attacks α- (1→4) glucosidic bonds within starch molecules to produce dextrins (short chains of glucose molecules) and a variety of sugars including maltotriose and maltose, and glucose. Being an endoenzyme, α-amylase rapidly solubilises starch to yield the smaller fragments, which is useful during the mashing stage in brewing, since it reduces the viscosity of the starch solution (reviewed by Palmer, 1989; Bamforth & Barclay, 1993; Lewis & Young, 1995). While β-amylase, an exoenzyme, releases maltose by hydrolysing the penultimate α- (1→4) glucosidic bond from the non-reducing

ends of the dextrins produced by the action of α-amylase (reviewed by Palmer, 1989; Bamforth & Barclay, 1993; Lewis & Young, 1995).

Free α-amino nitrogen (FAN) content, which consists of free amino acids and small peptides, produced by proteinase and peptidase activity in the malt, is an important component of malt quality, as it is required during the fermentation stage of the brewing process as a source of yeast nutrition (Baxter, 1981; Pickerell, 1986). Adequate FAN levels are especially important in lager beer brewing processes which use non-malted grain (sorghum or maize) with only a small amount of sorghum malt (Muts, 1991).

Malt hot water extract, which is a measure of the soluble solids in solution and gives an estimate of how much of the malt will solubilise during the brewing process (Briggs, Hough, Stevens & Young, 1981; Bamforth & Barclay, 1993; Briggs, 1998), is particularly important in lager beer brewing where, in some cases, an all-malt grist is used rather than an approximately 30 % malt grist which is generally the situation in opaque beer brewing (Palmer, 1989).

Modification, which is the term that signifies all the desirable changes that occur when grain is converted into malt (Briggs, 1998), is the measure of all the other malt quality parameters together. There are three aspects of modification, (1) accumulation of hydrolytic enzymes; (2) the variety of chemical changes that occur in the grains; and (3) the physical changes, which appear as a weakening and softening of the grains (Briggs, 1998).

2.3.4 Sorghum and Millet Malting

In barley, steeping is generally acknowledged as the most critical stage of the malting process (Briggs, Hough, Stevens & Young, 1981; reviewed by Briggs, 1998; reviewed by Taylor & Dewar, 2001). For many years, this malting stage

has been considered to be relatively unimportant for sorghum, finger and pearl millet. This perhaps because in sorghum and finger and pearl millet malting it is necessary to water the grain during the germination step, whereas in barley malting the grain must receive all the water it requires for germination during the steeping stage (Novellie, 1962a; reviewed by Taylor & Dewar, 2001). According to a review by Taylor & Dewar (2001), under controlled conditions, sorghum is steeped from 4-6 h to a maximum of 24 h. The optimum steeping time for finger and pearl millet, reported in the literature, is relatively short, i.e. between 6 and 16 h (Nout & Davies, 1982; Malleshi & Desikachar, 1986b, 1986c; Gomez, Obilana, Martin, Madzvamuse & Monyo, 1997; Muoria & Bechtel, 1998). The short steeping time used for finger and pearl millet may have to do with the smaller size of these grains compared to sorghum since smaller kernels have a proportionally larger surface area than larger ones.

Studies by Dewar, Taylor & Berjak (1997a) showed that as is the case for barley, the steeping stage is critical stage of the malting process for sorghum and that the conditions of steep should be controlled in order to optimise the quality of the resulting malt. The latter authors also found that the quality of malt, i.e. Diastatic Power (DP), free amino nitrogen (FAN) and hot water extract are significantly affected by steeping time and temperature. In addition, aeration during steeping has been found to further improve the quality of the malt produced in sorghum (Ezeogu & Okolo, 1995; Okolo & Ezeogu, 1995; Dewar, Taylor & Berjak, 1997a), in finger millet (Nout & Davies, 1982; Malleshi & Desikachar, 1986c) and in pearl millet (Gomez, Obilana, Martin, Madzvamuse & Monyo, 1997; Muoria & Bechtel, 1998). No studies have been reported to investigate the effect of steeping time and temperature on pearl millet malt quality.

Studies by many authors established the optimum conditions for the germination stage of sorghum malting (Novellie, 1962a; Morrall, Boyd, Taylor & Van der Walt, 1986; Dewar, Taylor & Berjak, 1997b; reviewed by Taylor & Dewar, 2001). In terms of both steeping (Dewar, Taylor & Berjak, 1997a) and

germination (Novellie, 1962a; Morrall, Boyd, Taylor & Van der Walt, 1986; Dewar, Taylor & Berjak, 1997b), the optimum temperature for sorghum, is between 24-30 °C. A temperature of 18 °C (and possible lower), reported as optimal for barley malting (Briggs, Hough, Stevens & Young, 1981; reviewed by Briggs, 1998), is considered suboptimal for sorghum (Dewar, Taylor & Berjak, 1997a), as are temperatures of 32 °C and higher (Morrall, Boyd, Taylor & Van der Walt, 1986). In the case of both finger and pearl millet, very little research has been done. Workers reported different optimum germination conditions for subsequent malt quality, 20-25 °C for 5-6 days (Nout & Davies, 1982, working with finger millet); 15-20 °C for 4-5 days (Malleshi & Desikachar, 1986b, working with finger millet); 22 °C for 3 days (Muoria & Bechtel, 1998, working with pearl millet) and 25 °C for 3-5 days (Gomez, Obilana, Martin, Madzvamuse & Monyo, 1997, working with pearl millet).

The moisture content of sorghum, both at the end of steeping (Dewar, Taylor & Berjak, 1997a) and of the green malt (malt prior to the drying process) (Dewar, Taylor & Berjak, 1997b) are positively correlated with malt quality in terms of DP, FAN and hot water extract. Therefore, for brewing purposes the moisture content of sorghum during the malting process is an important indicator of malt quality (reviewed by Taylor & Dewar, 2001).

Non-germinated sorghum grain shows virtually no β-amylase activity (Taylor & Robbins, 1993). Beta-amylase is influenced by germination time and temperature. A rapid increase in β-amylase activity occurs within the first 2 days of germination (Pal, Wagle & Sheorain, 1976; Taylor & Robbins, 1993) and subsequently declines in rate of increase up to 7 days (Taylor & Robbins, 1993). It is reported that β-amylase activity is inversely related to germination temperature over the 24-32 °C range, the highest activity being at 24 °C and 5 days (Taylor & Robbins, 1993).

Sorghum has little α-amylase in the non-germinated grains. Germination of sorghum leads to the production of both α- and β-amylases with α-amylase predominating. Morrall, Boyd, Taylor & Van der Walt (1986) and Dewar, Taylor & Berjak (1997b) found that the optimum conditions for high amylase activity in sorghum are germination at 24-30 °C for at least 4 days.

The proteolytic activity of pearl millet malt in 3- day germinated grains was found to be nearly eight times that of non-germinated grains (Pal, Wagle & Sheorain, 1976). Morrall, Boyd, Taylor & Van der Walt (1986) and Dewar, Taylor & Berjak (1997b) found that malting sorghum at 24-30 °C, for 6 days, at high watering treatment, gave optimum FAN. An increase in proteolytic activity during malting is desirable for nutritional improvement of cereals because it leads to hydrolysis of prolamins, and the liberated amino acids such as glutamic acid and proline are converted to limiting amino acid such as lysine (reviewed by Chavan & Kadam, 1989). FAN development is reported to vary among cultivars probably because of differences in major enzyme characteristics and rate of protein metabolism during sorghum malting as well as variations in grain protein structure and degradability (reviewed by Owuama, 1999). The reason for the increase of FAN during the malting process could be the fact that nitrogen is transferred from endosperm to embryos (axes and scutella). Nitrogen may also move from root to embryo by physiological mechanisms (Agu & Palmer, 1996).

During the germination phase, the moist grain is allowed to grow in a humid atmosphere under controlled conditions. When the degradation of the endosperm, which naturally sustains the development of the growing embryo (germ) during germination, has progressed to only a limited extent, the malster terminates both its degradation and the growth of the germ to produce a shelf-stable product, by drying the grain (Briggs, Hough, Stevens & Young, 1981; Taylor & Dewar, 1992; reviewed by Taylor & Dewar, 2001). In barley, the malt is kilned (rather than only dried), in that it is not only dehydrated, but partly cooked. This procedure partially or wholly destroys some of the

hydrolytic enzymes developed during malting. It also develops colour and flavour in the final product (Briggs, Hough, Stevens & Young, 1981). In tropical cereals like sorghum and pearl millet, however, because the hydrolytic activity of the malt (particularly the β-amylase activity) is inherently lower than that of barley (Novellie, 1960; Jayatissa, Pathirana & Sivayogasunderam, 1980; Aniche & Palmer, 1990), the malt is dried at a relatively low temperature (50 °C) as opposed to being kilned, as higher drying temperatures significantly reduce the already-low amylase activity (Novellie, 1962a; Okon & Uwaifo, 1985). The main aim of sorghum, finger and pearl millet malt is to conserve as much of the enzyme activity of the malt as possible whilst producing a shelf-stable product.

Alkaline steeping with final warm water steep in general improves substantially α-amylase activity in sorghum. However, in some cultivars it reduces it. The reason for this variation with cultivars is unclear but may be related to α-amylase polymorphism (reviewed by Owuama, 1999). Alkali is known to disrupt the molecular structure of the non-starch polysaccharides, which make up the cell wall (Verbruggen, Beldman & Voragen, 1995). It was suggested that NaOH disrupts the sorghum pericarp cell wall structure and, consequently allows water to enter the grain more rapidly during steeping, but not at a rate causing any significant imbibitional damage. Enhanced imbibitional hydration of the grain, brought about by steeping in dilute NaOH, could facilitate the onset of the stage of active metabolic activity more rapidly, thereby producing the malt quality required more quickly (Dewar, Taylor & Orovan, 1997; reviewed by Taylor & Dewar, 2001). Alkaline steeping causes a highly significant increase in sorghum malt FAN (Okolo & Ezeogu, 1996). No research has been reported on the effect of alkali treatment on the improvement of pearl millet malt quality parameters, probably because most varieties of pearl millet are low in tannins.

Exogenous GA_3, however, is not used in the sorghum malt industry, largely because of the evidence that its application does not significantly improve the amylase activity of this grain (Daiber & Novellie, 1968; Aisien & Palmer, 1983; Aisien, Palmer & Stark, 1983).

Earlier, Nout & Davies (1982) found that two levels of bromate (15 and 150 ppm) caused a reduction of approximately 30% malting loss in barley, however, the response in finger millet and sorghum was not significant. They also found that gibberellic acid treatment (0.2 ppm) was characterised by an accelerated development of α-amylase activity, resulting in increased diastatic power values.

Agu & Okeke (1991, 1992) and Agu & Ezeanalue (1993) studied the effect of potassium bromate and gibberellic acid on malting of pearl millet. They found that both additives, singly or in combination, improved the quality of pearl millet malt compared to untreated control. Potassium bromate was the most effective treatment followed by gibberellic acid and the combined treatment. Agu, Okeke, Nwufo, Ude & Onwumelu (1993) compared the effect of these additives on both sorghum and pearl millet. For millet the highest diastase and cellulase activities were observed on the 5^{th} day of germination (0.20 mg/l gibberellic acid applied at steep-out), while sorghum showed highest activities of the enzymes on 4^{th} day of germination for the same concentration of gibberellic acid.

Nzelibe & Nwasike (1995) reported that in two varieties of pearl millet, 12 ppm potassium bromate treatment did not significantly reduce the malting losses. However, at 120 ppm potassium bromate showed a significant reduction in malting losses in millet and sorghum at the 6^{th} day of malting. These authors also reported that gibberellic acid stimulated the production and activity of diastatic enzymes in pearl millet and sorghum. The effect of gibberellic acid was apparent from the 2^{nd} day of malting, corresponding to the mobilisation of natural gibberellins in the grain.

Germination significantly increases the total soluble sugars, reducing and non-reducing sugar contents of finger and pearl millet with a parallel decrease in its starch content, Malleshi & Desikachar (1986c) and Sripriya, Antony & Chandra (1997), working with finger millet and Opoku, Ohenhen & Ejiofor (1981) and Khetarpaul & Chauhan (1990a), working with pearl millet. Both soaking and sprouting periods have been found to influence the loss of starch and accumulation of sugars in sorghum. The soaking of grains for 10 h followed by sprouting for 24 h improved the starch digestibility significantly. However, prolonged soaking and germination beyond 10 and 24 h, respectively, apparently caused adverse effects on the susceptibility of residual starch to α-amylase (reviewed by Chavan & Kadam, 1989).

Malleshi & Desikachar (1986a) considered an increase in dietary fibre content, during malting of finger, foxtail and pearl millet for 4 days at 25 °C, as apparent and due to mainly the disappearance of starch. There is no indication that fibre components like cellulose and lignin are synthesized from carbohydrates during sprouting.

Malting has been found to decrease the fat content in sorghum germinated for 3 days at 25 and 30 °C by Aucamp, Grieff, Novellie, Papendick, Schwartz & Steer (1961) and Bhise, Chavan & Kadam (1988), respectively; in pearl millet germinated for 3 days at the temperatures of 22, 25 and 30 °C by Opoku, Ohenhen & Ejifor (1981); Opoku, Osagie & Ekperigin (1983); Mtebe, Nabikunze, Bangu & Mwanezi (1993) and Pawar & Pawar (1997) and in finger millet by Malleshi & Desikachar (1986a). The lipids of pearl millet grains are implicated in the decreased of its palatability. Hence, any reduction in fat content during malting will be advantageous, since it may contribute to an increase in pearl millet consumption.

Contradictory data have been reported on the effect of germination on the mineral (ash) content of cereals. When malting at 30 °C for 96 h, Wu & Wall (1980) observed a decrease in the initial 3 days of germination ash (mineral)

content of wheat, oats, and sorghum followed by an increase. However, no such differences or definite trends were observed for millets (Malleshi & Desikachar, 1986c) and sorghum (Aucamp, Grieff, Novellie, Papendick, Schwartz & Steer, 1961; Wu & Wall, 1980) during malting. An increase in ash content upon malting found by these authors is probably due to the loss of starch, while a decrease can be attributed to leaching losses during soaking and rinsing.

Sankara Rao & Deosthale (1983) reported that there were significant mineral losses when pearl millet is malted for 96 h at 25 °C: iron and manganese 40%, copper, 30%, and phosphorus, 25% and when finger millet is malted: calcium, 40%, zinc, 30% and copper, 25%.

Most reports agree that malting of cereal grains generally improves their vitamin value. However, the quantitative increase in each vitamin may be small and its practical significance in meeting the nutritional requirements of cereal-based diets is difficult to evaluate (reviewed by Chavan & Kadam, 1989). An increase in the content of vitamins of B-group through simple processing like sprouting is nutritionally desirable since cereal grains are an important source of these vitamins (reviewed by Chavan & Kadam, 1989). The sprouts obtained from maize, sorghum, finger millet and pearl millet have been reported to contain higher levels of niacin and riboflavin than the respective non-germinated grain (Aucamp, Grieff, Novellie, Papendick, Schwartz & Steer, 1961; reviewed by Chavan & Kadam, 1989). During malting of pearl millet for 96 h at 22 °C in the dark (Opoku, Ohenhen & Ejiofor, 1981) observed significantly higher values for riboflavin, thiamin, ascorbic acid, carotene, and tocopherol in the malt than in grains.

Data on changes in total protein content in cereal grains due to malting are contradictory. Some reports (Dalby & Tsai, 1976; Opoku, Ohenhen & Ejiofor, 1981); Taylor, 1983; Sripriya, Antony & Chandra, 1997) have indicated an increase while others (Hwang & Bushuk, 1973; Wu & Wall; 1980; Bhise, Chavan & Kadam, 1988; Subramaniam, Sambasiva, Rao, Jambunathan,

Murty & Reddy, 1995) have shown a decrease in protein content upon malting of cereals. Figure 1 gives data of changes in the protein content in several cereals grains during 5 days of malting at 28 °C in the dark (Dalby & Tsai, 1976). The total protein content increased steadily with time of malting in all cereals except the oats. Similarly, Hamad & Fields (1979), working with wheat; Wu & Wall (1980) and Taylor (1983), working with sorghum; Opoku, Ohenhen & Ejiofor (1981) and Sripriya, Antony & Chandra (1997), working with pearl and finger millet, respectively, reported an increase in protein content during malting. The increase in protein content has been attributed to loss in dry weight, particularly carbohydrates through respiration during germination.

Contradictory data were reported by Hwang & Bushuk (1973), working with wheat, Bhise, Chavan & Kadam (1988), Subramaniam, Sambasiva, Rao, Jambunathan, Murty & Reddy (1995), working with sorghum, and Opoku, Ohenhen & Ejiofor (1981), working with pearl millet, when studying loss in protein of the malts. The reduction in protein content is attributed to prolonged soaking of grains, where the protein may have been leached out.

The time of soaking the grains prior to germination and sprouting will greatly affect the changes in the protein fractions (Lorenz, 1980). Prolamin changes during malting for 5 days of some cereal grains are shown in Figure 2.

Taylor (1983) showed that the prolamins show the greatest decrease during malting of sorghum at 28 °C for 7 days, initially accounting for 45% of the total nitrogen in the grain, they decline to a mere 16% of their original quality. However, this author found that electrophoretic prolamin bands remained unchanged during malting which was the indication that prolamins are degraded directly to small peptides and amino acids. The glutelin fraction which accounted for some 27% of the total nitrogen in the non-malted sorghum declined to 57% of its original quantity after malting.

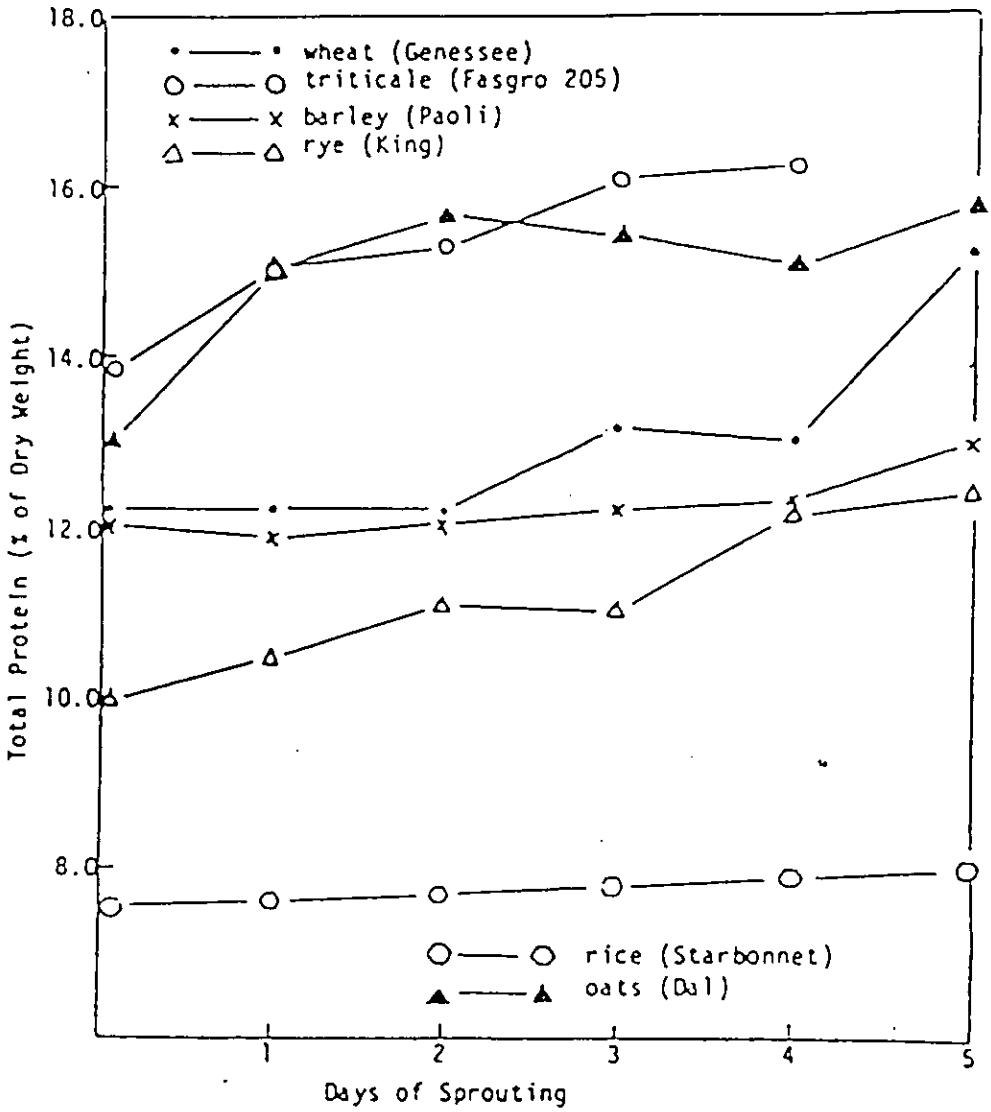


Figure 1- Changes in total protein during germination of cereal grains (Dalby & Tsai, 1976)

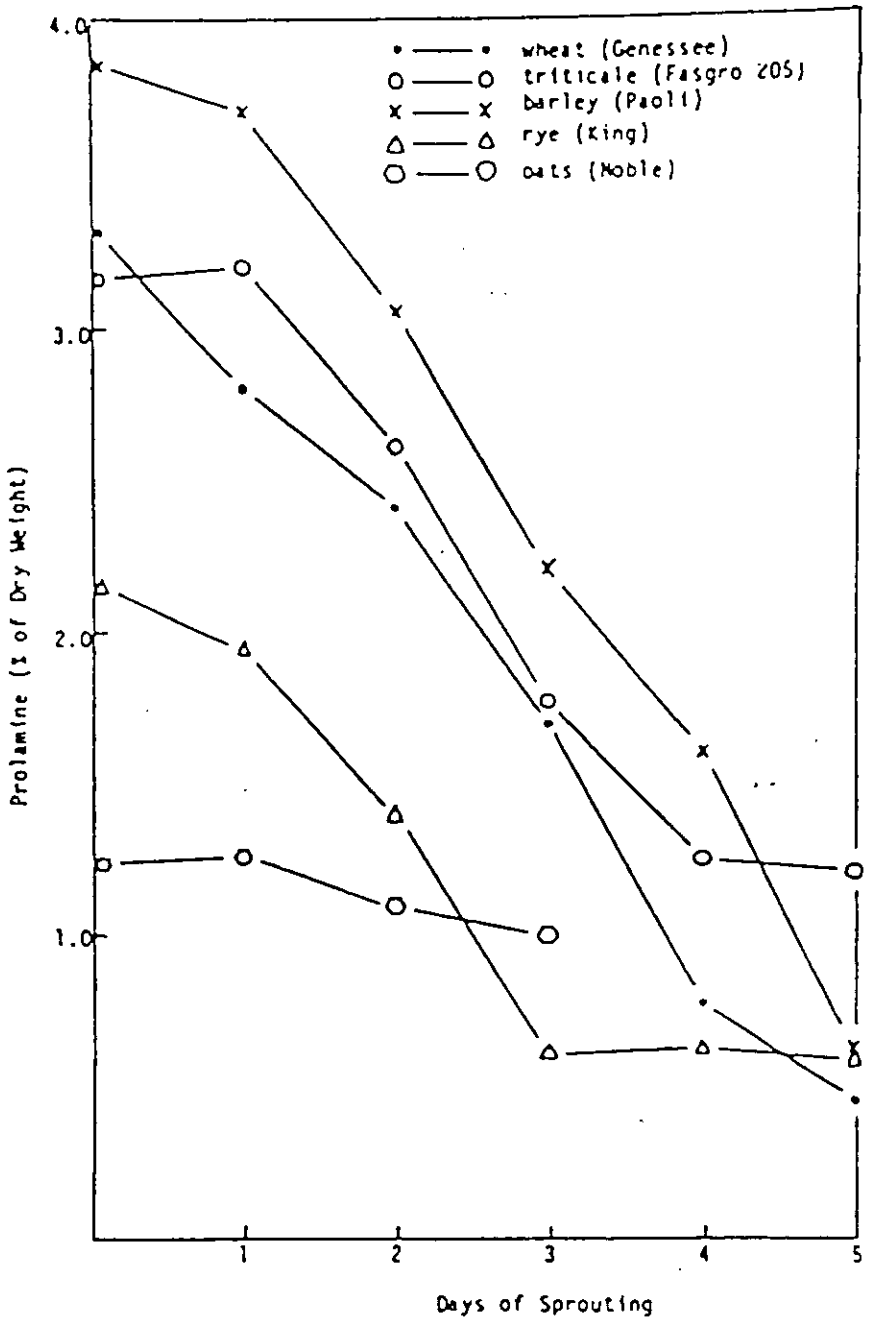


Figure 2- Changes in prolamin during germination of cereal grains (Dalby & Tsai, 1976)

Germination and sprouting increase the amounts of essential amino acids in cereal grains and that is of nutritional importance (Lorenz, 1980). Dalby & Tsai (1976) reported increases in lysine and tryptophan when germinating the cereal grains for 5 days at 28 °C in the dark (Figures 3 and 4, respectively). These increases were found to be inversely related to the amount of prolamin present in the cereal grains studied. Taylor (1983) reported a lysine increase of 4-fold during malting of sorghum and stated that the improvement in the nutritional quality of free amino nitrogen mirrors the changes that occur in the total amino acid composition of sorghum during the germination. The increase in lysine was also reported by Almeida-Dominguez, Serna-Saldivar, Gomez-Machado & Rooney (1993). They reported an increase in lysine from 2.2 to 3.2 and 3.0 to 7.8 g/100 g of protein when normal and high lysine-sorghum were germinated. Germination of the finger, pearl and foxtail millets for 48 h enhanced the lysine content of their proteins from 3.5% to 4.0, 3.7% to 4.3% and 3.0% to 3.5%, respectively; however, the threonine and the sulphur amino acid content were not altered appreciable (Malleshi & Desikachar, 1986a). Udayasekhara Rao (1994) did not find differences in the amino acid compositions of brown and white finger millet proteins after malting.

Malting at 25 °C for at least 48 h has been reported to reduce the phytic acid content thereby improving the nutritional quality of finger millet (Hulse, Laing & Pearson, 1980; Kumar & Kapoor, 1984; Shukla, Gupta, Swarkar, Tomar & Sharma, 1986) and pearl millet (Opoku, Ohenhen & Ejiofor, 1981; Kumar & Kapoor, 1984). Calcium and phosphorus contents were also lower in the germinated samples. This could be due to metabolic/leaching losses and also transfer of nutrients to the growing embryo (Malleshi & Desikachar, 1986a).

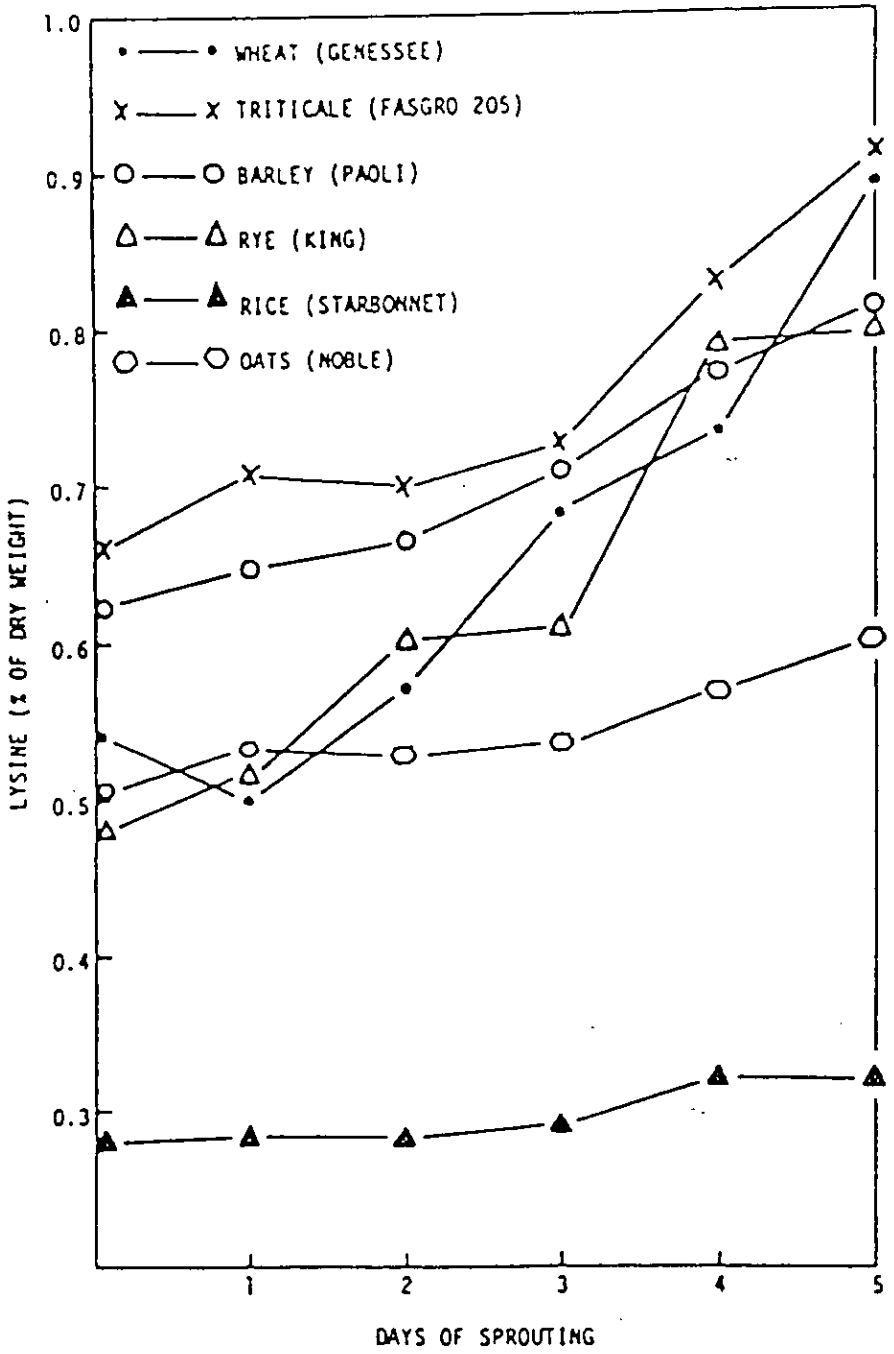


Figure 3- Changes in lysine content of cereal grains during sprouting (Dalby & Tsai, 1976)

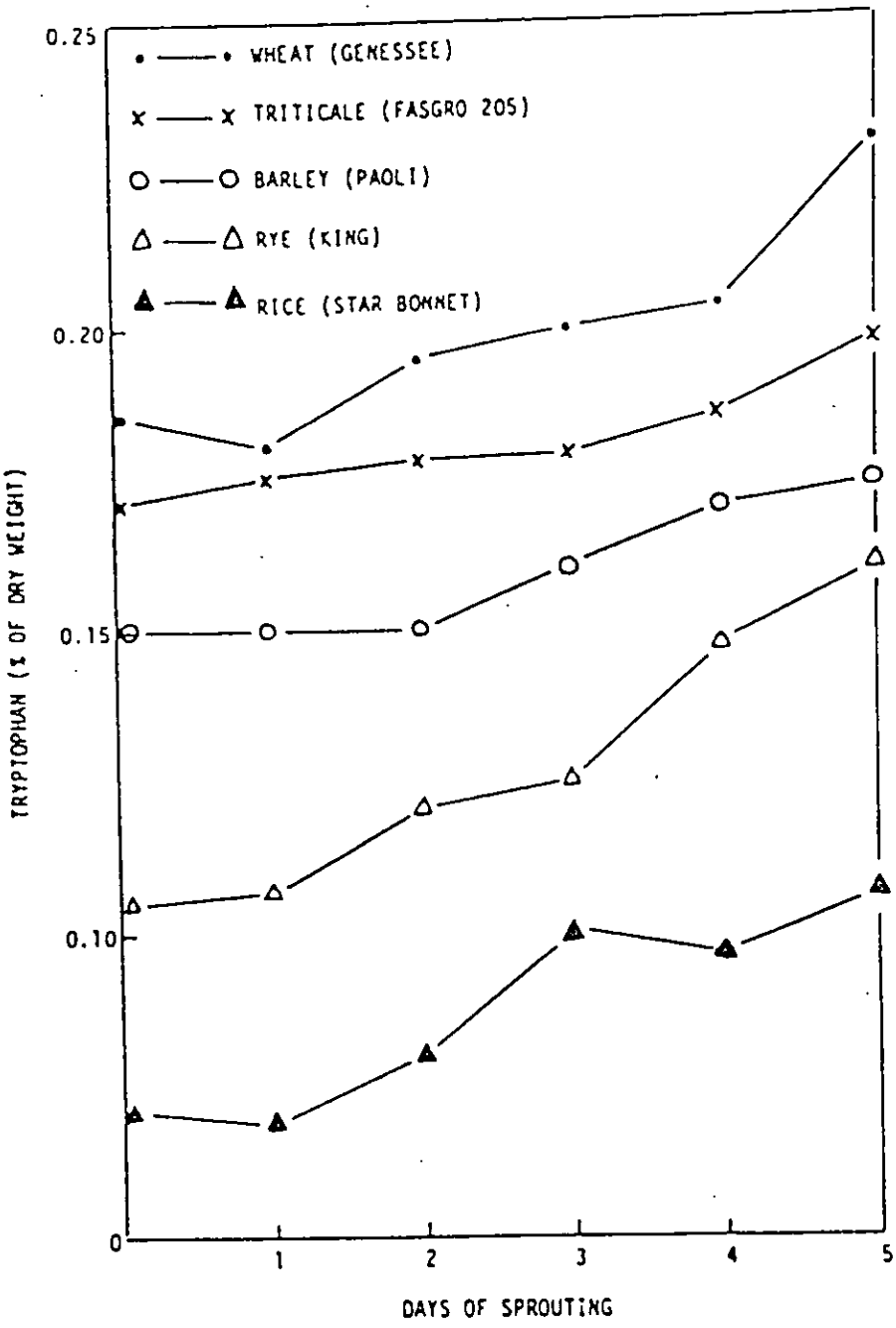


Figure 4- Changes in tryptophan content of cereal grains during sprouting (Dalby & Tsai, 1976)

Germination of finger millet, pearl and foxtail millets for 96 h at 25 °C reduced the phytate phosphorus levels (Malleshi & Desikachar, 1986b). Germination of pearl millet at 30 °C for 48 h significantly increases inorganic P, non-phytate P and phosphorus extractable in 0.03 M HCl, the concentration of acid found in human stomach, with a corresponding decline in phytate P of pearl millet grains germination (Khetarpaul & Chauhan, 1989). HCl-extractability of calcium, ion, zinc, copper and manganese, an index of their bioavalability to humans, was also significantly improved by germination (Khetarpaul & Chauhan, 1989).

McGrath, Kaluza, Daiber, Van der Riet & Glennie (1982) reported that during sorghum malting, for 5 days at 25 °C, the roots and shoots developed a large complement of polyphenols and the properties of the tannins changed. Glennie (1984) observed that tannins in bird-resistant (high tannin) sorghum formed complexes with proteins during malting at 25 °C for 6 days, although the tannins did not affect the percentage of germination. These reports indicate that sprouting does not decrease the tannin content of grain, but favours the formation of complexes between testa tannins and endosperm proteins.

2.4 SUMMARY

Most research has been done on the effect of malting on temperate cereal grains. Sorghum is the only tropical cereal grain, that malting has been investigated to any great extent for industrial utilisation.

Although some research has been done on malting conditions of millets, temperature and time of both steeping and germination, most of it concentrates on the effects of germination/sprouting on chemical composition, nutritional properties, enzymatic activities and the effects of additives on quality of malts of millets. Little research is reported on the semi- and

industrial malting of pearl millet, especially that which would suit southern African food industries. Also, there has been little research on the effect of malting on the pearl millet grain structure modification; as well as the effect of malting on the off-odour which appears in pearl millet when it is ground. It was therefore decided to devise this project to accommodate these aspects. For better understanding of the pearl millet malting process it was also thought necessary to give emphasis to the functionality of its starch, since starch is the most important functional food biopolymer in cereal grains.

CHAPTER 3

EXPERIMENTAL

3.1 EXPERIMENTAL DESIGN

The effects of malting temperature (steeping and germination) and time and moisture (germination) conditions were investigated using two pearl millet varieties. Various quality parameters were measured, including: those for opaque and conventional beer brewing, and physico-chemical, nutritional and functional parameters. The assays were replicated three times.

The flow chart of the experimental design is shown in Figure 5.

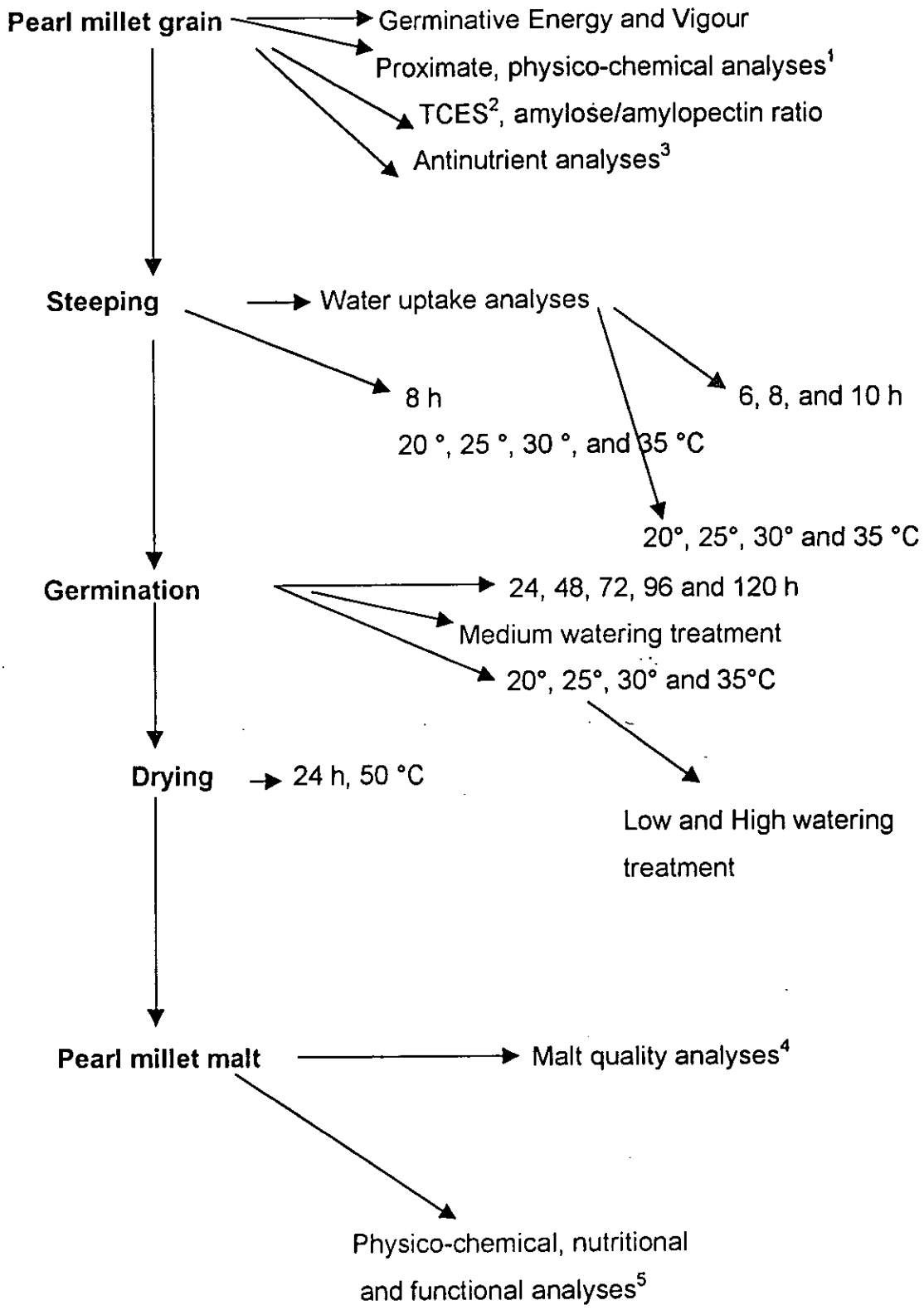


Figure 5. - Flow chart of the experimental design of pearl millet malting

Notes:

[1]Proximate and physico-chemical analyses: Moisture content, protein, crude fat, ash, total carbohydrate, the percentage of TCES, odour generation and evaluation of the odour generated.

[2]TCES: percentage of the total carbohydrate which was enzyme susceptible.

[3]Antinutrient analyses: Phytic acid and polyphenols.

[4]Malt quality analyses: Green malt moisture, roots and shoots, Diastatic Power (DP), α- and β-amylase activity, free α-amino nitrogen (FAN), malting loss, hot water extract (HWE).

[5]Physico-chemical, nutritional and functional analyses: protein, crude fat, ash, total carbohydrate, the percentage of the TCES, pasting properties, Water Absorption Index (WAI), Water Solubility Index (WSI), soluble nitrogen, Nitrogen Solubility Index (NSI), *in vitro* protein digestibility, amino acids, grain structure modification, odour generation and evaluation of the odour generated.

3.2 MATERIALS

The raw materials used were:

(1) Pearl millet grain, variety SDMV 910018, planted at Estação Agrária de Chockwé in Gaza Province, Mozambique, in November 1996 and harvested in April 1997.

(2) Pearl millet grain, variety SDMV 89004, (released in Zimbabwe as PMV-2), kindly donated by SADC/ICRISAT, Matopos Research Station, Bulawayo, Zimbabwe, planted at Matopos Research Station, in November 1997 and harvested in April 1998.

(3) Sorghum malt, NK 283, a condensed tannin-free hybrid from South Africa, previously malted in laboratory conditions, was used as a standard.

(4) Barley malt, variety Arapiles from Australia, kindly donated by S.A. Malsters (Pty) Ltd. (Alrode, South Africa) was used as a standard.

The raw materials were kept under cold storage (≤ 10 °C) conditions until used.

Pearl millet grains of variety SDMV 89004 are shown in Figure 6.

Figure 6.- Pearl millet grains of variety SDMV 89004 (average mass of each kernel: 8.9 mg)

3.3 PEARL MILLET MALTING PROCESS

The malting of the two varieties of pearl millet was done at CSIR – Bio/Chemtek (Division of Food, Biological and Chemical Technologies), Pretoria, South Africa. The malting process comprised three main stages: Steeping, Germination and Drying.

3.3.1 Steeping

Samples of pearl millet grain (5 kg) were washed 4 to 5 times, in running tap water (22-24 °C) to remove foreign material. The grain was then put in 250 x 300 mm (2.5 kg) nylon bags and closed with rubber bands and spin-dried (30 s at 300 x g) to remove excess surface-held water. After the spin-drying process, exactly 500 g of grain per sample was placed in 150 x 200 mm nylon bags and closed with rubber bands. The bag held grain was re-weighed then steeped in static water at 20°, 25°, 30° or 35 °C, with a cycle of 2h wet, 2 h dry air rest for 8 h. During the dry stands the grain was held in still air at 20–22 °C. After the steeping period, the grain in nylon bags was spin-dried (30 s at 300 x g) and weighed.

3.3.2 Germination

The steeped grain was then germinated in the nylon bags for 1 to 5 d at one of four different temperatures: 20°, 25°, 30° and 35 °C. Germination was carried out in a water-jacketed incubator (Forma Scientific, Marrietta, Ohio, USA) in an atmosphere of near water-saturation with a continuous flow of moist air. The nylon malting bags were covered with wet cloths to maintain the water saturation. Twice daily, the bag held grain samples were removed from the incubator, weighed and steeped for 10 min in tap water (22–24 °C), then spin-dried (30 s at 300 x g), weighed and returned to the germination cabinet (Medium watering treatment).

At the germination temperature of 25 °C, beside this Medium watering regime, two additional regimes were used:

1) Low- a very small amount of water was applied to maintain the green malt at constant fresh weight.

2) High- as for Medium watering regime, but the spin-drying process was omitted; so as to keep the malt wet.

Figure 7 shows the green malt moisture content of the two pearl millet varieties.

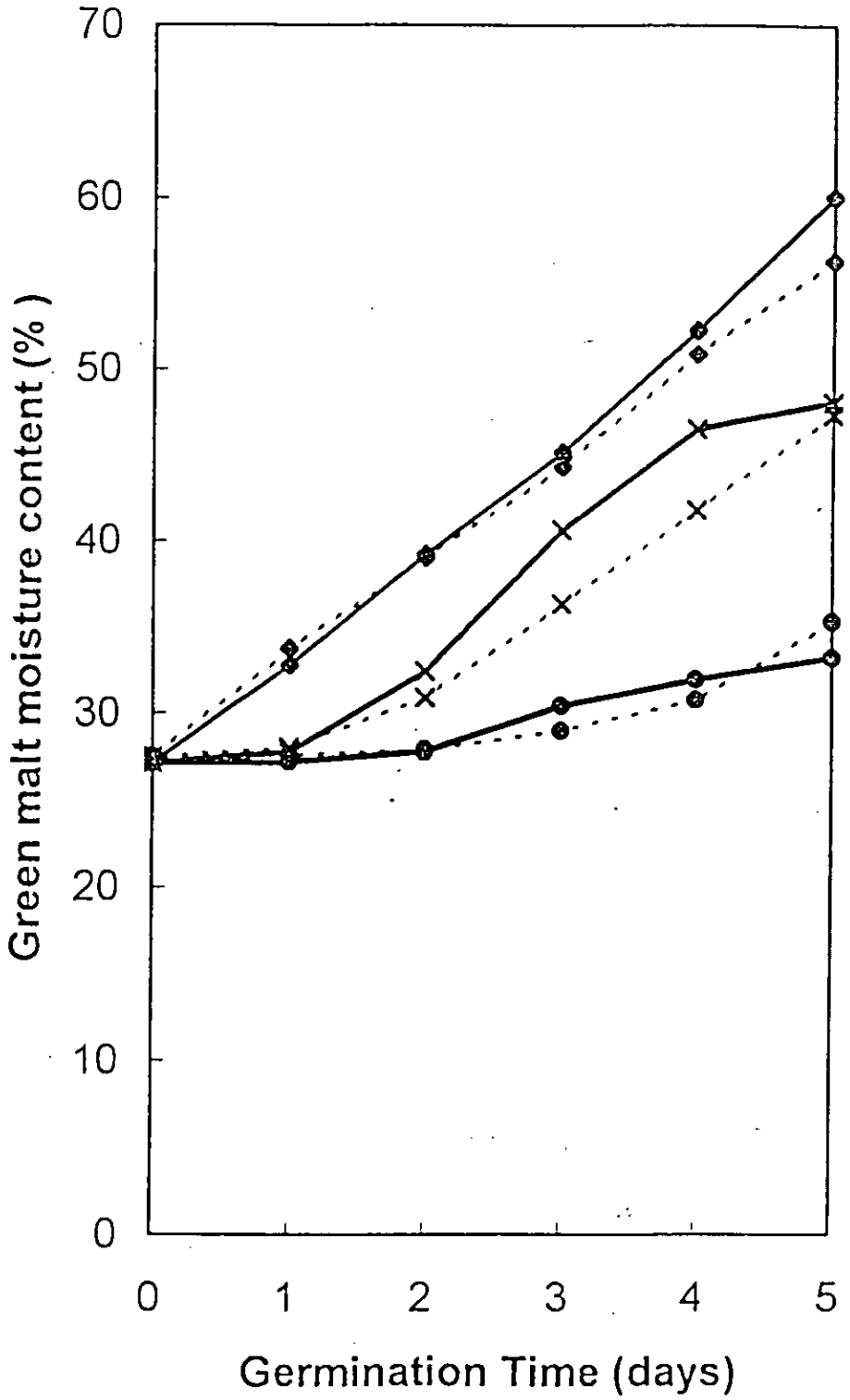


Figure 7.- Green malt moisture content at low, medium and high watering treatment of pearl millet at 25 °C (variety SDMV 89004(–) and variety SDMV 91018 (--) at various watering treatments (•- Low watering; x- Medium watering; ♦- High watering)

3.3.3 Drying

After the pre-determined malting times, two samples of the green malt were taken, weighed and placed in shallow stainless steel trays with fine mesh bottoms and dried in a forced draught oven (50 °C for 24 h). After the drying, the dry malt was allowed to cool for about and weighed, put in sealed plastic bags and stored at 4 °C.

3.4 ANALYTICAL METHODS

For analysis, the pearl millet malts, including roots and shoots, were milled for 45 s in a beater-type, water-cooled coffee mill (Janke and Kunkel, Staufen, Germany).

3.4.1 Moisture content

Moisture content of the raw materials, as well as of the malts was determined by AOAC Method 14.004 (Association of Official Analytical Chemists, 1980). In this method the difference between the initial and final weights should be equal to the weight of free moisture in the sample.

3.4.2 Protein

Protein content (N x 6.25) was determined by the Kjeldahl method, as modified in AOAC Method 2.057 (Association of Official Analytical Chemists, 1980). Samples are digested with concentrated sulphuric acid in the presence of Kjeldahl tablet (potassium sulphate, selenium and copper) catalyst for the conversion of nitrogen to ammonium hydrogen sulphate. Then the digested samples are neutralised with concentrated NaOH and volatile ammonium was distilled off into a solution of boric acid. An amount of borate anions equivalent to the ammonium is formed which is then titrated against standard hydrochloric acid. The distillation and titration steps were performed using an automated Büchi 322 Distillation Unit (Büchi, Flawil, Switzerland).

3.4.3 Fat

Fat content was determined by AOAC Method 14.018, procedure 7.056 (Association of Official Analytical Chemists, 1980). This method is based on the principle of gravimetric extraction of fat from a sample by petroleum solvent, followed by recovery of the fat by evaporation of the solvent.

3.4.4 Ash

Ash was determined by AOAC Method 14.006 (Association of Official Analytical Chemists, 1980) by differential weighing after incinerating at 550 °C until a light grey ash was obtained to constant weight.

3.4.5 Total carbohydrate and the total carbohydrate which was enzyme-susceptible

To determine total carbohydrate and the percentage of the TCES an enzymic method using α-amylase and amyloglucosidase was used (Taylor, 1992). This method measures total starch by gelatinization of the starch by pressure-cooking followed by hydrolysis to glucose by α-amylase and amyloglucosidase and colorimetric determination of glucose. The percentage of the total carbohydrate which was enzyme susceptible was measured in the same way except that the gelatinization step was omitted.

3.4.6 Fibre

Fibre was calculated by difference:

100 - (% moisture + % carbohydrate + % fat + % ash + % protein)

3.4.7 Phytic acid

The determination of phytic acid was done by a modification of the method of Garcia-Villanova, Garcia-Villanova & Ruiz de Lope (1982). The method involves reaction of phytic acid with iron (III) solution. Excess iron (III) is complexed with sulphosalicylic acid and determined by complexometric back

tritation with standard disodium ethylenedinitrilotetraacetate (EDTA) solution. The time allowed to stand after adding HCl and Na_2SO_4 was 2 h instead of 90 min.

3.4.8 Total polyphenols

Before the determination of the total polyphenol content, the pearl millet grains were submitted to the Chlorox bleach test. In this test, the grain is immersed in a sodium hypochlorite solution (bleach) containing alkali. The solution at 70 °C dissolves away the out pericarp layer of grain, revealing the presence of a black pigmented testa layer in the case of tannin grains, or its absence in the case of non-tannin grains (Waniska, Hugo & Rooney, 1992).

To determine the total polyphenol content of the samples, the modified method of the International Standardisation Organisation (ISO), (1988) was used. In this method the Jerumanis ferric ammonium citrate reacts with the phenolic compounds under alkaline conditions and the absorbance of the products is linearly related to the concentration of the phenolic acid compound in the samples.

3.4.9 Amylose/amylopectin ratio

The amylose/amylopectin ratio in the native starches was measured colorimetrically by a method of Knutson (1986) as modified by Faulks & Bailey (1990). This method is based on the principle of the reaction of iodine when it is dissolved in mixtures of water and dimethyl sulphoxide where triicodide ion

is formed. This reaction is utilised to form the blue amylose-iodine complex. Amylose is then determined by measurement of the absorbance of the complex at 620 nm.

3.4.10 Germinative Energy (GE) and Germinative Vigour (GV)

Germinative Energy and Germinative Vigour were measured as described by Dewar, Joustra & Taylor (1993). In order to produce good quality malt, a prerequesite is that a high (>90%) proportion of the grain is germinable. Germinative Energy (GE) is a measure of the percentage of grains, which can be expected to germinate if the grain is malted normally at the time of the test. Germinative Vigour (GV) is a measure of how energetically the grain germinates.

3.4.11 Malting loss

The total malting loss was calculated according to the method described by Gomez, Obilana, Martin, Madzvamuse & Monyo (1997):

Malting loss (%)=[(Initial dry grain weight- Dry malt weight)/(Initial dry grain weight)] x 100

3.4.12 Roots and shoots

Where stated, these were separated from the pearl millet malt kernels by rubbing the grain in a nylon bag of coarse mesh size, which allowed the roots and shoots to escape while retaining the kernels, as described by Morrall, Boyd, Taylor & Van der Walt (1986). The weight of roots and shoots was expressed as a percentage of the total malt weight.

3.4.13 Green malt moisture

The green malt moisture content was calculated as follows, Gomez, Obilana, Martin, Madzvamuse & Monyo (1997):

Green malt moisture (%) = [(Green malt weight – Dry malt weight)/Green malt weight] x 100

3.4.14 Water uptake

Water uptake of the pearl millet grains was calculated as a percentage of the weight gained by the pre-washed, non-steeped grain at the end of the 8 h of steeping.

3.4.15 Grain hardness

Pearl millet grain hardness was estimated according to Rooney & Miller (1982). Pearl millet grains were cut into halves longitudinally. One half was viewed with the naked eye and the proportion of horny endosperm was determined by reference to a standard. On the basis of the proportion of horny endosperm, grains were classified into hard, intermediate and soft. The hardness was rated in a scale of 1 to 5, with a 1 rating meaning that the kernel contains very little floury endosperm (almost completely horny) and a 5 rating meaning essentially all floury.

3.4.16 Diastatic Power (DP)

Diastatic Power is a measure of the joint α- and β-amylase activity. The standard method for sorghum malt Diastatic Power (South African Bureau of Standards (SABS), 1970) was used. In this Diastatic Power determination, an aqueous extract of milled malt is prepared which contains amylase enzymes. The enzyme extract is reacted with an excess of starch under specific conditions of time, temperature and pH. The amount of sugar product produced by hydrolysis of starch is determined by titration in terms of its reducing power.

The following amendments to the SABS method were included:

The peptone extraction was replaced by distilled water extraction since the pearl millet malt used was prepared from tannin-free grains.

Two g whole malt flour was used instead of 25 g and the extraction volume

was 40 ml instead of 500 ml.

The extraction was carried out in centrifuge tubes and the extraction time was reduced from 2.5 to 2 h.

The results were expressed as Pearl Millet Diastatic Units (PMDU/g dry wt malt), where PMDU is equivalent to Kaffircorn Diastatic Units (KDU).

3.4.17 Alpha-amylase activity

Determined according to the method of Novellie (1962a) by inactivating the β-amylase activity at 70 °C in the presence of Ca^{2+} ions. The malt extracts were prepared as for DP. Calcium acetate (0.02 g) was added to the 10.0 ml of the malt extract. The extracts were incubated at 70 °C for 15 min. After incubation, extracts were cooled in ice for 10–15 min at 30 °C. Enzyme activity was then determined by the DP procedure and expressed as PMDU.

3.4.18 Beta-amylase activity (by inactivation of α-amylase)

The contribution of β-amylase to the diastatic activity was estimated by inactivating α-amylase activity with 0.2 M ammonium oxalate which binds with calcium ions in the α-amylase molecule (Taylor & Von Benecke, 1990). Pearl millet extracts were prepared with 1.00 g malt and 40.0 ml 0.2 M ammonium oxalate. Enzyme activity was then determined by the DP procedure and expressed as PMDU.

3.4.19 Beta-amylase activity (Betamyl method)

The Betamyl Method (beta-amylase assay kit, from Megazyme, Ireland) was used. This method measures soluble and total β-amylase activity using a mixture of p-nitrophenyl maltopentaoside and p-nitro maltohexaoside as substrate (McCleary & Codd, 1989). Cysteine is used to extract the "insoluble" β-amylase. This method is based on hydrolysis of p-nitrophenyl maltopentaoside to maltose and p-nitrophenyl maltotrioside by β-amylase, the nitrophenyl trioside is immediately cleaved to glucose and free p-nitropehenyl by the α-glucosidase present in the substrate mixture. Thus, the rate of the release of p-nitrophenol relates directly to the rate of release of maltose by β-amylase (Mathewson & Seabourn, 1983). Beta-amylase activity was expressed as Betamyl Units.

3.4.20 Free α-amino nitrogen (FAN)

Free amino nitrogen consists of free amino acids and small peptides produced by proteinase and peptidase activity in the malt. The FAN content was determined according to the ninhydrin method described by Morrall, Boyd, Taylor & Van der Walt (1986), except that 1 g of the sample was used instead of 5 g. This method measures the pre-formed FAN malt. An aqueous extract of milled malt is heated with ninhydrin and the colour produced measured colorimetrically. Ninhydrin is an oxidising agent and causes oxidative decarboxylation of α-amino acids, producing CO_2, NH_3 and an aldehyde with one less carbon atom than the parent amino acid. The reduced ninhydrin then reacts with the unreduced ninhydrin and the liberated ammonia, forming a purple complex.

The intensity of purple colour is determined colorimetrically and the amount of FAN calculated by the use of a standardised solution of an amino acid. The results were expressed as mg/ 100 g dry weight malt.

3.4.21 Hot water extract (HWE)

Hot water extract is a measure of the soluble solids in solution and gives an estimate of how much of the malt will solubilise during the brewing process (Briggs, Hough, Stevens & Young, 1981). Hot water extract gives an indication of the modification of the malt during the malting process. Hot water extract was measured as described by Morrall, Boyd, Taylor & Van der Walt (1986). In this method the amount of malt solubilised is measured by determining the specific gravity of clear wort obtained by centrifugation under standard conditions.

3.4.22 Water Absorption Index (WAI)

Water Absorption Index was determined according to the method of Anderson, Conway, Pfeifer & Griffin (1969). This method is based on the determination of the amount of pearl millet starch dispersed in an excess of water (supernatant) after centrifugation.

3.4.23 Water Solubility Index (WSI)

Water Solubility Index was measured according to the method of Anderson, Conway, Pfeifer & Griffin (1969). In this method the amount of milled pearl millet solubilised in water is measured by determining the amount of dry matter recovered after the supernatant is evaporated from Water Absorption determination (3.4.22).

3.4.24 Percentage of soluble nitrogen and Nitrogen Solubility Index (NSI)

Nitrogen Solubility Index is a measure of the amount of water soluble nitrogen in a sample. The amount of the total nitrogen and water soluble nitrogen in non- and germinated pearl millet was determined according to AACC Method 46-23 (American Association of Cereal Chemists, 1983). This method is based on the determination of the amount of nitrogen in water extract from pearl millet samples after centrifugation.

3.4.25 *In vitro* protein digestibility

The *in vitro* protein digestibility method of Mertz, Hassen, Cairns-Whittern, Kirlies, Tu & Axtell (1984) as modified by Hamaker, Kirleis, Butler, Axtell & Mertz (1987) was used. The principle of this method is that pepsin is used to hydrolyse the insoluble protein into soluble amino acids and peptides. The

percentage protein hydrolysis is related to the protein digestibility. Protein was determined by the Kjeldahl method (3.4.2).

The percentage of protein digestibility was calculated as:

$$\%\ Protein\ Digestibility = (Total\ protein - Residual\ protein)/Total\ protein \times 100$$

3.4.26 Amino acid analysis

The amino acid content of non- and malted pearl millet was analysed using the Pico.Tag Method (Bidlingmeyer, Cohen & Tarvin, 1984) in a Pico.Tag Column for hydrolysate amino acid analysis Part no. 88131 (3.9 mm x 15 cm). This method is based upon formation of a phenylthiocarbamyl derivative of the amino acids for analysis of free amino acids from acid-hydrolysed proteins. The amino acids are separated based on their hydrophobicity. Hydrochloric acid is used to hydrolyse proteins into amino acids. This derivatization method is very sensitive and specific for primary and secondary amino acids in protein hydrolyzates. The reversed-phase liquid column chromatographic system allows for the rapid, bonded-phase separation with ultraviolet detection of the common amino acids with 12-min analysis time and a 1-pmol sensitivity (Bidlingmeyer, Cohen & Tarvin, 1984).

3.4.27 Scanning electron microscopy (SEM)

Samples for SEM were germinated for 1 to 7 days as described in Germinative Energy and Germinative Vigour methods (3.4.10). Grains of non- and germinated pearl millet were fixed in 2.5% gluteraldehyde in 0.075 M phosphate buffer; pH 7.4-7.6 for 1 h. Samples were rinsed 3 times, 15 min each in 0.075 M phosphate buffer. After this, samples were dehydrated in 50%, 70% and 90% ethanol for 15 min each concentration, then dehydrated 3 times with 100% ethanol for 15 min each, respectively. After fixing the samples were fractured by cutting longitudinally using a sharp blade.

The samples were subjected to critical point drying in liquid CO_2. The critical-point-dried samples were mounted on stubs and sputtered coated with gold. The samples were examined in a JEOL JSM-U3 microscope (JEOL, Tokyo, Japan).

3.4.28 Odour generation and evaluation of the odour generated

Non- and germinated milled pearl millet were mixed with water 30% (w/v) and placed in Petri dishes. The samples were then air-dried in a fume cupboard at ambient temperature overnight. About 3 g samples were sealed in 27 ml glass bottles "polytops" for at least 1 h before being evaluated for odour generation. Twelve trained panellists carried out evaluations of treated pearl millet samples. These panellists were trained using fresh samples (no odour) and treated, i.e. wetted and dried grits (odour present). The trained panellists were asked to sniff the head space over treated samples and compare it with a control samples. Panellists were asked to rate each coded sample on numeric

scale of 1 (least intense, i.e. no odour) to 9 (most intense, i.e. strong odour present). Appendix A shows the score sheet used to evaluate the odour generated by pearl millet.

3.4.29 Pasting properties

The pasting behaviour of the grain and germinated pearl millet was determined using a Rapid Visco-Analyser model 3D (RVA) (Newport Scientific, Warriewood, Australia). Four g whole flour of grain and germinated of pearl millet (14% moisture basis) were separately mixed with 25 ml of distilled water. A programmed heating and cooling cycle was used. The suspension was held at 50 °C for 1 min, heated to 90 °C for 7.5 min at the rate of 6 °C/min, held at 90 °C for 5 min before cooling to 50 °C in 7.5 min and holding at 50 °C for 1 min. Peak viscosity (PV), hot paste viscosity (shear thinning) and setback were recorded.

3.4.30 Statistical analysis

Analysis of variance (ANOVA) with the least significant difference test (LSD-Test) was applied. The level of 95% was considered as significantly different.

CHAPTER 4

RESULTS

4.1 ANALYSIS OF RAW MATERIALS

4.1.1 Proximate Analysis of Pearl Millet Varieties

The proximate composition of the two pearl millet varieties used in this research work is shown in Table 6.

TABLE 6.- Proximate composition (%) of the two pearl millet varieties used in this investigation

Pearl Millet Variety	Moisture	Protein (N x 6.25)	Fat	Ash	Carbohydrate	Fibre (by difference)
SDMV 89004	9.4	10.6 (11.7)[a]	5.8 (6.4)[a]	1.3 (1.4)[a]	71.0 (78.4)[a]	1.9 (2.1)[a]
SDMV 91018	12.0	9.9 (11.3)[b]	5.6 (6.4)[a]	2.6 (2.9)[b]	66.3 (75.3)[b]	3.6 (4.1)[b]

Mean values with different letters in each column are significantly different from each other ($p < 0.001$).

Results in brackets are on dry basis.

Results are mean of three replicates.

Variety SDMV 89004 had significantly higher protein and carbohydrate contents than SDMV 91018. Variety SDMV 91018 had significantly higher ash and fibre contents. Fat was similar in both varieties.

4.1.2 Enzyme Susceptibility of Carbohydrate, Amylose-Amylopectin Ratio and Gelatinisation Temperature Range of Pearl Millet Varieties

The percentage of the TCES, the amylose-amylopectin ratio as well as the gelatinisation temperature range of pearl millet varieties are shown in Table 7.

TABLE 7.- Enzyme susceptibility of carbohydrate, amylose-amylopectin ratio and gelatinisation temperature range of pearl millet varieties used in this investigation

Pearl Millet Variety	% of Total Carbohydrate Enzyme-Susceptible	Amylose-Amylopectin Ratio (%)	Gelatinisation Temperature Range (°C)
SDMV 89004	10.6[a]	19.0/81.0[a]	63.5 - 70.9[a]
SDMV 91018	10.1[a]	20.4/79.6[a]	65.8 - 73.4[b]

Mean values with different letters in each column are significantly different from each other ($p < 0.001$).

Results are mean of three replicates.

There was no significant difference between the percentage of the TCES between the two pearl millet varieties. Variety did not have any effect on the amylose-amylopectin ratio. SDMV 91018 had a significantly higher gelatinisation temperature than SDMV 89004.

4.1.3 Antinutritional Factors in Pearl Millet Varieties

The percentages of phytic acid and total polyphenols are shown in Table 8.

TABLE 8.- Phytic acid and polyphenol content of the two pearl millet varieties used in this investigation

Pearl Millet Variety	Phytic Acid (%)	Total Polyphenols (%)
SDMV 89004	0.22 (0.24)[a]	0.08 (0.09)[a]
SDMV 91018	0.24 (0.27)[b]	0.10 (0.11)[a]

Mean values with different letters in each column are significantly different from each other ($p < 0.001$).

Results in brackets are on dry basis.

Results are mean of three replicates.

The percentage of phytic acid of the variety SDMV 89004 was significantly lower than SDMV 91018.

After submiting the two millet varieties to the Chlorox bleach test no black kernels were observed, i.e. none of the grains were of the high tannin type.

There was no significant difference between the content of total polyphenols in the two pearl millet varieties. The level of polyphenols in both pearl millet varieties was low.

4.1.4 Endosperm Texture of Pearl Millet Varieties

After cutting the grains in two lengthwise halves using a sharp disposable scalpel, the texture of the endosperm of the pearl millet of variety SDMV 89004 was found to be softer compared with the SDMV 91018. Varieties SDMV 89004 and SDMV 91018 had an endosperm textures of 2.5 and 3.2, respectively.

4.1.5 Germinative Energy (GE) and Germinative Vigour (GV) of Pearl Millet Varieties

The percentage Germinative Energy and Germinative Vigour is shown in Table 9.

TABLE 9.- Germinative Energy and Germinative Vigour of the two pearl millet varieties used in this investigation

Pearl Millet Variety	Germinative Energy (%)			Germinative Vigour (%)		
	24h	48h	72h	24h	48h	72h
SDMV 89004	94.0[a]	97.9[a]	99.6[a]	92.0[a]	96.2[a]	98.0[a]
SDMV 91018	89.9[b]	93.1[b]	95.5[b]	86.9[b]	91.0[b]	92.5[b]

Mean values with different letters in each column are significantly different from each other ($p < 0.001$).

Results are mean of three replicates.

Both pearl millet varieties had high Germinative Vigour and high Germinative Energy. Variety SDMV 89004 had significantly higher Germinative Energy and Germinative Vigour than SDMV 91018.

4.1.6 Water Uptake of Pearl Millet Varieties

The effects of steeping time and temperature as well as variety on water uptake of the two pearl millet varieties are shown in Table 10.

Water uptake was significantly affected ($p < 0.001$) by time and temperature of steeping. In general, variety did not have any effect on water uptake ($p > 0.05$). Water uptake increased with steeping time for both pearl millet varieties. Water uptake increased with an increase in steeping temperature for both pearl millet varieties. The lowest percentage of water uptake 14.7% was observed at 6 h steeping time at 20 °C with SDMV 91018. The highest water uptake 25.3% was recorded at 10 h steeping time at 35 °C with variety SDMV 89004.

TABLE 10.- Water uptake of the two pearl millet varieties at various steeping temperatures and times

Steeping Temperature (°C)	Steeping Time (h)	Water Uptake (%) Variety SDMV 89004	SDMV 91018
20	6	15.9[a]	14.7[a]
	8	18.9[b]	16.1[b]
	10	19.0[b]	16.2[b]
25	6	17.0[a]	16.7[a]
	8	19.9[b]	19.5[b]
	10	19.7[b]	19.6[b]
30	6	19.1[a]	18.8[a]
	8	21.7[b]	21.2[b]
	10	22.3[c]	21.3[b]
35	6	22.2[a]	21.4[a]
	8	24.8[b]	21.8[b]
	10	25.3[c]	23.2[c]

Means values with different letters in each block are significantly different from each other ($p < 0.001$).

Results are mean of three replicates.

4.2 PEARL MILLET ENDOSPERM MODIFICATION DURING GERMINATION

Figure 8 shows pearl millet malts of variety SDMV 89004, germinated for 5 days, with long roots and shoots.

The pearl millet grains of both varieties were tear-shaped and the germ was large in proportion to the rest of the kernel. The kernel contained floury and horny endosperm portions (Figure 9).

In the non-germinated grains, the floury opaque endosperm contained spherical (round) or polygonal starch granules and many air spaces (Figure 10). There appeared to be no protein bodies in the opaque endosperm. The horny endosperm was devoid of air spaces and contained polygonal starch granules (Figure 11).

Initially (24 h), modification of starch and the protein matrix could be observed in the floury endosperm adjacent to the scutellum epithelium (Figure 12). At the beginning of germination, degradation appeared as pin holes in the starch granules and as germination progressed these holes became bigger and appeared to coalesce (Figure 13). As germination progressed, degradation was observed at further distances from the germ in the direction of the horny endosperm. The proportion of highly degraded starch granules decreased from the proximal to the distal end of the grain. There was not much modification on the cell walls, aleurone layer and horny endosperm during germination (Figure 9). Protein body degradation occurred to a lesser extent than starch granule degradation (Figure 14).

The pearl millet grains, which were germinated for longer periods, showed a seedling with long roots and shoots and almost completely empty starchy endosperm. In this research, both pearl millet varieties showed up signs of germination (chitting) between 8 and 10 h of germination.

Figure 8.- Pearl millet malts of variety SDMV 89004 germinated for 5 days (average mass of each kernel: 8.9 mg; average length of the roots and shoots: 15-20 mm)

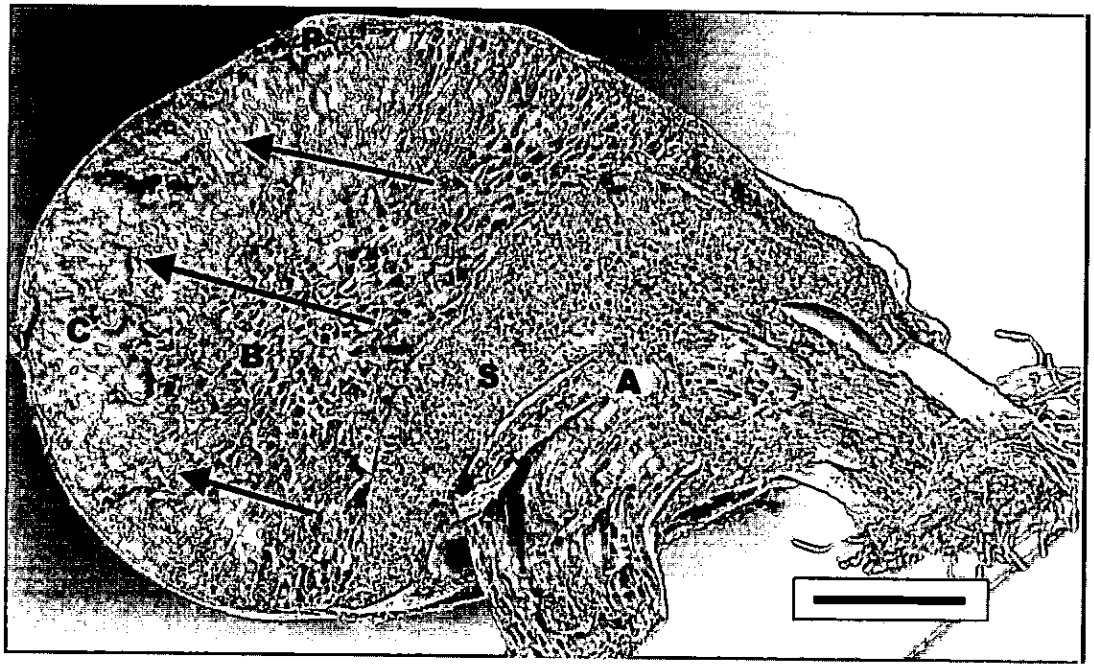


Figure 9.- Longitudinal section of pearl millet malt of variety SDMV 91018 germinated for 5 days showing the large germ in proportion to the rest of the kernel and the wave of modification (arrows) Germ; B- Floury endosperm; C- Horny endosperm; P- Pericarp; S- scutellum) (Bar = 600 μm)

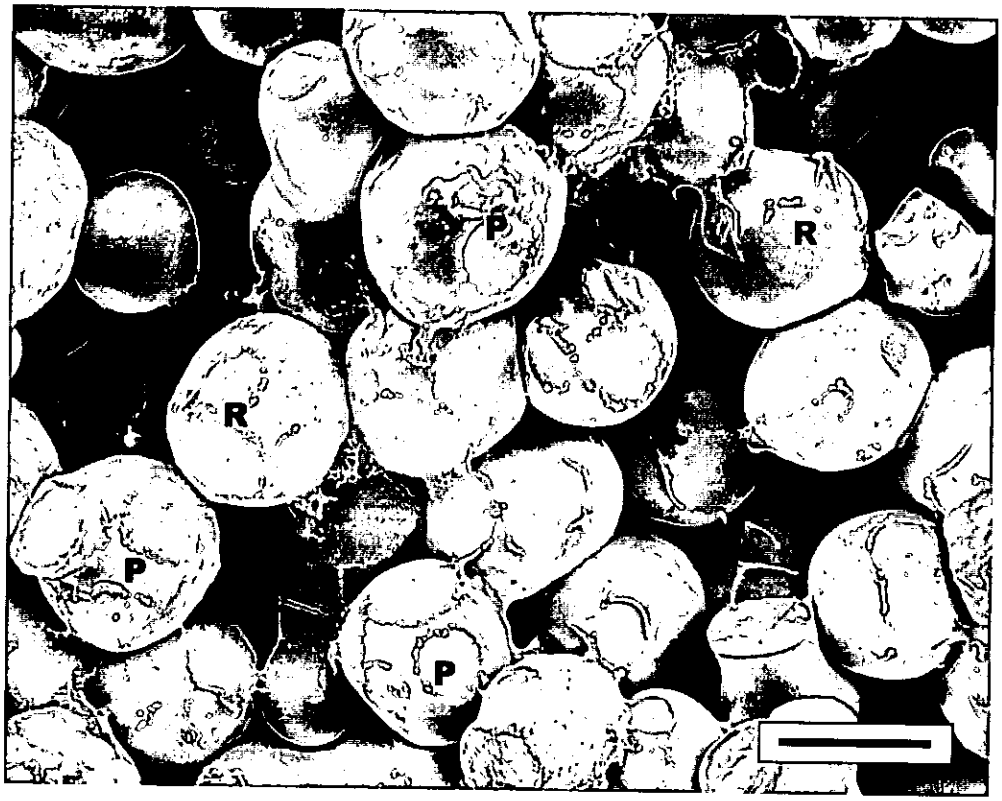


Figure 10.- Floury endosperm of non-germinated pearl millet of SDMV 89004 variety showing both rounded (R) and polygonal (P) starch granules (Bar = 8 μm)

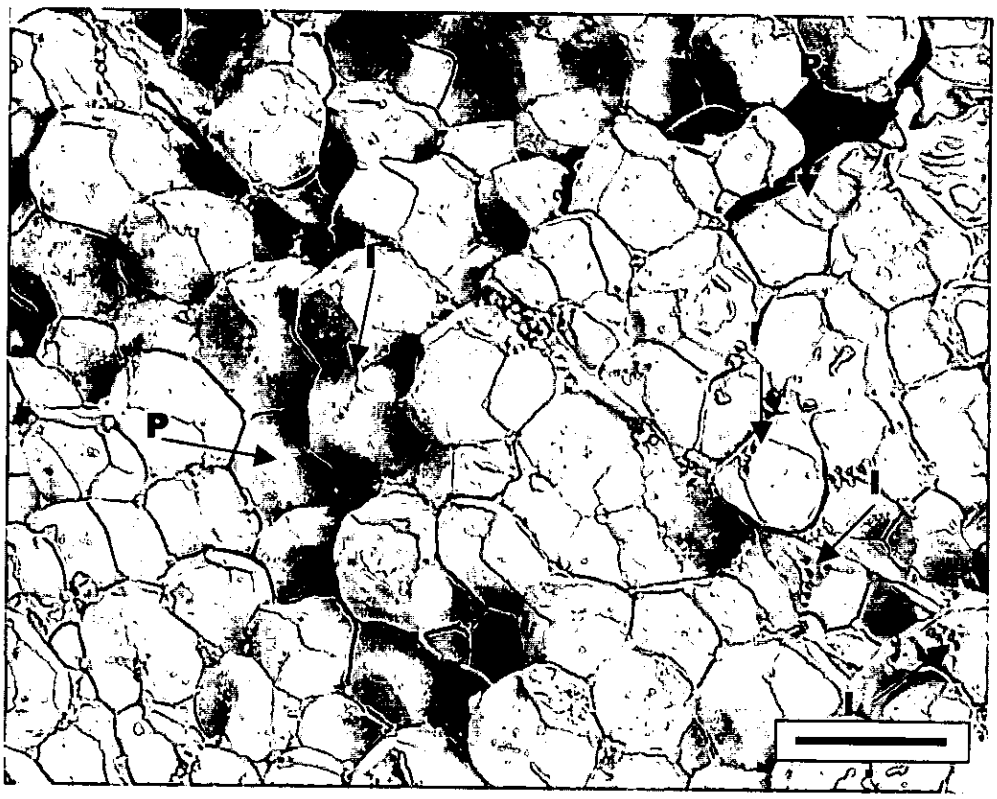


Figure 11.- Horny endosperm of non-germinated pearl millet grain of SDMV 91018 variety showing polygonal (P) starch granules and imprints (I) of protein bodies (Bar = 9 μm)

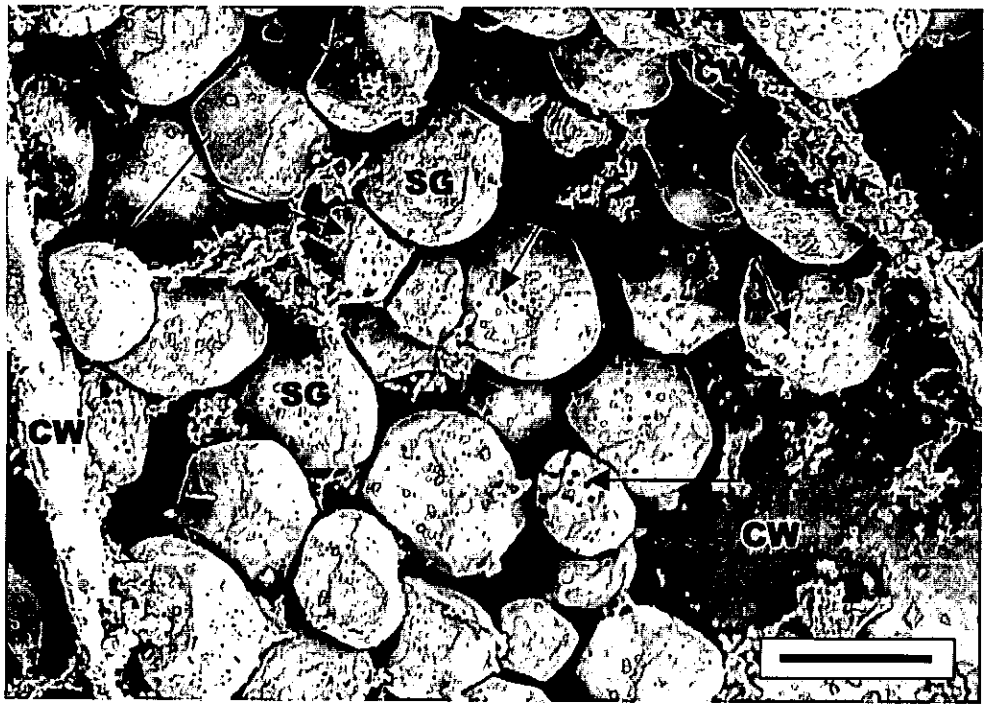


Figure 12.- Floury endosperm adjacent to scutellum epithelium of pearl millet of variety SDMV 89004 after 24 h of germination showing pin holes (arrows) in the starch granules (CW- Cell wall; SG- Starch ganule) (Bar = 8 μm)

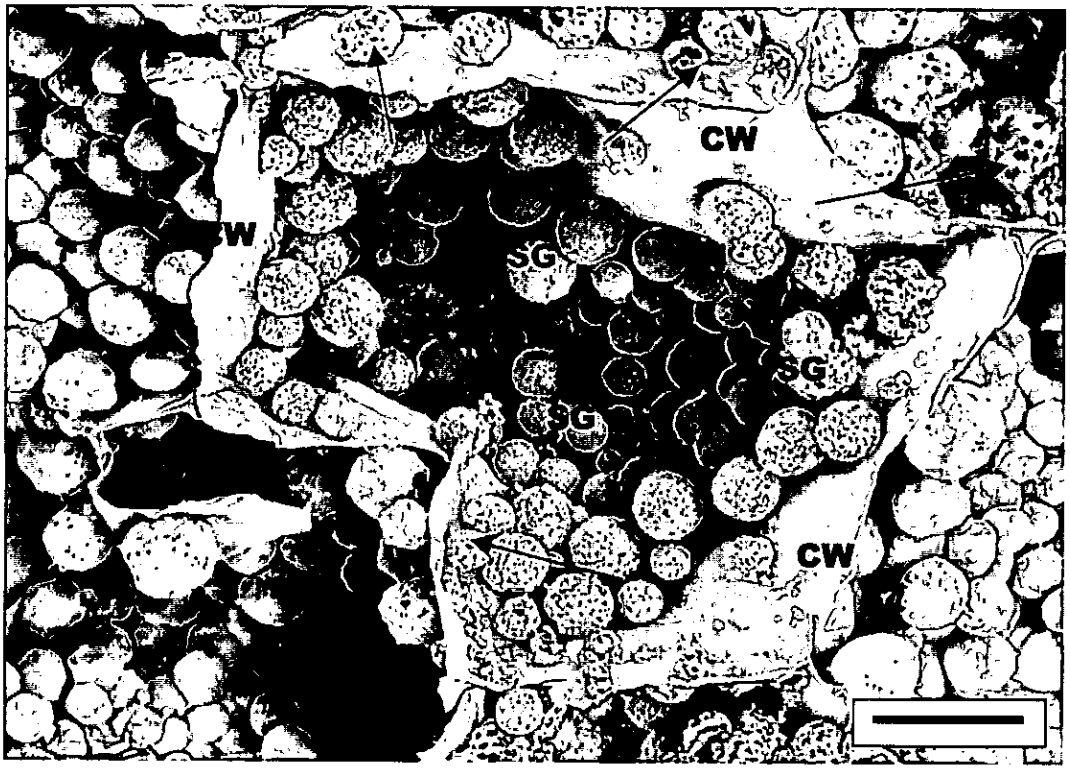


Figure 13.- Floury endosperm adjacent to scutellar epithelium of pearl millet of SDMV 89004 variety after 72 h of germination showing highly degraded starch granules (arrows) and intact cell wall (CW- Cell wall; SG- Starch granules) (Bar = 13 μm)

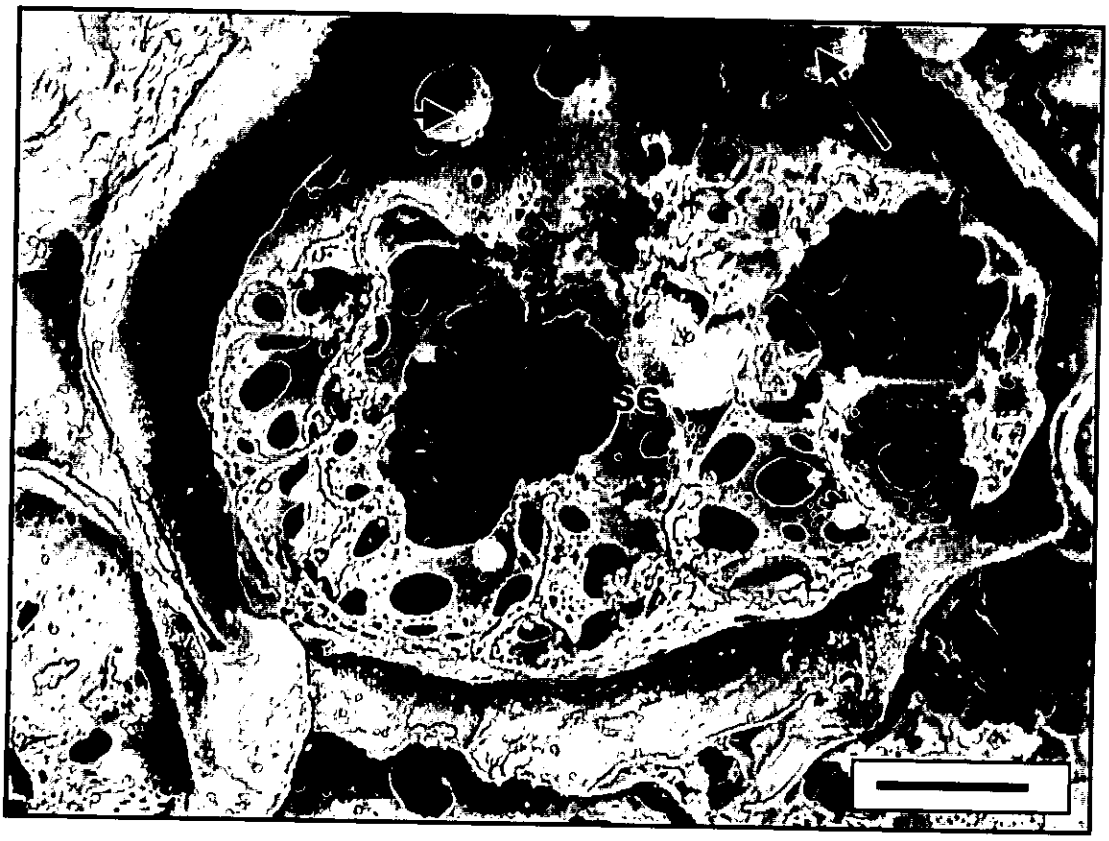


Figure 14.- Scanning electron micrograph of completely degraded starch granule in modified pearl millet malt of variety SDMV 89004 germinated for 48 h (Arrows- Protein body;SG- Starch granule) (Bar = 3 μm)

4.3 BREWING QUALITY ANALYSES OF MALTS

Pearl millet malts steeped for 8 h with a cycle of 2 h wet and 2 h dry air rest and germinated at 20 °, 25 °, 30 °, and 35 °C, medium watering treatment were analysed in terms of brewing quality, i.e. water uptake during germination, root and shoot growth during germination, Diastatic Power (DP), α- and β-amylase activity, free α-amino nitrogen (FAN), malt extract and malting loss. At the optimum germination temperature of 25 °C, high and low watering treatments, malts were analysed in terms of DP, total and soluble β-amylase activity (Betamyl assay), FAN, malt extract and malting loss. Total and soluble β-amylase activity (Betamyl assay) and malt extract of peal millet malts were compared to barley and sorghum malts standard.

4.3.1 Water uptake during steeping

The effects of steeping temperature and variety on water uptake during steeping of the two pearl millet varieties, at 8 h steeping time, are shown in Table 10 and described under 4.1.6.

4.3.2 Root and Shoot growth during germination

The effects of germination time, temperature and variety on root and shoot growth of the two pearl millet varieties are shown in Figure 15.

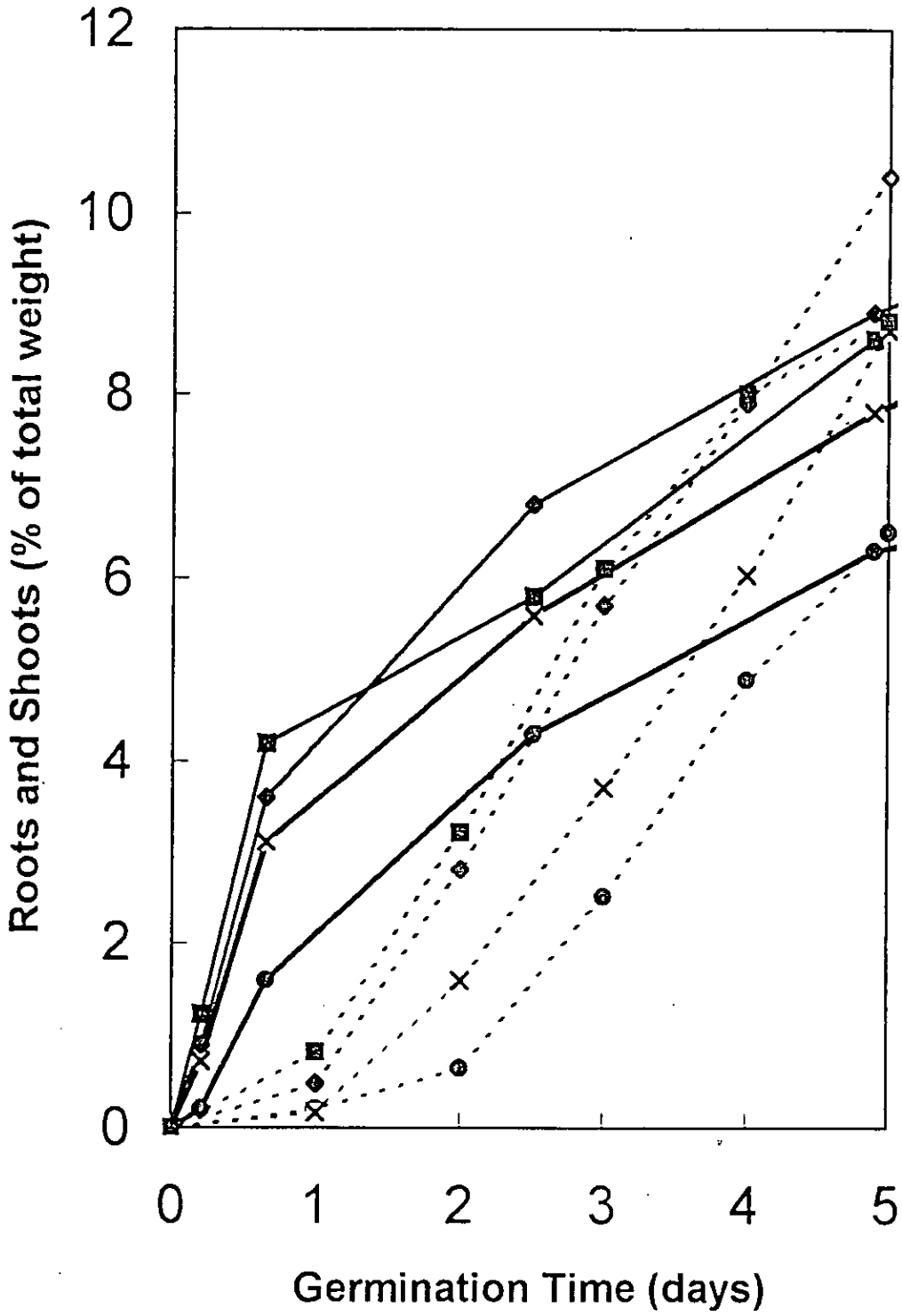


Figure 15.- Effects of germination time, temperature and variety on root and shoot growth of pearl millet (variety SDMV 89004 (–) and variety SDMV 91018 (--); (•- 20 °C; ×- 25 °C; ♦- 30 °C; ■- 35 °C)

The percentage of roots and shoots in pearl millet malt was significantly affected ($p < 0.001$) by germination time, temperature and variety. Roots and shoots increased with germination time. Germination at 20 °C gave the lowest root and shoot growth. Overall, germination at 30 ° and 35 °C gave the highest root and shoot growth ($p < 0.05$). Generally, variety SDMV 89004 had higher root and shoot growth than SDMV 91018. The highest percentage of roots and shoots 10.4 % was recorded at 5 days germination at 30 °C with variety SDMV 91018.

4.3.3 Diastatic Power (DP)

The effects of germination time, temperature and variety on the DP of the two pearl millet varieties are shown in Figure 16.

DP was also significantly affected ($p < 0.001$) by germination time and temperature and variety. Non-germinated pearl millet did not have any DP. DP increased as germination time increased. Generally, the DP of malt germinated at 20 °C was the lowest. Germination at 35 °C gave an initially higher level of DP. However, the rate of increase in DP at 35 °C declined over longer germination periods. Overall, germination at 30° and 35 °C gave the highest DP ($p < 0.05$). Variety SDMV 89004 gave about 30% higher DP than the SDMV 91018. The highest DP 40 PMDU/g was recorded at 5 days germination at 25 °C with variety SDMV 89004.

The effects of watering treatment, germination time and variety on DP of the two pearl millet varieties at the germination temperature of 25 °C are shown in Figure 17.

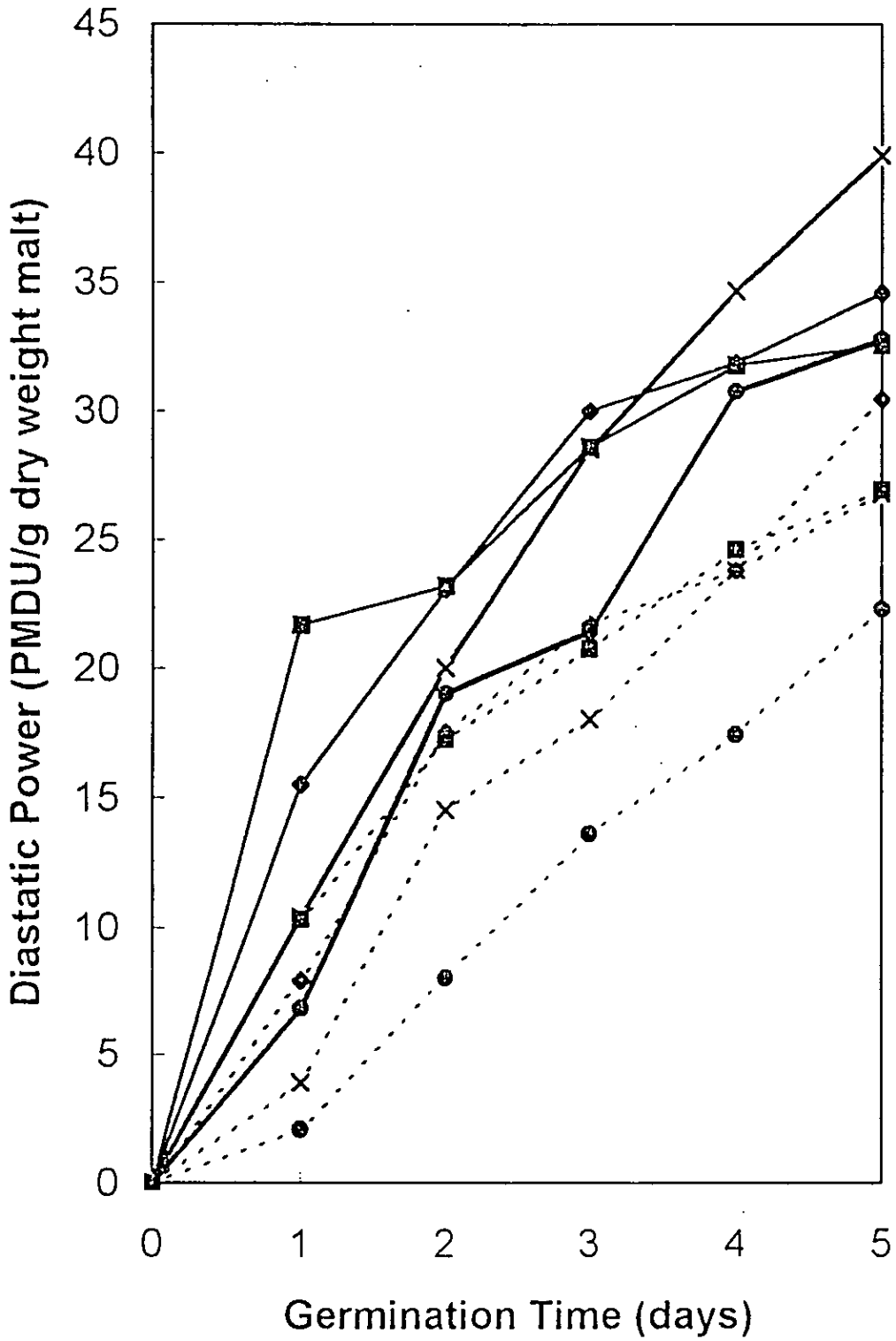


Figure 16.- Effects of germination time, temperature and variety on the DP of pearl millet (variety SDMV 89004 (–) and variety SDMV 91018 (--); (•-20 °C; ×- 25 °C; ♦- 30 °C; ■- 35 °C)

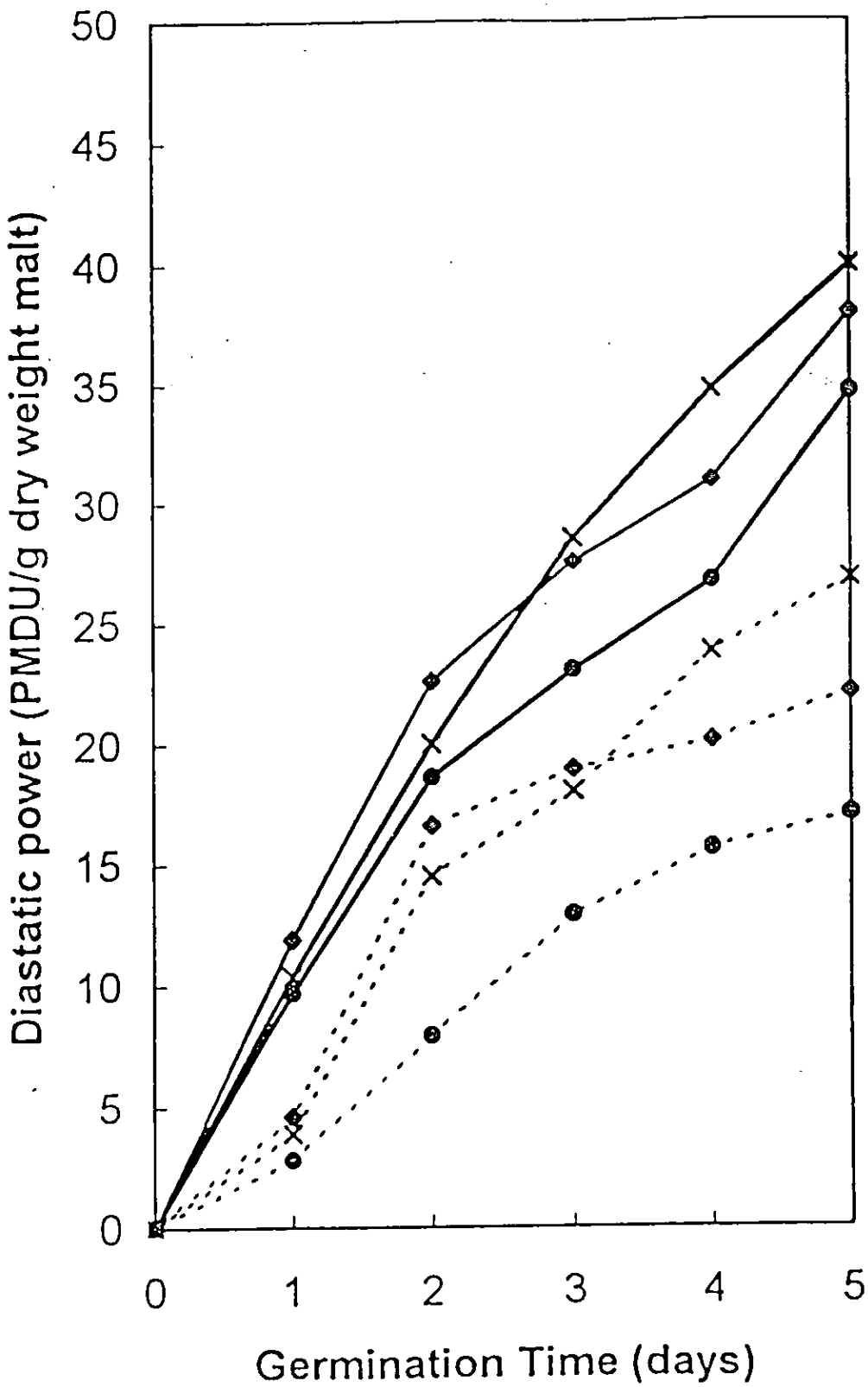


Figure 17.- Effects of germination time, watering treatment and variety on the DP of pearl millet at 25 °C (variety SDMV 89004(–) and variety SDMV 91018 (--) at various watering treatments (•- Low watering; ×- Medium watering; ♦- High watering)

DP was significantly affected ($p < 0.001$) by watering treatment and variety at 25 °C. DP increased as the germination moisture increased. At low watering treatment, DP was the lowest. Initially, germination at the high watering treatment gave the higher level of DP. However, the rate of increase in DP at high moisture levels declined over longer germination periods. Variety SDMV 89004 gave about 30% higher DP than the SDMV 91018. The highest DP 39.9 (PMDU/g) was recorded at 5 days germination at 25 °C, medium watering regime, with variety SDMV 89004.

4.3.4 Alpha-amylase activity

The effects of germination time, temperature and variety on the α-amylase activity of the two pearl millet varieties are shown in Figure 18.

As with DP, α-amylase activity was significantly affected ($p < 0.001$) by germination time, temperature and variety. There was practically no α-amylase activity in the non-germinated pearl millet. Alpha-amylase activity increased as the germination time increased. In general, α-amylase activity of pearl millet malt germinated at 20 °C was the lowest. Germination at 35 °C gave an initial higher level of α-amylase activity. However, as with DP, the rate of increase of α-amylase activity at 35 °C decreased as the germination process continued. Overall, germination at 30 ° and 35 °C gave the highest α-amylase activity, but 35 °C was not significantly different from 25 °C ($p > 0.05$). Variety SDMV 89004 gave about 40% higher α-amylase activity than the SDMV 91018. The highest α-amylase activity 33 PMDU/g was recorded at 5 days germination at 25°C with variety SDMV 89004.

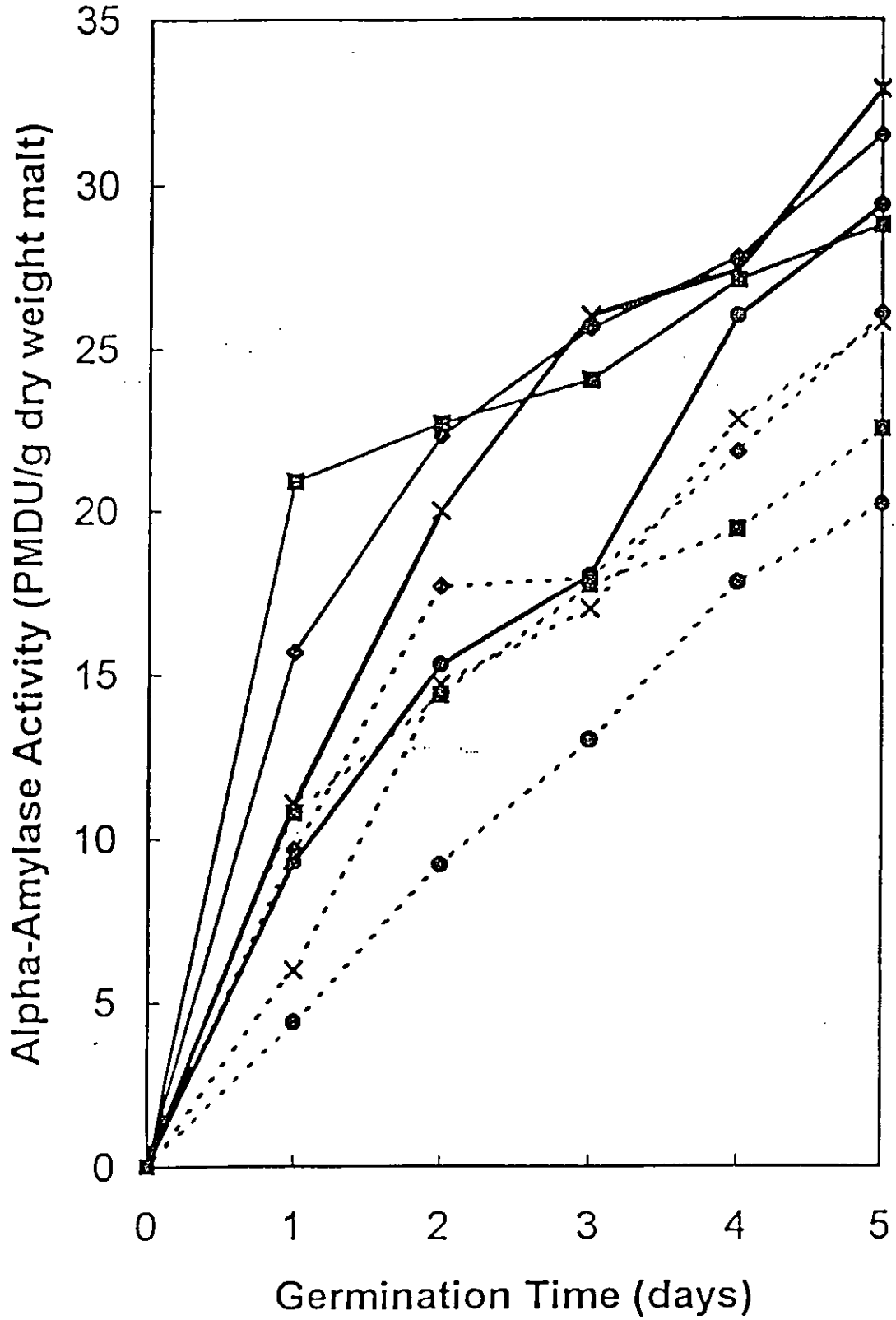


Figure 18.- Effects of germination time, temperature and variety on the α-amylase activity of pearl millet (variety SDMV 89004 (–) and variety SDMV 91018 (--); (• - 20 °C; × - 25 °C; ♦ - 30 °C; ■ - 35 °C)

4.3.5 Beta-amylase activity

The effects of germination time, temperature and variety on the β-amylase activity (measured by inactivation of α-amylase) of the two pearl millet varieties are shown in Figure 19.

As with DP and α-amylase, β-amylase was significantly affected ($p < 0.001$) by germination time and temperature and variety. The β-amylase activity of non-germinated pearl millet was negligible. Beta-amylase activity increased with germination time. As with DP and α-amylase, 20 °C gave the lowest β-amylase activity for both pearl millet varieties. Generally, germination at 35 °C produced a higher level of β-amylase activity during the initial germination period. However, as with DP and α-amylase, as the germination time progressed the rate of increase in β-amylase activity at 35 °C slowed down. Overall, germination at 30 ° and 35 °C gave the highest β-amylase activity ($p < 0.05$). However, at longer germination time 25 °C was the best. Variety SDMV 89004 exhibited almost 25% higher β-amylase activity than the SDMV 91018. The highest β-amylase activity 27 PMDU/g was recorded at 5 days germination at 35 °C with SDMV 89004 variety.

The effects of watering treatment, germination time and variety on total and soluble β-amylase activity (Betamyl assay) of the two pearl millet varieties at the germination temperature of 25 °C are shown in Figures 20 and 21.

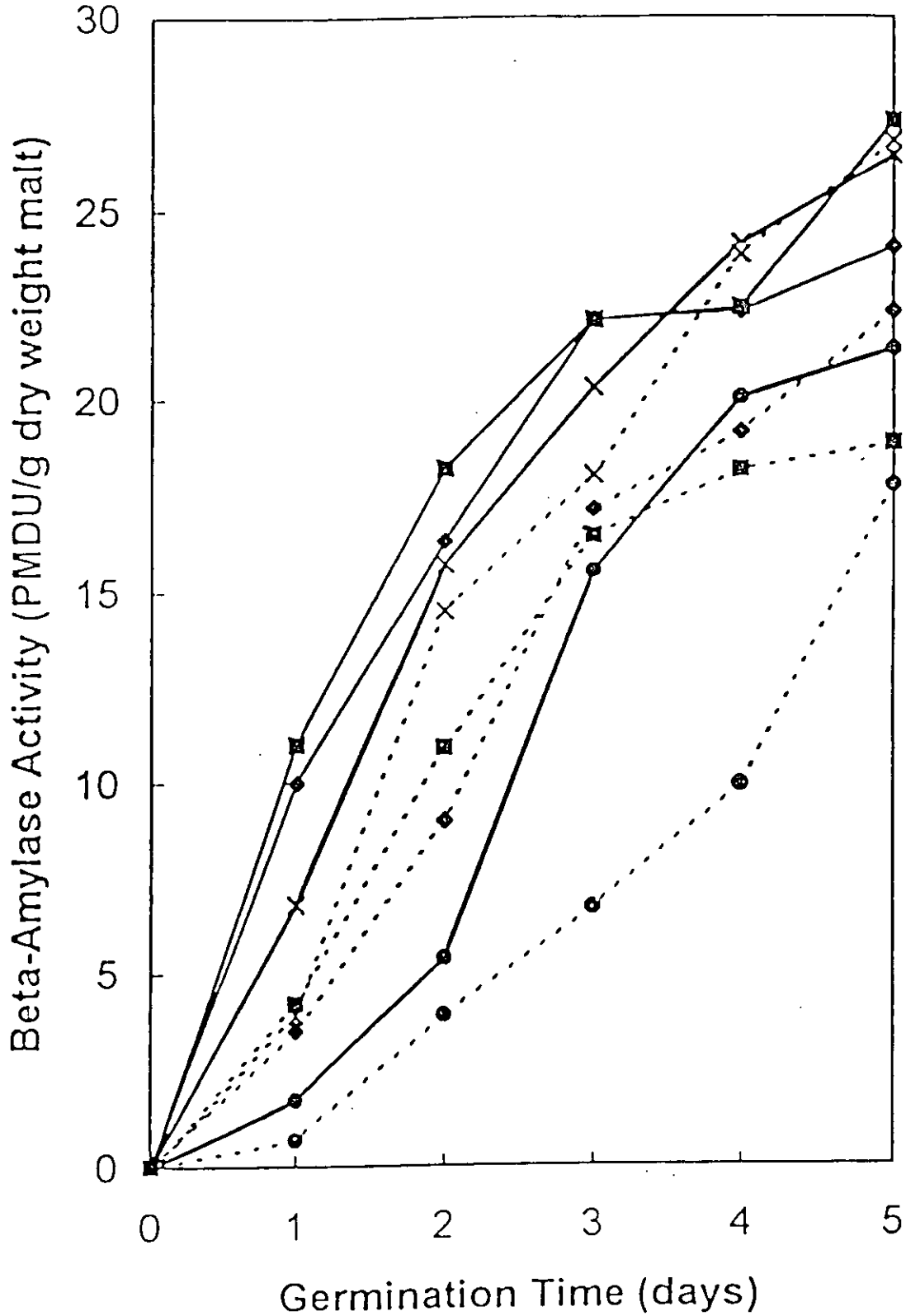


Figure 19.- Effects of germination time, temperature and variety on the β-amylase activity (measured by inactivation of α-amylase) of pearl millet (variety SDMV 89004 (–) and variety SDMV 91018 (--); (• - 20 °C; × - 25 °C; ♦ - 30 °C; ■ - 35 °C)

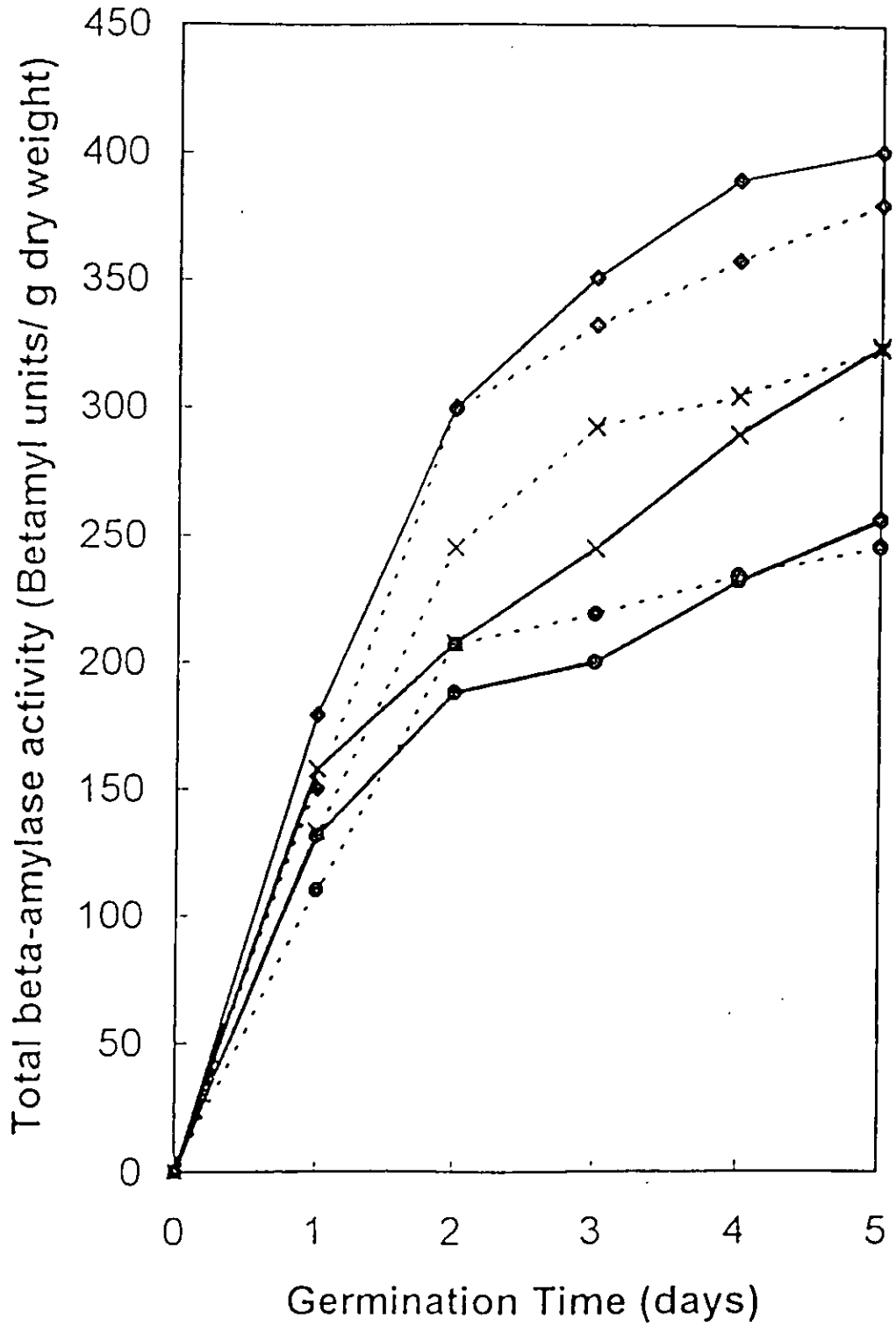


Figure 20.- Effects of germination time, watering treatment and variety on the total β-amylase activity (measured by Betamyl assay) of pearl millet at 25 °C (variety SDMV 89004(–) and variety SDMV 91018 (--) at various watering treatments (•- Low watering; ×- Medium watering; ♦- High watering)

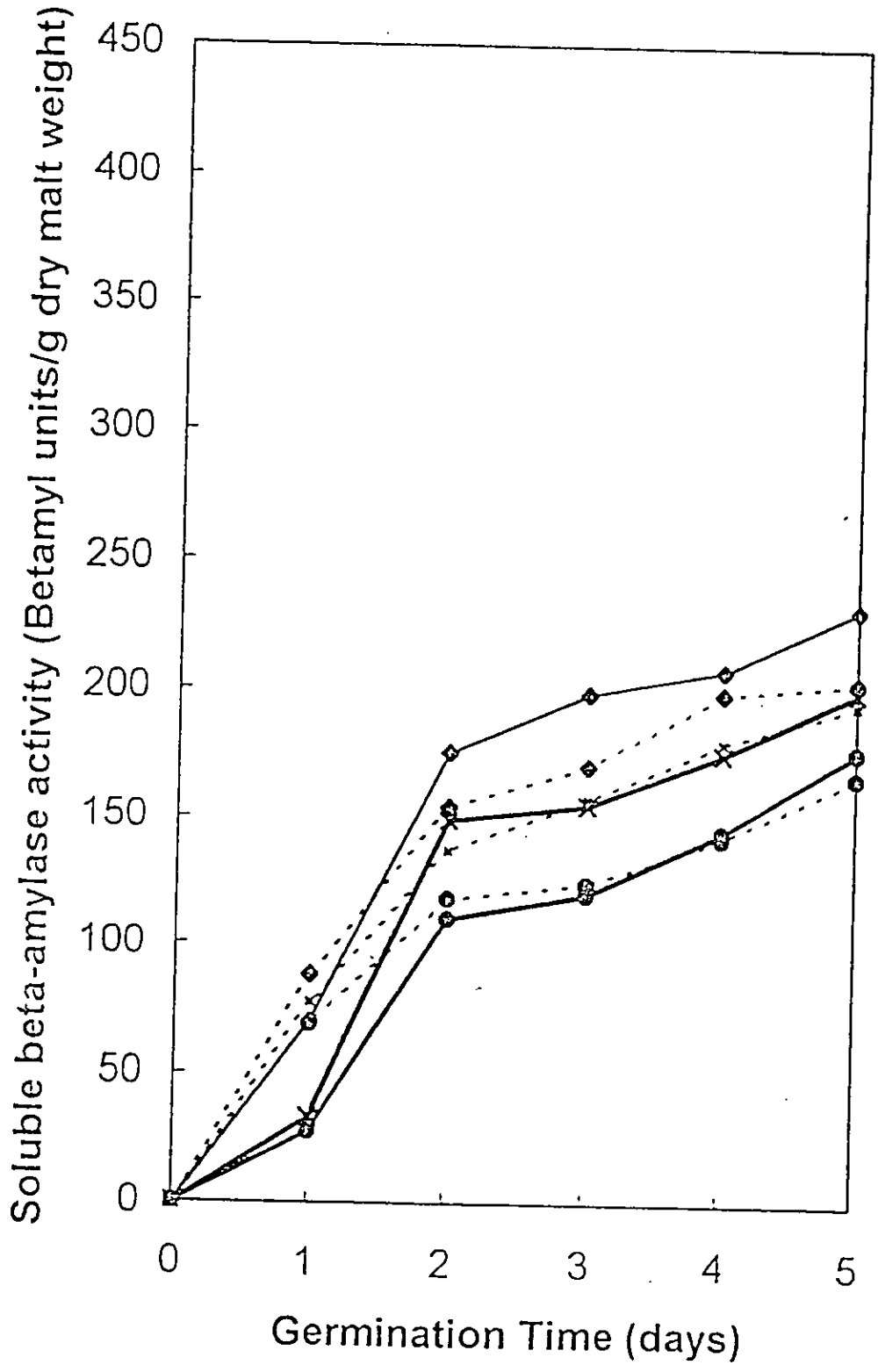


Figure 21.- Effects of germination time, watering treatment and variety on the soluble β-amylase activity (measured by Betamyl assay) of pearl millet at 25 °C (variety SDMV 89004(–) and variety SDMV 91018 (--) at various watering treatments (•- Low watering; ×- Medium watering; ♦- High watering)

At 25 °C, malt total and soluble β-amylase activity were significantly affected by watering treatment. Generally, variety did not affect total and soluble β-amylase activity. However, variety SDMV 89004 at high watering treatment had significantly higher ($p < 0.05$) total and soluble β-amylase activity than SDMV 91018. Total and soluble β-amylase increased with germination moisture and germination time. As with DP, at low watering treatment, total and soluble β-amylase activity was the lowest. Overall, germination at a high moisture content gave a higher level of total and soluble β-amylase activity, than the low and medium watering treatments. Total and soluble β-amylase activity of pearl millet malts was lower than that of barley malt standard, which had 727.7 and 375.5 (Betamyl units/ g), respectively, but higher than sorghum malt standard, which had 203.0 and 108.8 (Betamyl units/g), respectively.

Total β-amylase activity was about 50% higher than soluble β-amylase activity. The highest total and soluble β-amylase activity 400.1 and 232.0 (Betamyl units/g), respectively, were recorded at 5 days germination at high watering treatment with SDMV 89004 variety.

4.3.6 Free α-amino nitrogen (FAN)

The effects of germination time, temperature and variety on the malt FAN of the two pearl millet varieties are shown in Figure 22.

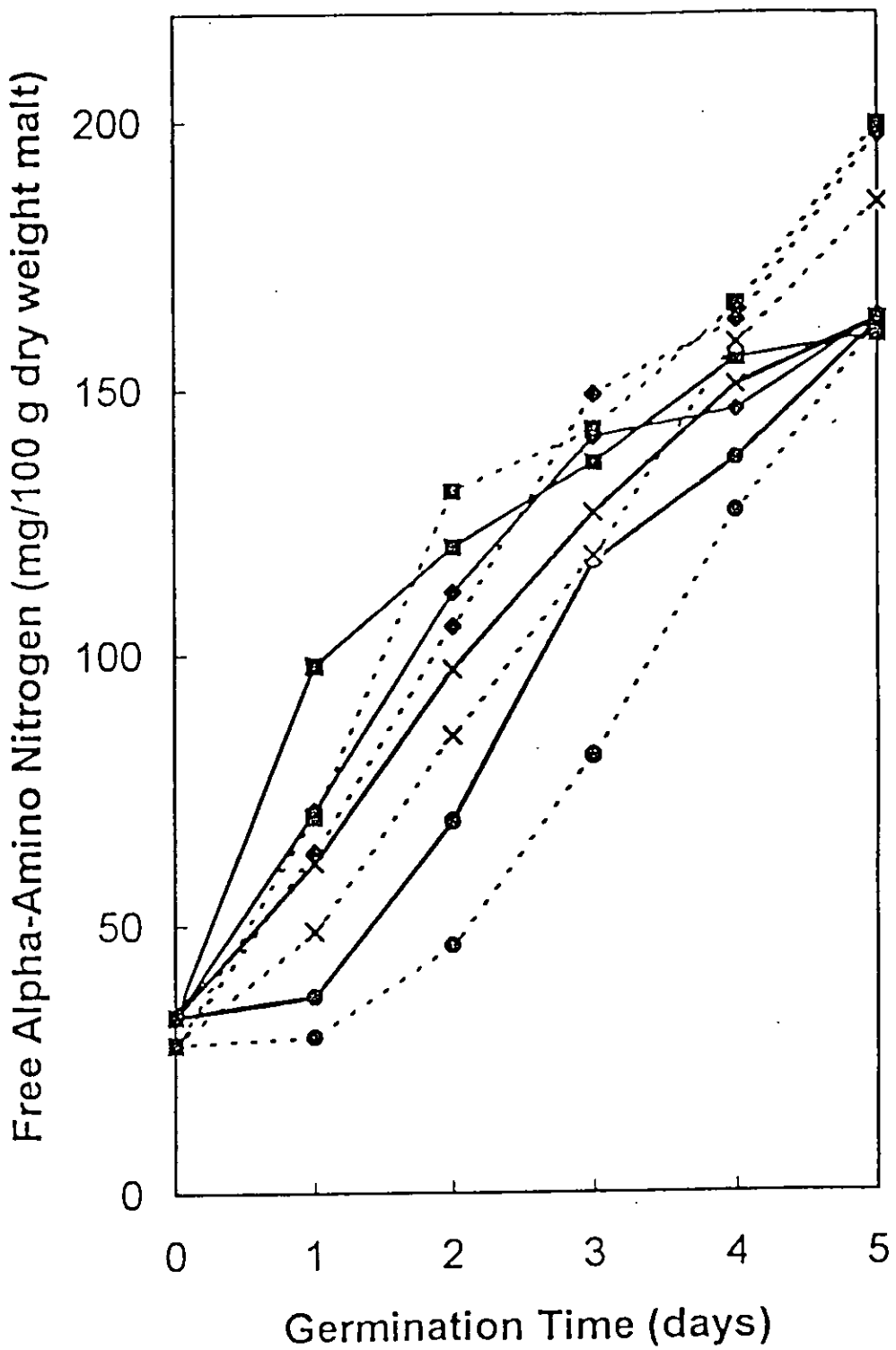


Figure 22.- Effects of germination time, temperature and variety on the malt FAN of pearl millet (variety SDMV 89004 (–) and variety SDMV 91018 (--); (•- 20 °C; ×- 25 °C; ♦- 30 °C; ■- 35 °C)

FAN was significantly affected ($p < 0.001$) by time and temperature of germination. Variety did not have a significant effect on malt FAN. Both varieties of non-germinated pearl millet grains contained some FAN, 32.9 and 27.6 mg/100 g for varieties SDMV 89004 and SDMV 91018, respectively. Malt FAN increased with germination time. The germination temperature of 20 °C generally gave the lowest FAN. Overall, germination at 35 °C gave the highest malt FAN ($p < 0.05$). Germination at 25 ° and 30 °C gave intermediate malt FAN. Although malt FAN was similar overall for both pearl millet varieties, malt FAN of SDMV 89004 at 5 days germination was generally lower than that of SDMV 91018. The highest level of FAN 199 mg FAN/100 g malt was recorded at 5 days germination at 35 °C with variety SDMV 91018.

The effects of watering treatment, germination time and variety at 25°C germination temperature on the malt FAN of the two pearl millet varieties are shown in Figure 23.

At 25 °C, FAN was significantly affected ($p < 0.001$) by watering treatment. Generally, variety did not have a significant effect ($p < 0.05$) on malt FAN at 25 °C. FAN increased as germination time and moisture increased. Germination at the low watering treatment gave the lowest malt FAN. Germination at high watering treatment gave the highest malt FAN. Generally, the malt FAN was similar for both pearl millet varieties. Except that malt FAN of SDMV 89004 at 5 days germination was generally lower than that of SDMV 91018. The highest level of FAN 191 mg FAN/100 g malt was recorded at 5 days germination with variety SDMV 91018 at high watering treatment.

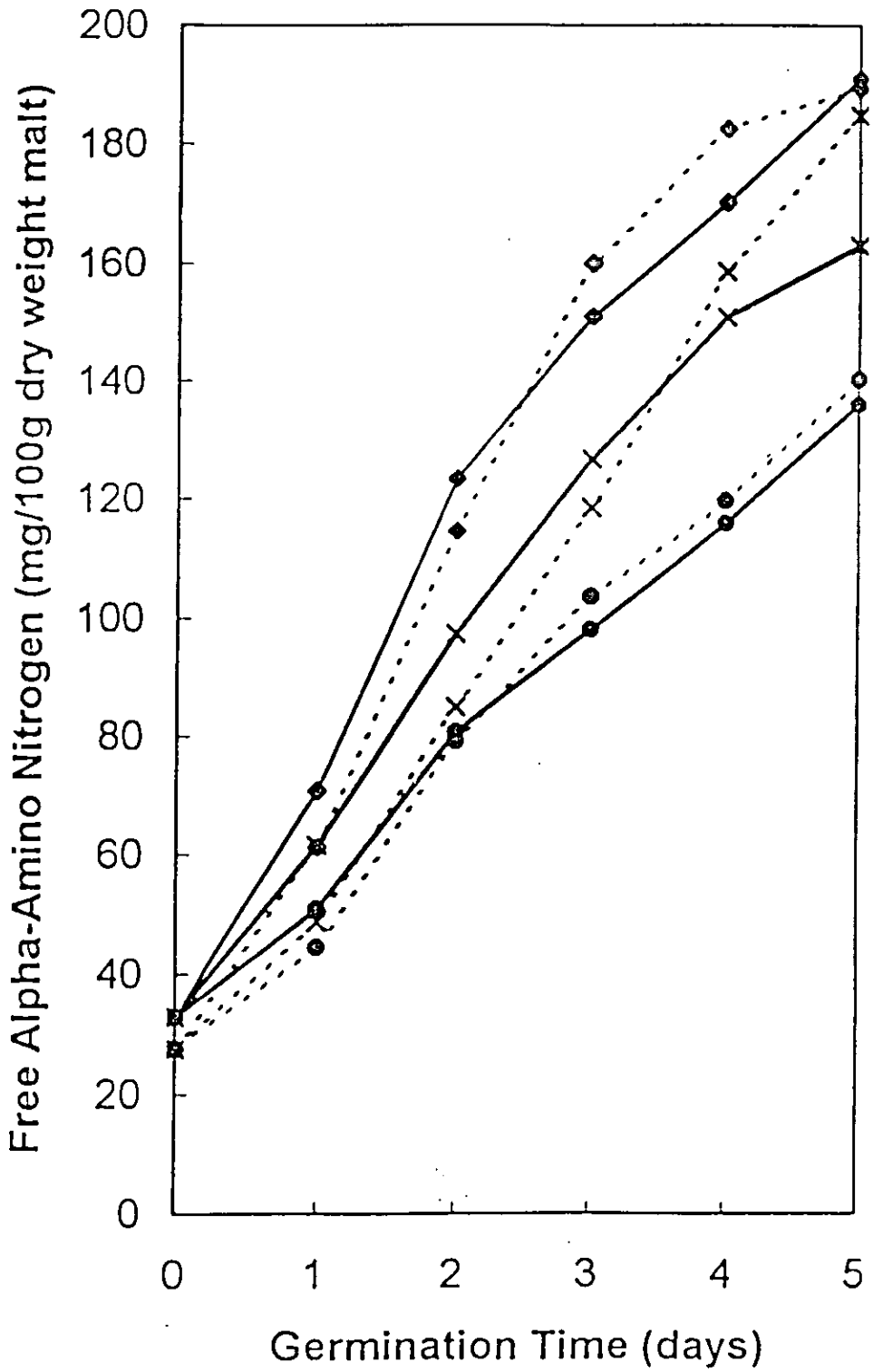


Figure 23.- Effects of germination time, watering treatment and variety on the malt FAN of pearl millet at 25 °C (variety SDMV 89004(–) and variety SDMV 91018 (--) at various watering treatments (•- Low watering; ×- Medium watering; ♦- High watering)

4.3.7 Hot water extract

The effects of watering treatment, germination time and variety on malt extract of the two pearl millet varieties at 25 °C germination temperature are shown in Figure 24.

Hot water malt extract was significantly affected ($p < 0.001$) by germination time, watering treatment and variety. Generally, variety SDMV 91018 had lower malt extract than SDMV 89004. Malt extract increased with germination time. Both non-germinated pearl millet varieties contained some extract, 18.6 and 18.5% for the varieties SDMV 89004 and SDMV 91018, respectively. Germination at the low moisture level gave continuously lower malt extract. Germination at the high moisture level gave the highest malt extract in both varieties. Malt extract of pearl millet malt was higher than that of the sorghum malt standard, which had 22.1% and generally lower than the barley malt standard, which had 59.4% malt extract. The highest malt extract 69.0 % was recorded at 5 days germination at high moisture germination, with variety SDMV 89004.

4.3.8 Malting loss

The effects of germination time, temperature and variety on malting loss of the two pearl millet varieties are shown in Figure 25.

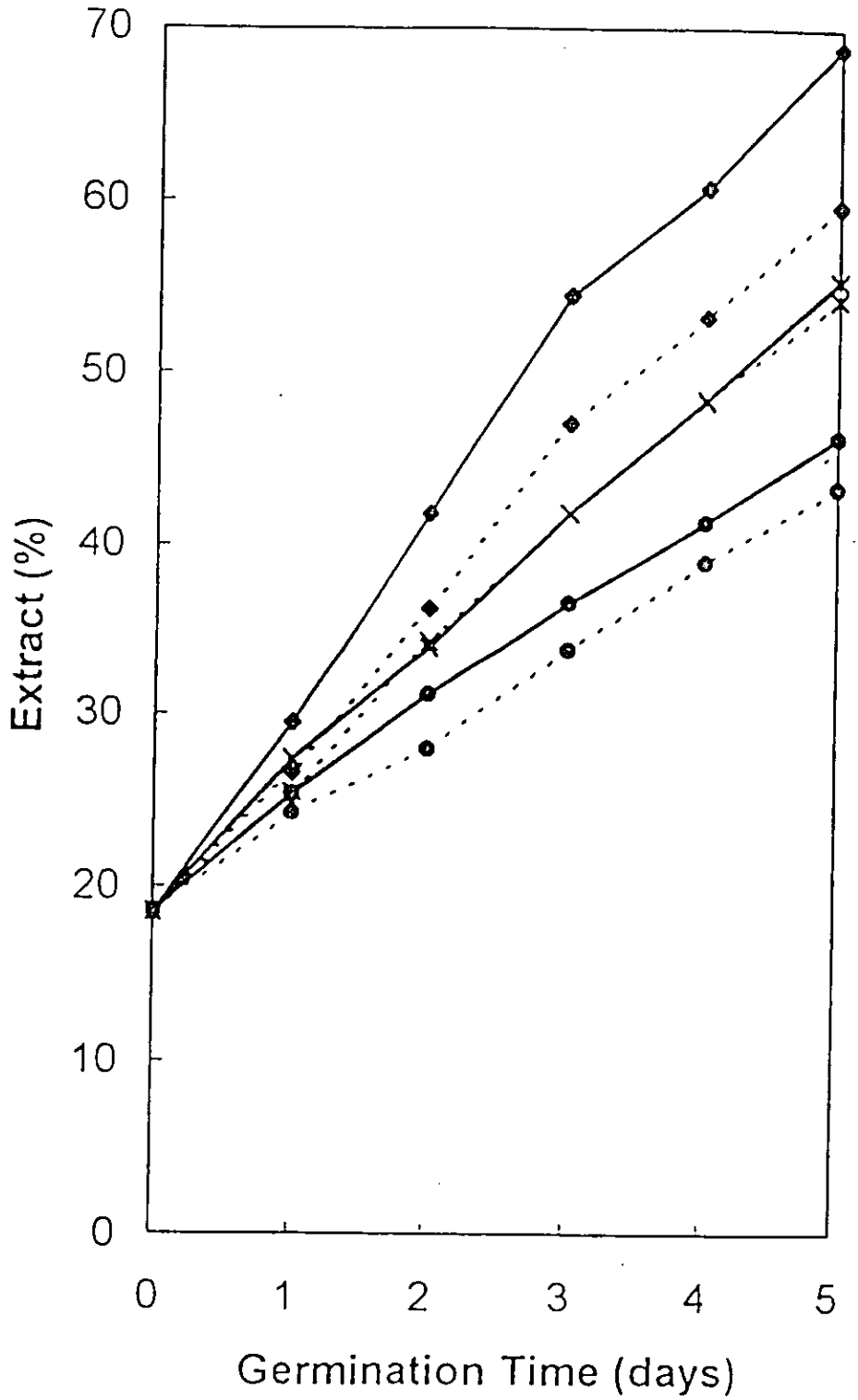


Figure 24.- Effects of germination time, watering treatment and variety on malt extract of pearl millet at 25 °C (variety SDMV 89004(–) and variety SDMV 91018 (--) at various watering treatments (•- Low watering; x- Medium watering; ♦- High watering)

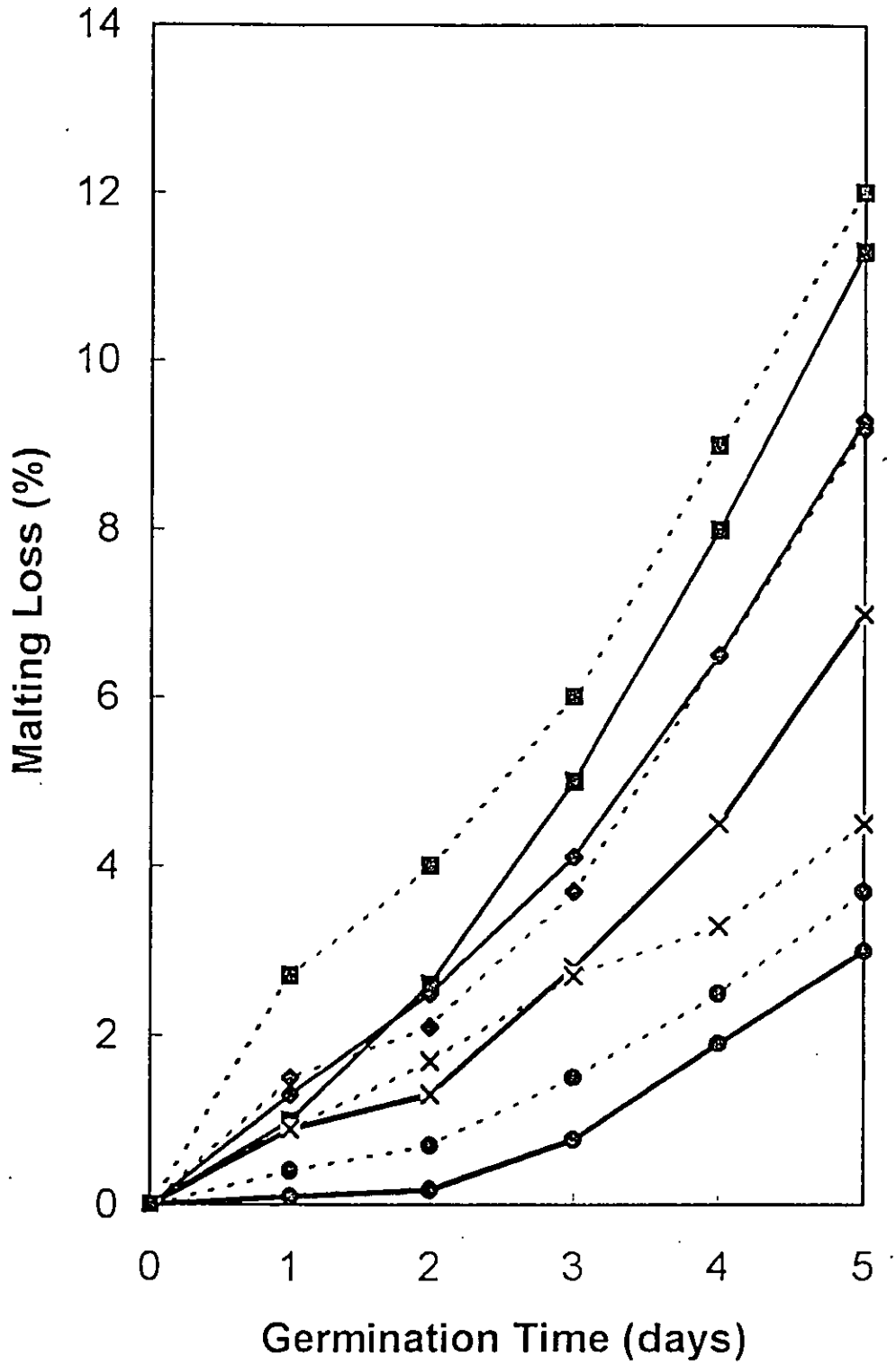


Figure 25.- Effects of germination time, temperature and variety on malting loss of pearl millet (variety SDMV 89004 (–) and variety SDMV 91018 (--); (•- 20 °C; ×- 25 °C; ♦- 30 °C; ■- 35 °C)

Malting loss was significantly affected ($p < 0.001$) by germination time and temperature. Overall, variety did not effect malting loss. Malting loss increased as the germination time increased. Germination at 20 °C gave the lowest malting loss. Malting loss was directly temperature dependent. Overall, germination at 35 °C gave the highest malting loss ($p < 0.05$). The germination temperature of 25 ° and 30 °C gave intermediate malting losses. The highest malting loss 12.0% was recorded at 5 days germination at 35 °C with variety SDMV 91018.

The effects of watering treatment, germination time and variety on malting loss of the two pearl millet varieties at 25 °C germination temperature are shown in Figure 26.

Malting loss at 25 °C was significantly affected by watering treatment. Malting loss, at 25 °C, increased as the germination moisture increased. Germination at low moisture content gave the lowest malting loss. Germination at high moisture content gave the highest malting loss. In general, malting loss was similar for both pearl millet varieties. The highest malting loss 9.2% was recorded at 5 days germination with variety SDMV 89004 at high watering treatment.

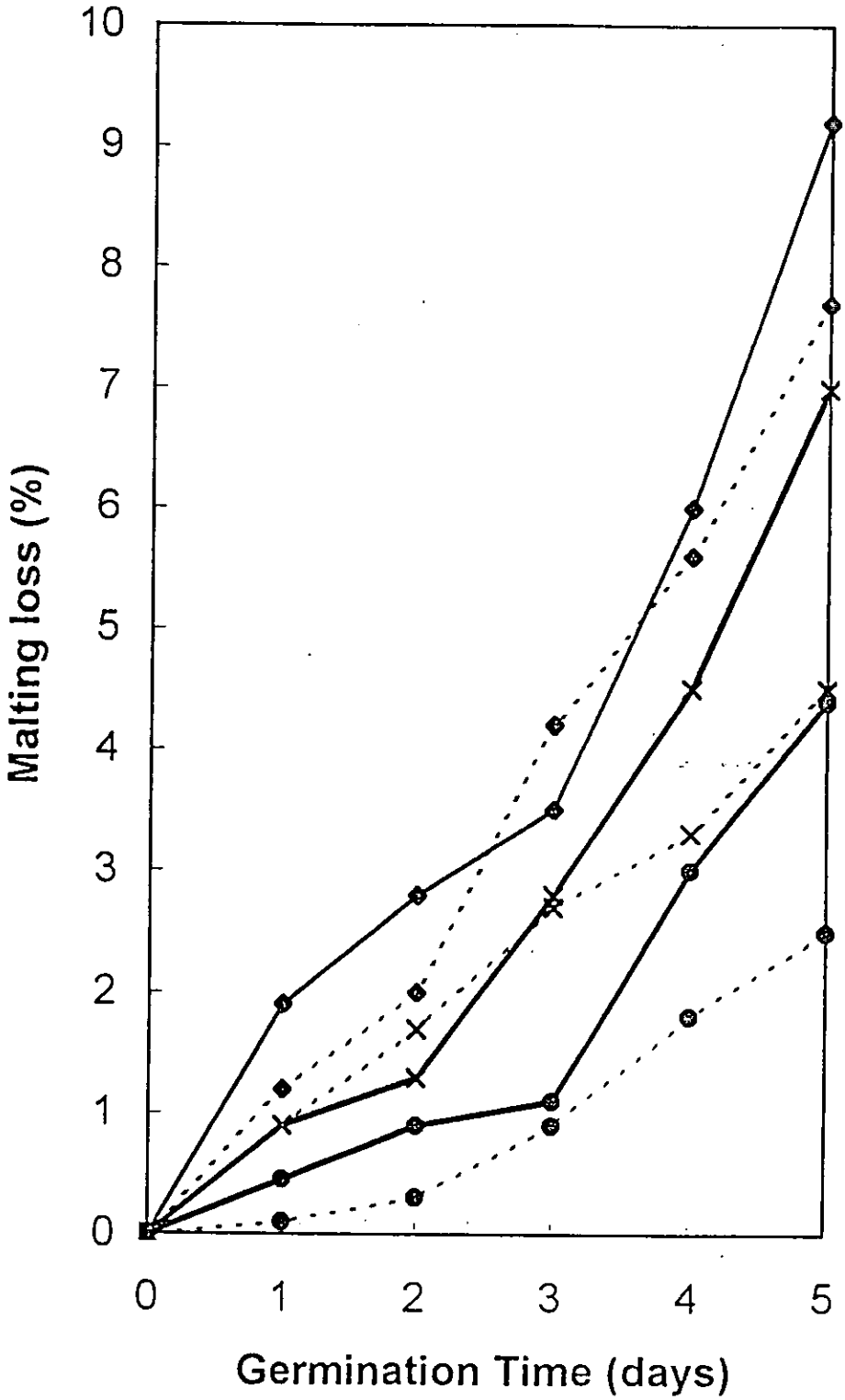


Figure 26.- Effects of germination time, watering treatment and variety on malting loss of pearl millet at 25 °C (variety SDMV 89004(–) and variety SDMV 91018 (--) at various watering treatments (•- Low watering; ×- Medium watering; ♦- High watering)

4.4 NUTRITIONAL AND FUNCTIONAL ANALYSES OF MALTS

Pearl millet malts germinated at the optimum temperature of 25 °C, medium watering treatment were analysed in terms of nutritional and functional properties.

4.4.1 Total Carbohydrate content and the percentage of the total carbohydrate which was enzyme susceptible

The effects of germination time and variety on total carbohydrate content and on the percentage of the TCES in the two pearl millet varieties are shown in Figure 27.

The total carbohydrate content was significantly affected ($p < 0.001$) by germination time and variety. Non-germinated pearl millet grains had a total carbohydrate content of 78.4% and 75.3% for varieties SDMV 89004 and SDMV 91018, respectively. The total carbohydrate content decreased with germination time for both pearl millet varieties. Generally, germinated pearl millet of variety SDMV 89004 had higher carbohydrate content than SDMV 91018. The lowest total carbohydrate content 53.8% was observed at 5 days germination with variety SDMV 89004.

The percentage of the TCES was significantly affected ($p < 0.001$) by germination time and variety. Non-germinated pearl millet grains had a percentage of the TCES of 10.6% and 10.1% for the varieties SDMV 89004 and SDMV 91018, respectively. The percentage of the TCES increased with germination time for both pearl millet varieties. The percentage of the TCES was higher in SDMV 89004.

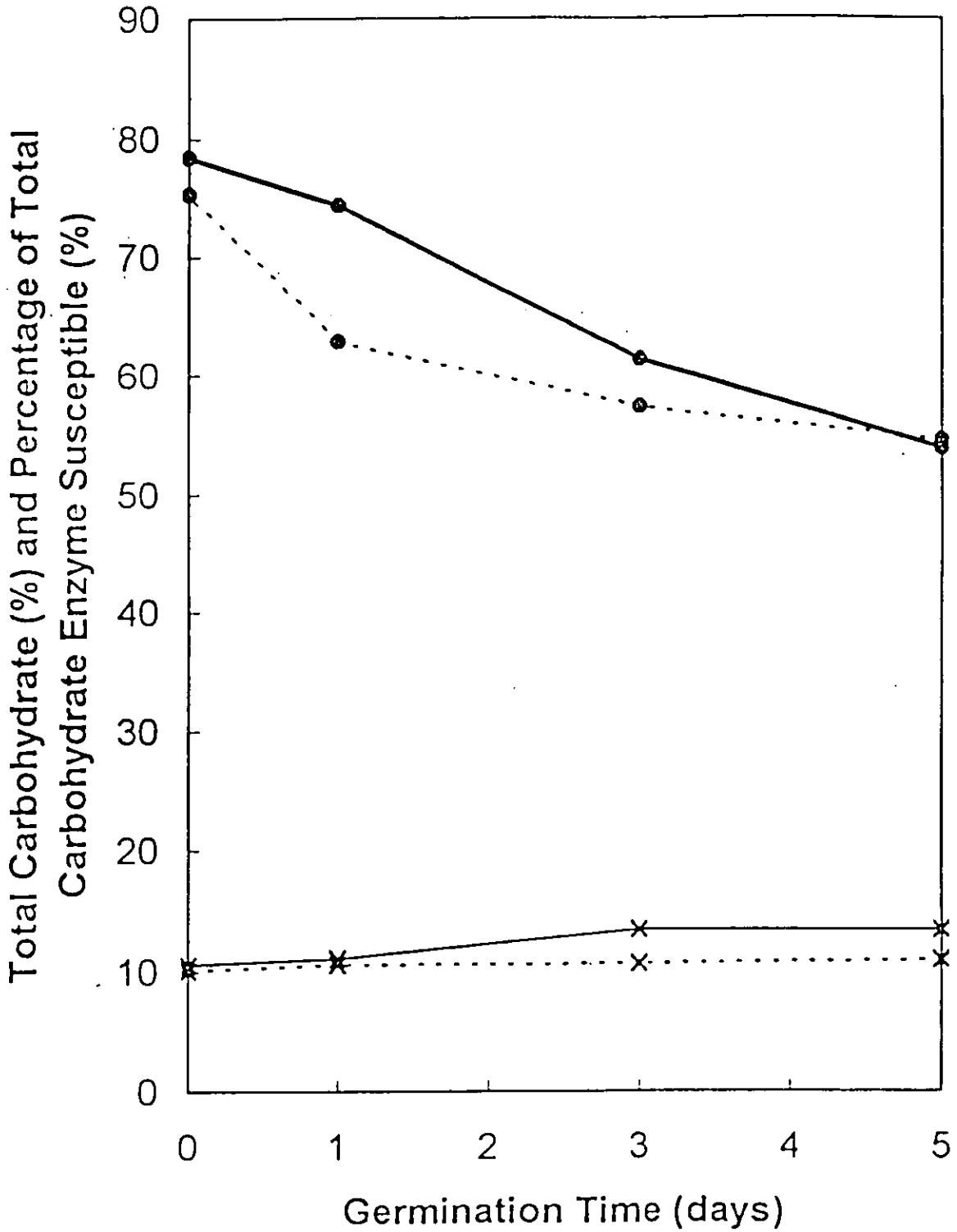


Figure 27.- Effects of germination time and variety on the total carbohydrate content and the percentage of the total carbohydrate which was enzyme susceptible of pearl millet (variety SDMV 89004(–) and variety SDMV 91018 (--); total carbohydrate (•-); the total carbohydrate which was enzyme susceptible (×-))

The highest percentage of the TCES of 13.5% was recorded at 3 and 5 days germination with variety SDMV 89004.

4.4.2 Water Absorption Index (WAI) and Water Solubility Index (WSI)

The effects of germination time and variety on Water Absorption Index and Water Solubility Index of the two pearl millet varieties are shown in Figure 28.

WAI was significantly affected ($p < 0.001$) by germination time in both varieties. Non-germinated pearl millet had a WAI of 10.4 (g/g) and 10.6 (g/g) for variety SDMV 89004 and SDMV 91018, respectively. Variety did not affect WAI. WAI decreased as the germination time increased. The lowest WAI 9.0 (g/g) was recorded at 5 days germination with variety SDMV 89004.

WSI was significantly affected ($p < 0.001$) by germination time in both pearl millet varieties. Non-germinated pearl millet had a WSI of 10.0% and 10.6% for the varieties SDMV 89004 and SDMV 91018, respectively. Variety did not affect WSI. WSI increased as the germination time increased. The highest WSI 11.6% was record at 5 days germination with variety SDMV 91018.

4.4.3 Fat

The effects of germination time and variety on the fat content of the two pearl millet varieties are shown in the Figure 29.

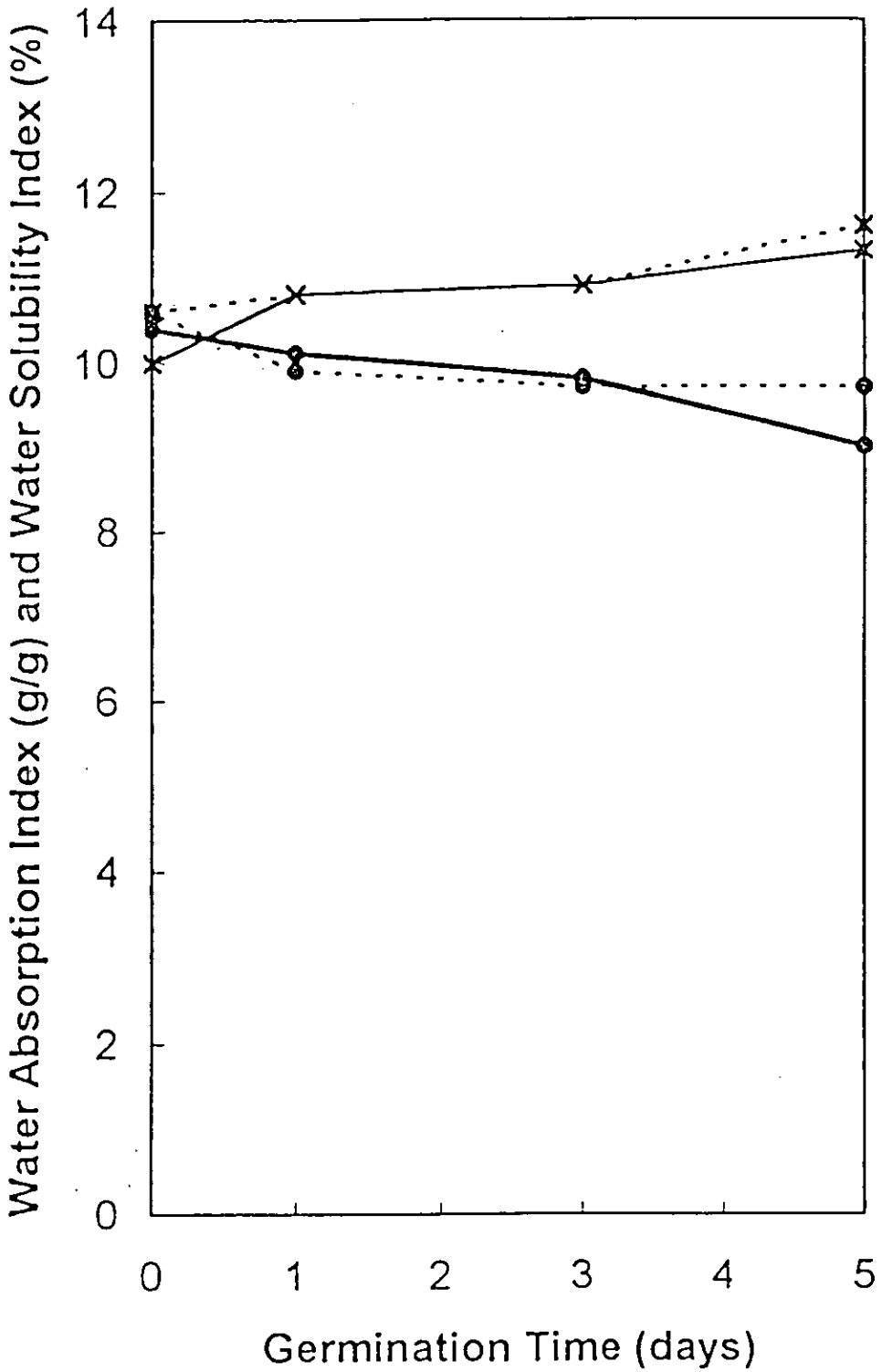


Figure 28.- Effects of germination time and variety on WAI and WSI of two pearl millet (variety SDMV 89004(–) and variety SDMV 91018 (--); water absorption index (•-); water solubility index (×-))

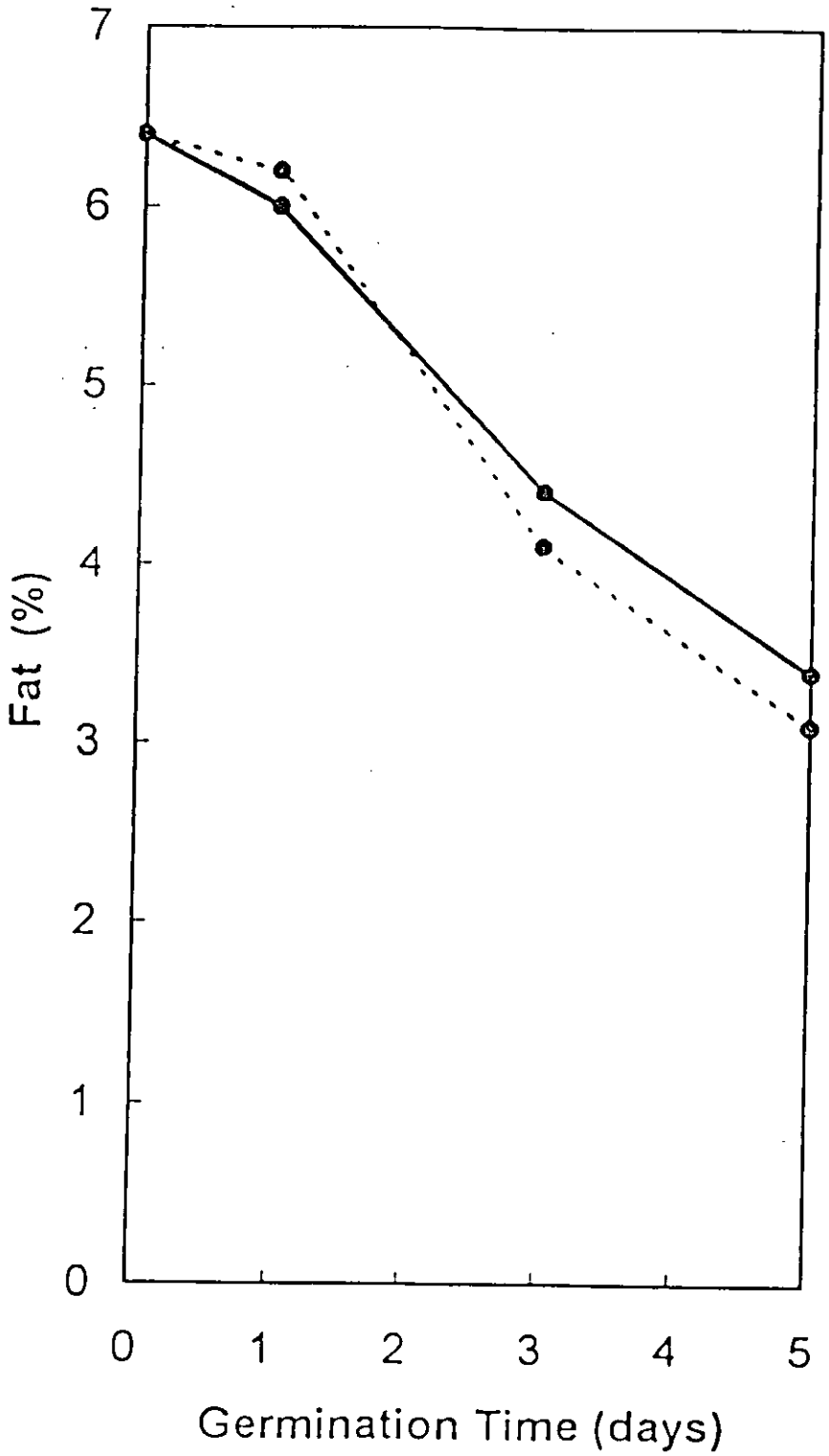


Figure 29.- Effects of germination time and variety on the fat content of pearl millet (variety SDMV 89004(–) and variety SDMV 91018 (--))

Germination time significantly affected ($p < 0.001$) the fat content of both pearl millet varieties. Non-germinated pearl millet grains had a crude fat content of 6.4% for both varieties. In general, variety did not have any effect on the fat content. The percentage fat content decreased with germination time. The lowest fat content 3.1% was recorded at 5 days germination with variety SDMV 91018.

4.4.4 Protein

The effects of germination time and variety on the protein content of the two pearl millet varieties are shown in Figure 30.

The protein content of pearl millet malts was affected by germination time and variety ($p < 0.05$). Non-germinated pearl millet grains had a protein content of 11.7% and 11.3% for varieties SDMV 89004 and SDMV 91018, respectively. There was a decrease in protein content of the malts compared with control samples. Germinated SDMV 89004 had higher protein than that of SDMV 91018 variety. The lowest protein content 10.8% was recorded at 3 and 5 days germination with variety SDMV 91018.

4.4.5 Nitrogen Solubility Index (NSI) and soluble nitrogen

The effects of germination time and variety on the NSI and soluble nitrogen of the two pearl millet are shown in Figures 31 and 32, respectively.

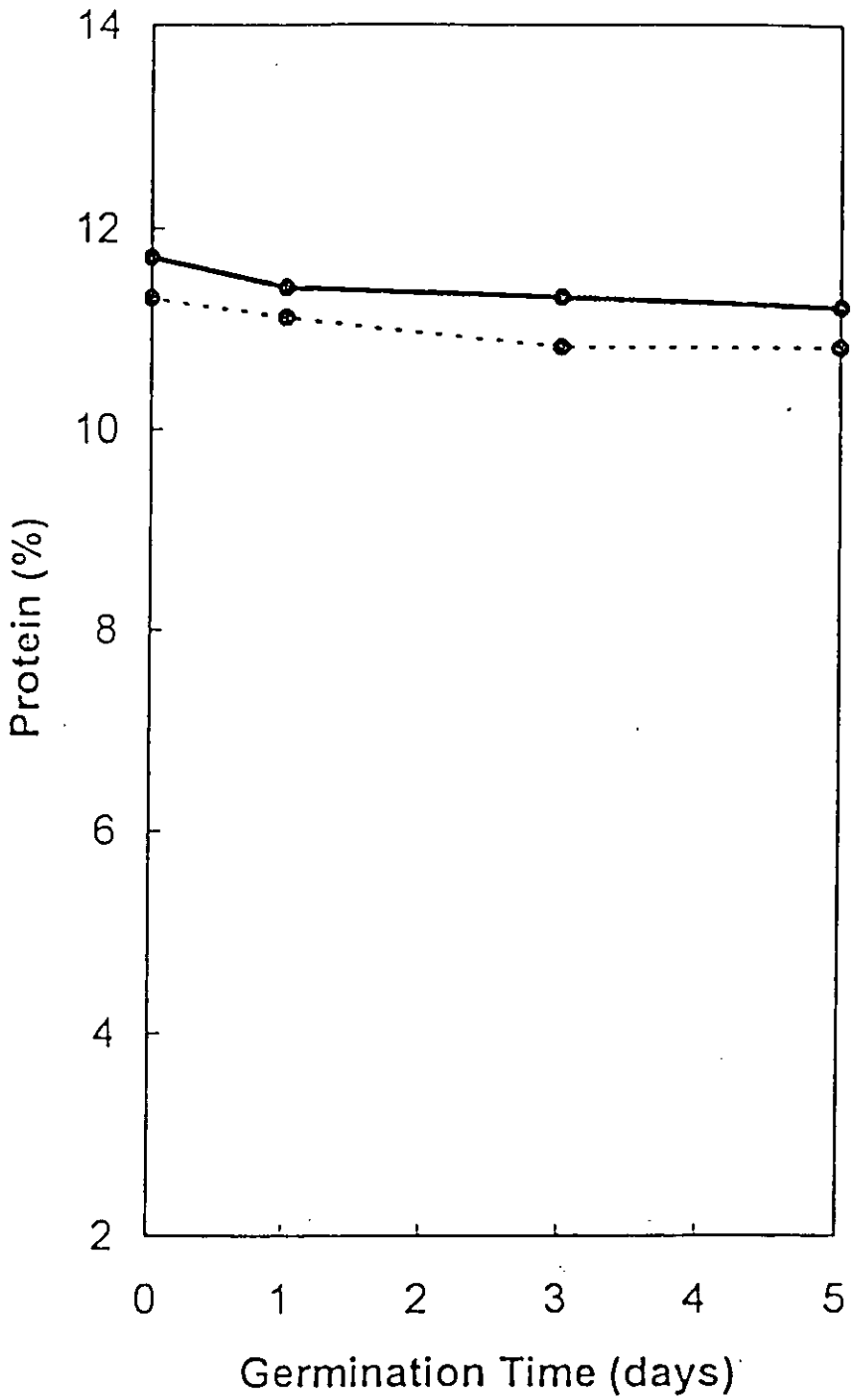


Figure 30.- Effects of germination time and variety on the protein content of pearl millet (variety SDMV 89004(–) and variety SDMV 91018 (--))

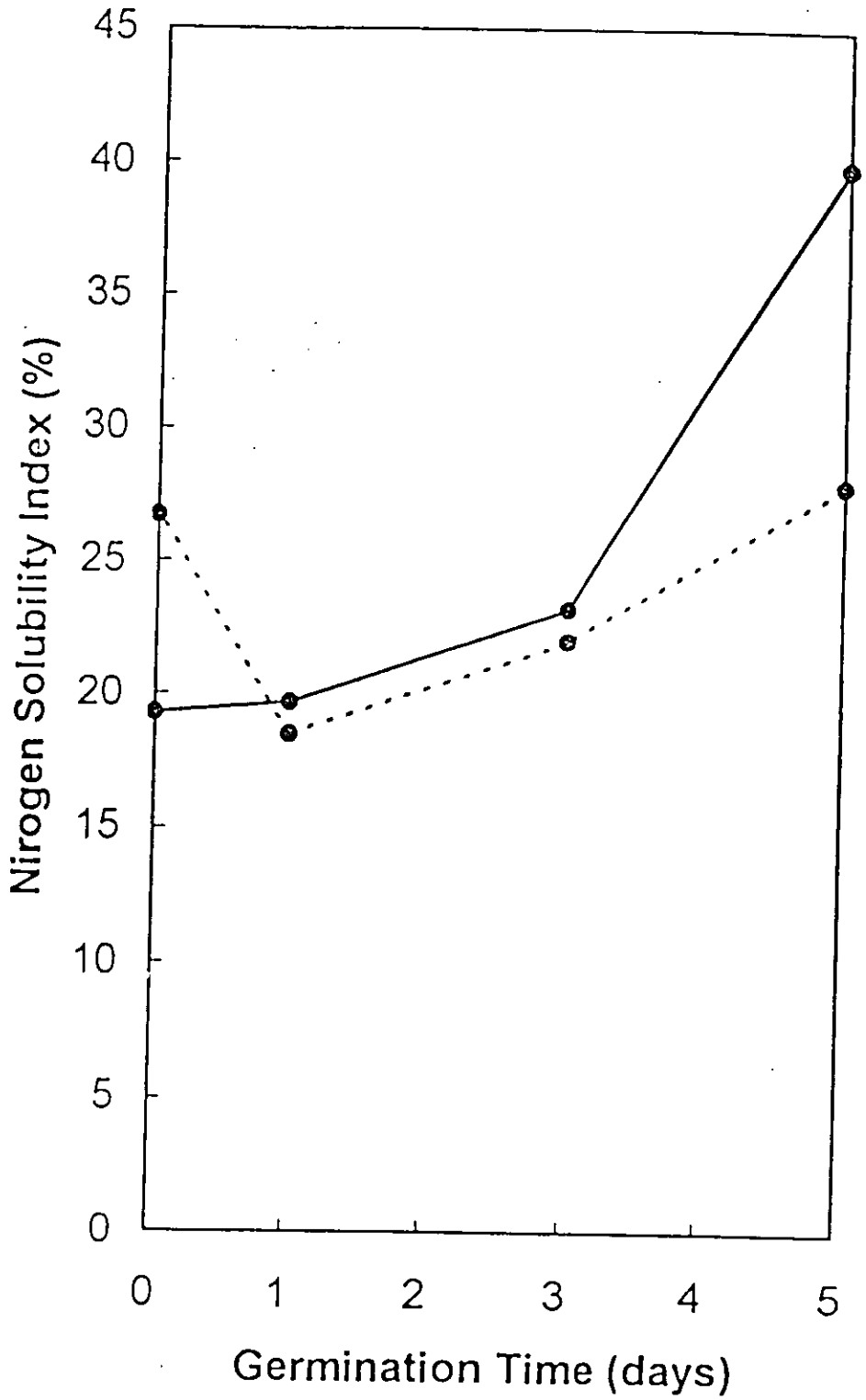


Figure 31.- Effects of germination time and variety on the Nitrogen Solubility Index of pearl millet (variety SDMV 89004(–) and variety SDMV 91018 (--))

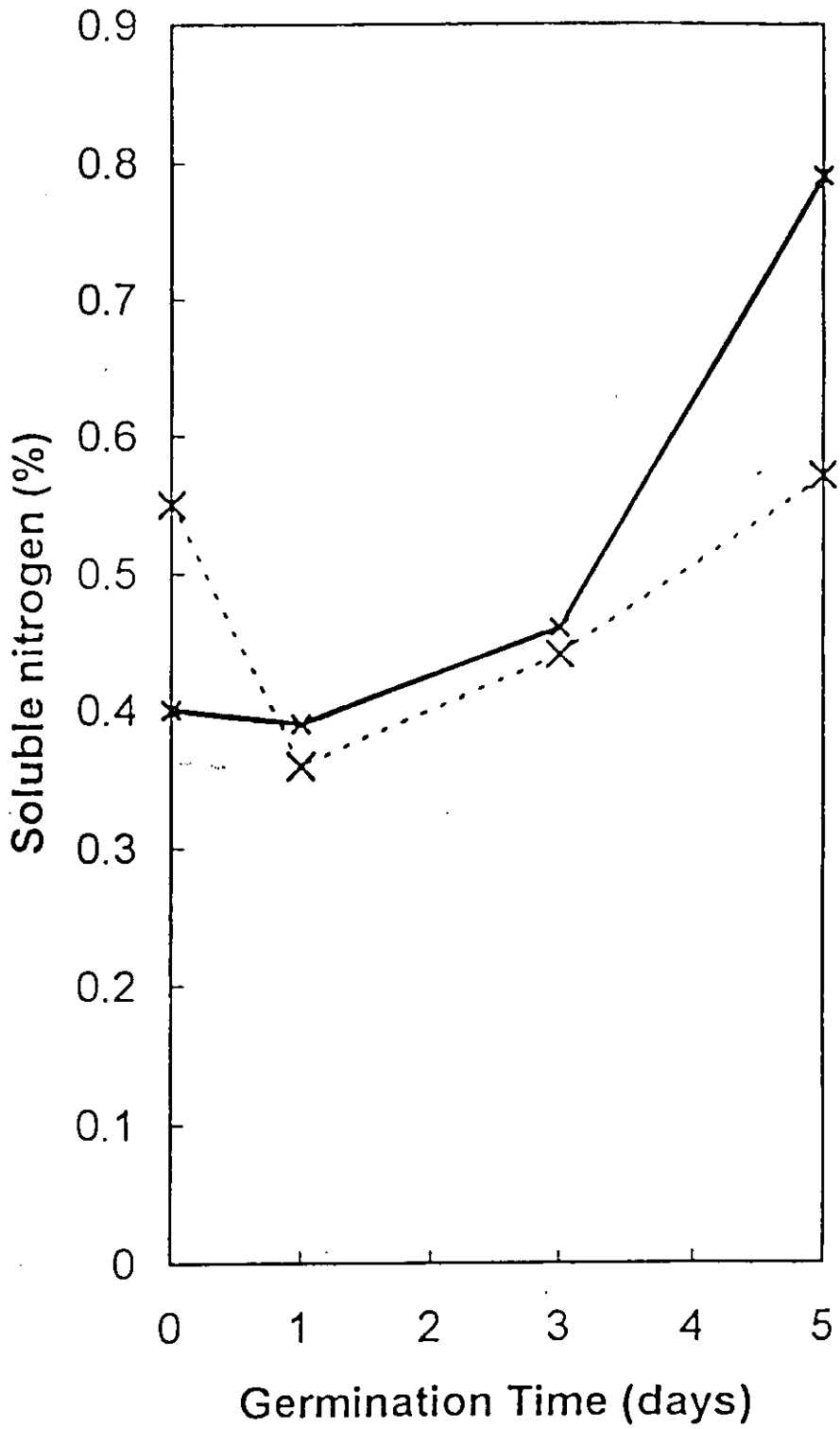


Figure 32.- Effects of germination time and variety on the percentage of soluble nitrogen of pearl millet (variety SDMV 89004(–) and variety SDMV 91018 (--))

NSI and soluble nitrogen were significantly affected ($p < 0.001$) by germination time and variety. Pearl millet grains had a NSI of 19.3% and 26.7% for varieties SDMV 89004 and SDMV 91018, respectively. NSI increased with germination time for both pearl millet varieties. However, non-germinated pearl millet grains of SDMV 91018 had higher NSI than grains germinated at 1 and 3 days. The highest NSI 39.8% was recorded at 5 days germination with variety SDMV 89004.

As germination time increased the amount of soluble nitrogen increased for both varieties. Non-germinated pearl millet grains had soluble nitrogen contents of 0.40% and 0.55% for varieties SDMV 89004 and SDMV 91018, respectively. In general, soluble nitrogen increased with germination time. Germinated pearl millet of SDMV 89004 variety had a higher percentage of soluble nitrogen than SDMV 91018. As with NSI, non-germinated pearl millet grains of SDMV 89004 variety had higher soluble nitrogen than that germinated for 1 and 3 days. The highest percentage of soluble nitrogen 0.79% was recorded at 5 days germination with variety SDMV 89004.

4.4.6 *In vitro* protein digestibility

The effects of germination time and variety on the *in vitro* protein digestibility of the two pearl millet varieties are shown in Figure 33.

In vitro protein digestibility was significantly affected ($p < 0.001$) by germination time and variety. Pearl millet grains had *in vitro* protein digestibilities of 68.8% and 58.5% for varieties SDMV 89004 and SDMV 91018, respectively. *In vitro* protein digestibility increased with germination time. Variety SDMV 89004 had higher *in vitro* protein digestibility than SDMV 91018. The highest percentage of *in vitro* protein digestibility 95.5% was recorded at 5 days germination with variety SDMV 89004.

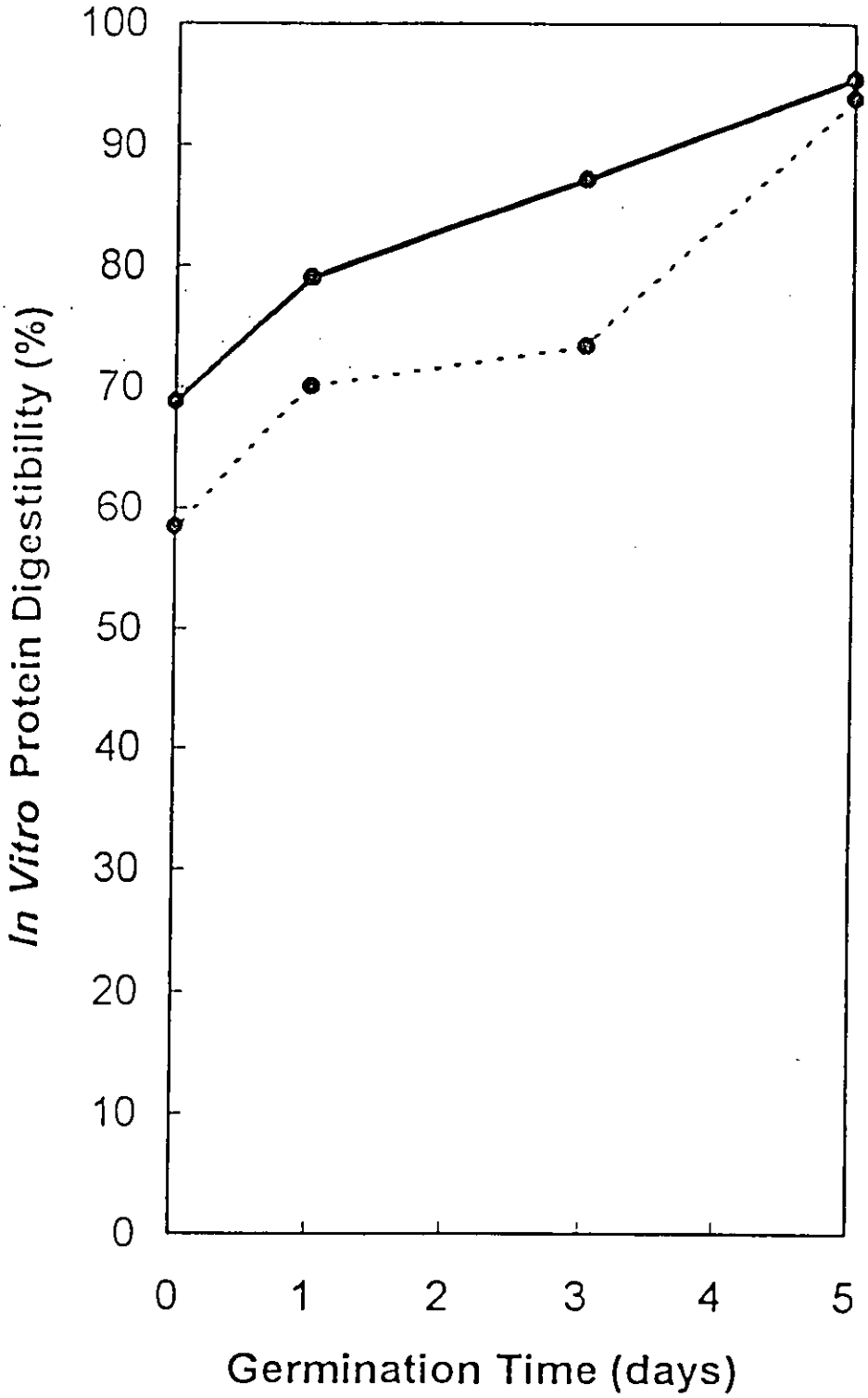


Figure 33.- Effects of germination time and variety on the *in vitro* protein digestibility of pearl millet (variety SDMV 89004(–) and variety SDMV 91018 (--))

4.4.7 Amino acid composition

The effects of germination time and variety on the amino acid composition of the protein of the two pearl millet varieties are shown in Table 11.

Generally, the non- and essential amino acids were significantly affected by germination time and variety. Lysine increased with germination time in the variety SDMV 91018. Variety SDMV 89004 showed a non expected decrease in lysine content with germination time.

In general, the levels of essential amino acid such as histidine, threonine, valine, isoleucine, leucine and phenylalanine showed little increase with germination time in both varieties, except that the levels of valine, isoleucine, leucine and isoleucine decreased on longer germination periods for the variety SDMV 89004. Methionine did not show any significant change for the variety SDMV 91018, but it decreased significantly for the variety SDMV 89004. Overall, the protein of pearl millet of variety SDMV 91018 showed a higher essential amino acid content than SDMV 89004.

In general, the non-essential amino acids contents decreased with germination time. However, non-essential amino acids, such as, glutamic acid, arginine and proline, increased during initial germination, but decreased with longer germination periods, except that, the level of aspartic acid increased with germination time for both varieties. Tyrosine, serine, glycine and alanine showed little changes with germination time. There was not a significant difference between varieties investigated.

The levels of essential amino acids in the proteins of non-germinated pearl millet in both varieties were generally lower than the FAO Scoring Pattern. However, pearl millet grains non-germinated and germinated for 1 day met the FAO Scoring Pattern for leucine. The lysine content of the protein of

TABLE 11.- Amino acid composition of the protein of the two pearl millet varieties (g/100 g protein)

Amino Acid	Control () [a]	Germination Time (Days) 1	3	5	FAO Scoring Pattern [b]
Essential amino acids					
Valine	(4.8-7.0)				5.0
SDMV 89004	3.6	3.7	3.5	2.9	
SDMV 91018	3.2	3.2	3.8	3.6	
Methionine	(1.5-2.9)				3.5
SDMV 89004	1.9	1.6	1.6	1.1	
SDMV 91018	1.5	1.5	1.6	1.5	
Isoleucine	(3.6-5.9)				4.0
SDMV 89004	2.2	2.2	2.0	1.5	
SDMV 91018	1.7	1.7	2.6	2.4	
Leucine	(8.0-25.1)				7.0
SDMV 89004	8.2	8.5	7.4	5.7	
SDMV 91018	7.7	7.5	8.6	8.1	
Phenylalanine	(4.4-5.6)				6.0
SDMV 89004	4.3	4.8	4.6	3.9	
SDMV 91018	4.3	4.3	4.6	4.5	
Lysine	(1.7-6.5)				5.5
SDMV 89004	3.3	3.2	2.9	2.9	
SDMV 91018	2.7	2.7	4.0	4.1	
Histidine	(1.8-2.6)				4.0
SDMV 89004	1.9	2.1	2.0	1.9	
SDMV 91018	1.9	2.0	2.2	2.2	
Threonine	(1.2-4.8)				4.0
SDMV 89004	4.2	4.7	4.3	4.0	
SDMV 91018	4.1	4.2	4.3	4.3	
Non-essential amino acids					
Aspartic acid	(4.9-10.3)				
SDMV 89004	8.2	8.9	9.9	10.3	
SDMV 91018	8.1	7.7	10.4	12.7	
Glutamic acid	(12.3-25.4)				
SDMV 89004	19.3	22.8	19.9	16.1	
SDMV 91018	19.8	20.1	18.2	17.0	
Serine	(3.7-5.6)				
SDMV 89004	6.6	7.1	6.6	6.0	
SDMV 91018	6.6	6.6	6.4	6.5	
Glycine	(2.8-5.8)				
SDMV 89004	4.3	4.1	3.5	3.6	
SDMV 91018	3.8	3.8	3.8	3.8	
Arginine	(3.2-8.1)				
SDMV 89004	6.0	6.0	5.3	4.9	
SDMV 91018	5.3	5.3	5.5	5.2	
Alanine	(7.5-10.5)				
SDMV 89004	8.7	9.1	8.7	8.0	
SDMV 91018	8.6	8.9	8.2	8.2	
Proline	(5.9-14.2)				
SDMV 89004	6.4	7.1	7.0	6.1	
SDMV 91018	6.4	6.4	6.4	6.3	
Tyrosine	(1.7-4.8)				
SDMV 89004	3.0	2.9	2.8	2.7	
SDMV 91018	2.5	2.5	3.5	3.4	

[a]Data in brackets are of pearl millet amino acids range from a review by Serna-Saldivar & Rooney (1995). [b]Food and Agriculture Organisation of the United Nations, according to Serna-Saldivar, McDonough & Rooney (1990) and Hoseney (1994).

germinated pearl millet of SDMV 91018 variety accounted for about 75.0 % of the FAO Scoring Pattern.

4.4.8 Phytic acid

The effects of germination time and variety on the phytic acid content of the two pearl millet are shown in Figure 34.

Phytic acid was significantly affected ($p < 0.001$) by germination time and variety. Non-germinated pearl millet grains had a phytic acid content of 0.24% and 0.27% for the varieties SDMV 89004 and SDMV 91018, respectively. Phytic acid decreased greatly with germination time in both pearl millet varieties investigated. Germinated pearl millet of SDMV 89004 variety had a lower percentage of phytic acid than SDMV 91018 throughout germination. The lowest percentage of phytic acid 0.024% was recorded at 5 days germination with variety SDMV 89004.

4.4.9 Pasting properties

The effects of germination time and variety on the pasting properties of the flours of the two pearl millet varieties are shown in Figures 35 and 36 for the variety SDMV 89004 and SDMV 91018, respectively.

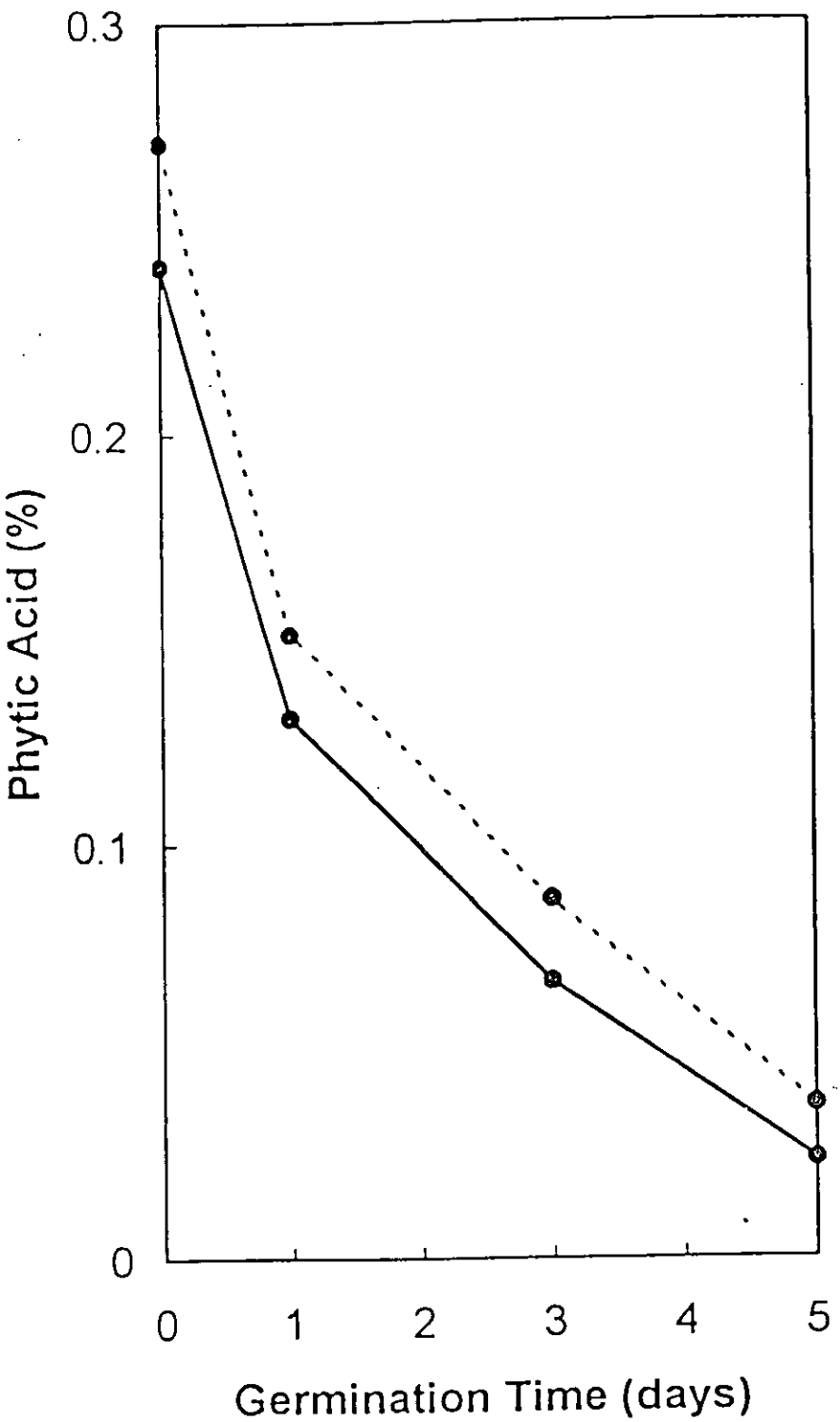


Figure 34.- Effects of germination time and variety on the phytic acid content of pearl millet (variety SDMV 89004(–) and variety SDMV 91018 (--))

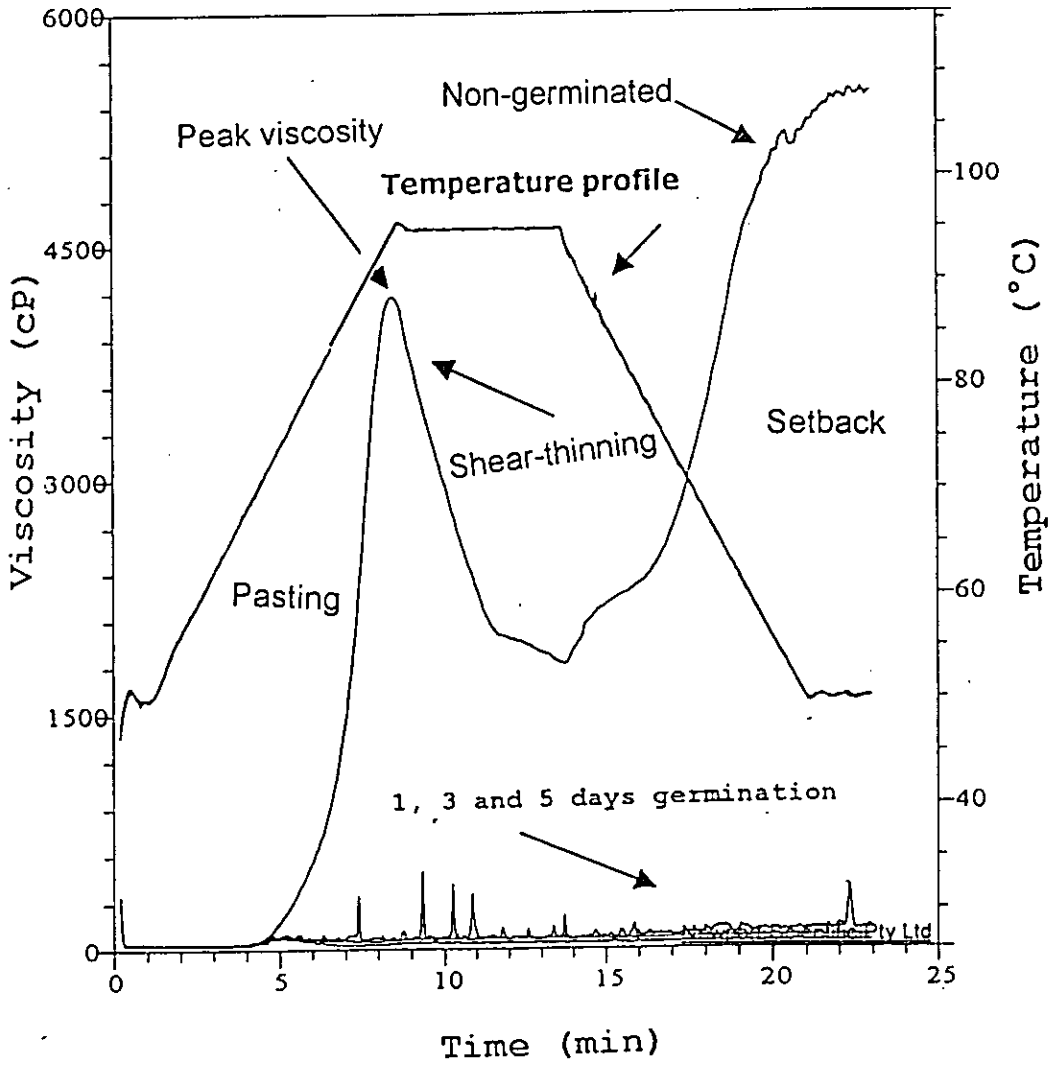


Figure 35.- Effect of germination time on pasting profiles of pearl millet flour of SDMV 89004 variety

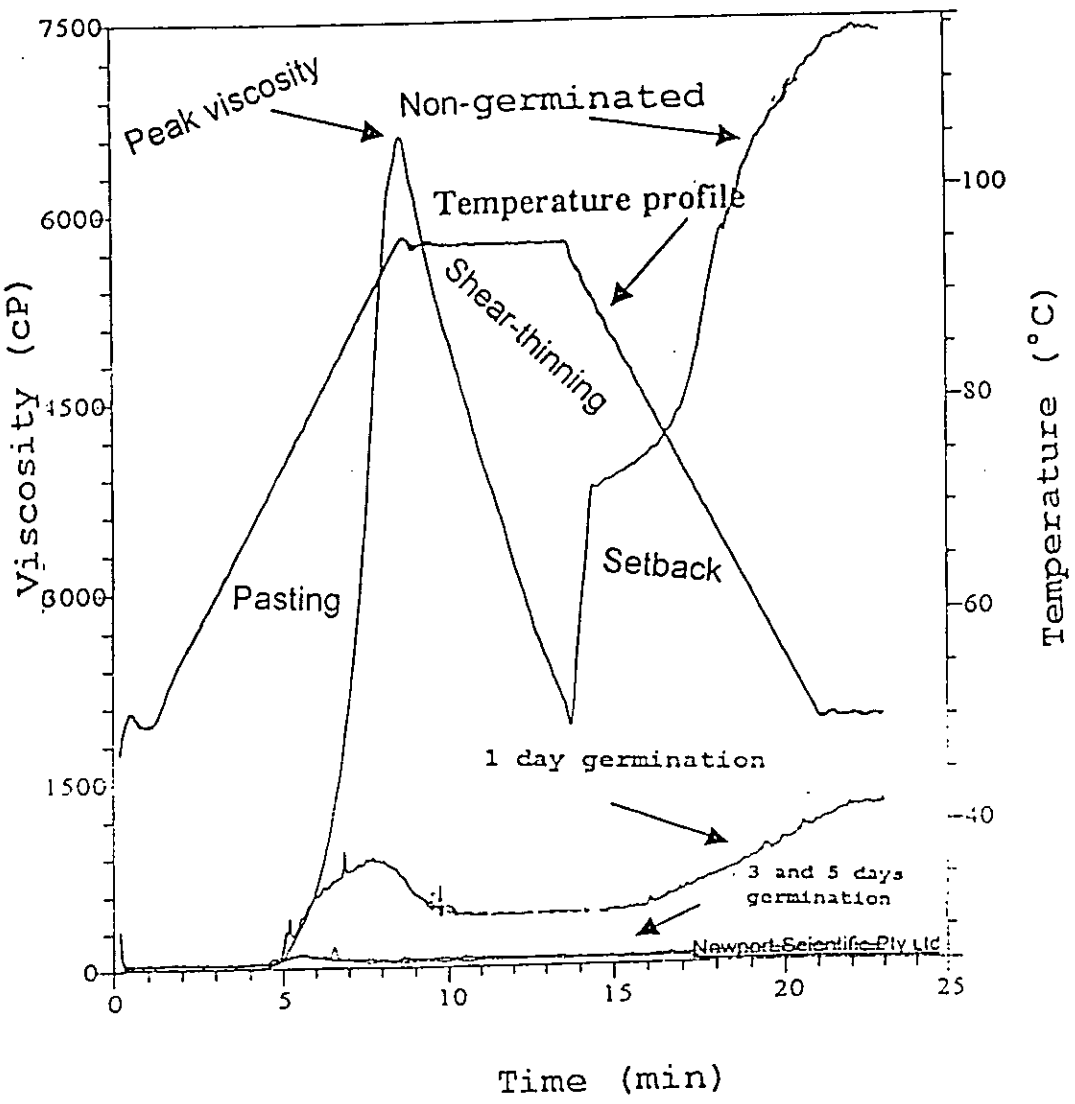


Figure 36.- Effect of germination time on pasting profiles of pearl millet flour of SDMV 91018 variety

Peak viscosity of pearl millet flour was significantly affected ($p < 0.001$) by germination time and variety. Peak viscosity of germinated pearl millet flour decreased dramatically with germination time for both varieties investigated. Peak viscosity of the malted pearl millet flours were very much lower than that of flour of grain in both varieties. Peak viscosity of pearl millet germinated for periods longer than 1 day was very low and the viscosities almost as low as water.

The effects of germination time and variety on peak viscosity, hot peak viscosity, cool peak viscosity and setback of the two pearl millet varieties are shown in Table 12. Grains and malts of the variety SDMV 91018 had significantly higher ($p < 0.001$) peak viscosity compared to SDMV 89004 (Table 12).

The peak viscosity, hot peak viscosity (ability of starch to withstand heating and shear stress), cool paste viscosity (final viscosity) and setback, a rapid increase in viscosity due to cooling of starch (cool paste viscosity – hot peak viscosity), decreased as the germination time increased. The lowest peak viscosity, hot peak viscosity, cool paste viscosity, setback and peak time 84, 36, 48, 12 (cP), and 4.9 min, respectively, were recorded at 5 days germination, with variety SDMV 89004. In general, both grain and malt of variety SDMV 89004 had lower hot peak viscosity, cool paste viscosity and set back as well as peak time compared to SDMV 91018.

4.4.10 Sensory evaluation of the mousy odour in pearl millet varieties

The Table 13 shows the results of the mousy odour evaluation by 12 trained panelists of the Department of Food Science of the University of Pretoria.

TABLE 13.- Sensory evaluation of mousy odour in non- and germinated pearl millet varieties

Panelists	Variety SDMV 89004				Variety SDMV 91018			
	Germination Time (days)							
	0	1	3	5	0	1	3	5
1	9	5	3	2	8	4	2	2
2	8	4	4	1	8	6	2	2
3	8	4	2	1	9	4	2	1
4	9	4	2	1	9	5	2	1
5	9	5	3	2	8	4	3	2
6	9	5	4	2	8	5	3	1
7	9	4	3	1	7	5	2	1
8	8	4	2	1	8	4	2	2
9	9	5	3	1	8	5	2	2
10	9	5	2	1	9	5	2	2
11	9	5	1	1	8	4	1	2
12	9	4	3	1	9	5	2	1
Mean	8.8^{a}	4.6^{b}	2.7^{c}	1.3^{d}	7.5^{a}	4.7^{b}	2.1^{c}	1.6^{d}

The level of intensity in the scale is 1– least intense and 9 – most intense mousy odour. Mean values with different letters in each variety are significantly different from each other ($p < 0.001$).

The mousy odour of pearl millet grains was significantly affected ($p < 0.001$) by germination time and variety. Non-germinated pearl millet grains had a rate of intensity of mousy odour of 8.8 and 7.5, on the scale of 1 least intense and 9 most intense mousy odour, for variety SDMV 89004 and SDMV 91018, respectively. Mousy odour decreased with germination time in both pearl millet varieties. Non-germinated pearl millet grains of SDMV 89004 variety had a significantly more intense mousy odour than SDMV 91018. However, in variety SDMV 91018 the reduction of the mousy odour due to malting was less than in SDMV 89004. The lowest level of intensity of mousy odour 1.3 was recorded at 5 days germination with variety SDMV 89004.

CHAPTER 5

DISCUSSION

In this discussion, the findings of this research on pearl millet malting will be compared primarily with those of research into sorghum and barley malting, since these are the major malting cereals. Additionally, an attempt will be made to explain the results in terms of what is known about the biochemistry of germination and malting of these cereals.

In this research, it was observed that the proportion of highly degraded starch granules decreased from the proximal to the distal end in malted pearl millet grain. Modification of horny endosperm was less intense compared to floury endosperm. Similar results, which showed that the degradative enzymes diffuse out from their origin in the scutellum, were found in wheat, rye, oats, maize (Okamoto, Kitano & Akazawa, 1980) and in sorghum (Glennie, Harris & Liebenberg, 1983; Glennie, 1984).

Unlike barley and sorghum, modification of endosperm structure during the germination of pearl millet has received little attention. Much of the information about modification during the process of malting is on barley; probably because barley is the most suitable cereal for malting (Briggs, 1972; Okamoto, Kitano & Akazawa, 1980; Palmer, 1980; MacGregor & Matsuo, 1982; Fretzdorff, Pomeranz & Bechtel, 1982; Briggs & MacDonald, 1983; Fincher & Stone, 1993); and to some extent on sorghum (Hoseney, Varriano-Marston & Dendy, 1981; Aisien, 1982; Glennie, Harris & Liebenberg, 1983; Glennie, 1984). During germination of pearl millet, α-amylase enzyme is known to preferentially attack the spherical granules instead of polygonal granules of the grain and that the starch hydrolysis is more vigorous at the centre of the grain than at the periphery (Hoseney, Varriano-Marston & Dendy, 1981).

In germinating barley, endosperm breakdown begins in the region adjacent to the scutellum and proceeds, roughly parallel to the scutellar epithelium, from the proximal to the distal end of the grain (Brown & Morris, 1890 according to Palmer, 1989; Gibbons, 1981; Ranki, 1990). Briggs & MacDonald (1983) also reported that barley modification begins beneath the scutellum and advances with a "front" roughly parallel to the scutellum "face". These results were taken to indicate that the enzymes that catalyse initial modification of the endosperm come from the scutellum. Later, Duffus (1987) stated that there was also probably sufficient evidence to suggest that in barley, modification begins under the aleurone cells at the acrospire end.

Another noteworthy finding of this study was that the degradation of starch granules appeared to be more intense than degradation of protein bodies. Glennie, Harris & Liebenberg (1983) also reported this for sorghum.

In this present study, it appears that the aleurone layer, cell walls and horny endosperm were not greatly involved in the modification process. This hypothesis is supported by the fact that these anatomical parts of pearl millet grain remained almost intact throughout the course of germination. The findings for pearl millet are contrary to that for barley, where the cell walls are degraded at a relatively early stage (one to two days), but the breakdown of starch granules is not in evidence until 3 to 4 days after modification begin (Palmer & Mackenzie, 1986; Duffus, 1987). The general pattern of modification in germinating barley was established as breakdown of cell walls, protein and starch hydrolysis (Fretzdorff, Pomeranz & Bechtel, 1982). Lewis & Young (1995) also stated that, in barley, the enzymes modify the endosperm cell walls, and the protein and starch granules contained therein, by partially hydrolysing them to release low-molecular weight products that can diffuse and/or be transported back to the scutellum, the cells of which elongate to aid absorption, and hence to the developing embryo. Since the starch and protein is combined within cells, the walls of these cells hydrolyse first. In sorghum,

the endosperm cell wall fraction was found to be more resistant to enzyme attack than the more soluble cell walls of barley; hence, sorghum endosperm cell walls were reported not to be readily degraded during germination (Glennie, 1984). MacGregor & Matsuo (1982) and Briggs (1998) believe that it is the germ which has a dominant role in the modification of barley, not the aleurone layer.

As stated in this study the endosperm cell walls of pearl millet did not show any significant changes due to germination. It is therefore suggested that the mechanisms by which the enzymes impart modification of the grain structure in pearl millet may be similar to that occurring in sorghum, where although the cell walls persist during germination, their chemical composition and solubility patterns change (Glennie, 1984). The fact that the degradation of starch and protein reserves occurred without degradation of cell walls was also observed in sorghum and wheat malts by Glennie, Harris & Liebenberg (1983) and Palmer (1989), respectively. It was suggested that localised "portals" exist in the normal endosperm cell walls of these cereals, through which proteolytic and amylolytic enzymes migrate during malting. This is contrary to what happens in barley where presumably, cell wall disruption as well as protein matrix degradation are required before starch hydrolysis can take place (Slack, Baxter & Wainwright, 1979).

It appears therefore that from all pearl millet grain structure modification observed in this research, the enzymes attack started from the germ, or more precisely from the scutellum, rather than from the aleurone layer. This suggests that pearl millet grain modification is controlled by the scutellum. Thus, it appears that pearl millet is the same as sorghum (Koeher, 1981; Aisien, 1982; Glennie, Harris & Liebenberg, 1983; Dufour, Mélotte & Srebrnik, 1992) in these respects.

In barley, MacGregor & Matsuo (1982) reported, using scanning electron microscopy, that starch degradation starts near the ventral crease and moves along the endosperm-embryo junction to the dorsal edge of the kernel. These observations are at variance with other reported data. However, they were interpreted to mean that the site of initial α-amylase synthesis is not the aleurone layer but the embryo.

The fact that the findings of this research on pearl millet grain structure modification during germination are the same as that of sorghum is consistent with their structural similarity and the fact that these grains are both tropical cereals.

The objectives of steeping are to clean the grain and to hydrate it to the correct extent and to prepare it so that it grows steadily and modifies uniformly during the germination stage (Briggs, 1998). The steeping conditions used in this research were a steeping time of 8 h, with a cycle of 2 h wet, 2 h dry air rest, at four different temperatures, 20 °, 25 °, 30 ° and 35 °C. This steeping time was found to be reasonable and was based on the fact that in the germinability tests, both pearl millet varieties started showing signs of germination after 8 to 10 h. Opoku, Osagie & Ekperigin (1983) showed that pearl millet germinates relatively less rapidly, which was indicated by the appearance of the radicle within the 15-18 h. The difference in the results of this research and that of Opoku, Osagie & Ekperigin (1983) is probably related to the variety differences. Similar results were found by Sheorain & Wagle (1973) who reported that pearl millet varieties began to show evidence of germination (chitting) after 7 or 8 h, while in the case of barley varieties, these authors found that it only occurred after about 24 h. The temperature of steeping was chosen on the basis that it was within the temperature range used by other authors working with tropical cereals: Malleshi & Desikachar (1986c) and Nout & Davies (1982) (finger millet); Gomez, Obilana, Martin, Madzvamuse & Monyo (1997); Muoria & Bechtel (1998) (pearl millet and

sorghum); Morrall, Boyd, Taylor & Van de Walt (1986) and Dewar, Taylor & Berjak (1997b) (sorghum).

In order to germinate, the grain must absorb water during steeping. Hence, the germ becomes active and makes use of the oxygen dissolved in the steeping water. Steeping the pearl millet grain for 8 h resulted in a reasonable percentage of water uptake, compared to sorghum. There was not much difference in the percentage of water uptake between 8 and 10 h steeping time.

Water uptake represents the amount of water taken up by the grain during steeping. This is crucial since during steeping physical and biochemical changes take place. Such changes include swelling of grains and an increase in respiratory activity. The small size of the pearl millet grains may be responsible for the rapid increase in the levels of moisture and water uptake. Small kernels have a proportionally larger surface area than larger ones.

The fact that pearl millet of variety SDMV 89004 had a higher water uptake in steeping than SDMV 91018 may be related to the texture of the endosperm. Variety SDMV 89004 had a softer endosperm than SDMV 91018. Softer endosperm texture can be more easily penetrated by water during the steeping process of the grain (Bamforth & Barclay, 1993). Bamforth & Barclay (1993) also stated that water uptake into the starchy endosperm is critical before the food reserves of that tissue can be mobilised through the action of enzymes. Water uptake by grains is complex and is regulated by the porosity of the grain surface layers, the temperature, the osmotic driving force and related properties, the ease of spreading through the grain tissues and the resistance of the grain to swelling (Briggs, 1998). It is widely agreed that

grains which hydrate quickly malt better than those which hydrate more slowly (Bamforth & Barclay, 1993; Briggs, 1998). Similarly, pearl millet of SDMV 89004 variety which had slightly higher water uptake, generally had higher malt quality than SDMV 91018. According to Briggs (1998) some grains take up water more rapidly than others because of a higher osmotic pressure in the hydrated grain and/or a less dense (soft) structure that permits the grain to swell to accommodate the water taken up.

The finding that in both pearl millet varieties the green malt moisture content increased as the amount of water applied in the different watering treatments increased is of crucial importance, since steep-out moisture strongly influences the rate and extent of modification (Lewis & Young, 1995; Dewar, Taylor & Berjak, 1997b). At steep-out the mean moisture content of the grain was about 27% for both varieties. The reason for low steep-out moisture may be related to the size of pearl millet germ and the fact that pearl millet germ is rich in fat. Working with finger millet, Malleshi & Desikashar (1986c) reported steep-out moisture contents of 30-35%. By 5 days of malting the mean moisture contents were 33.2, 48.0 and 60%, for the variety SDMV 89004 and 35.3, 47.3 and 56.3%, for variety SDMV 91018, with the low, medium and high watering treatment, respectively. These results are relatively lower than those of Morrall, Boyd, Taylor & Van der Walt (1986) and Dewar, Taylor & Berjak (1997b) working with sorghum. The reason for this can be the difference in the steeping time used. These authors steeped sorghum with a cycle of 3 h wet, 1 h dry, for 16 and 24 h, respectively. In this research, pearl millet was steeped for 8 h with a cycle of 2 h wet and 2 h dry rest. These conditions were chosen because of the size of the pearl millet grains, and the fact that pearl millet grains started chitting between 8 and 10 h. However, future work should be done to investigate steeping for longer periods of time as was done for sorghum by Dewar (1997).

When malting tropical cereals, the grains are liberally watered during germination and this watering compensates for the relatively low moisture content of the grain after the short steeping time (Novellie, 1962a; Morrall, Boyd, Taylor & Van der Walt, 1986; Dewar, Taylor & Berjak, 1997b). Both pearl millet varieties examined germinated at lower moisture contents compared with barley and sorghum. Bamforth & Barclay (1993) stated that, non-barley grains will germinate below approximately 30% moisture, whereas the most intransigent batches may require a final moisture approaching 50 % to achieve uniform germination and that most barleys require a steeping regime that takes them to 42-46% moisture.

In order to produce malts of good and consistent quality, a pre-requisite is that a good grain for malting must show vigorous and uniform growth and must have a high percentage of germination. To be considered "ready for malting" maltsters would expect sorghum grain to germinate at least 50% by day 2 and 95-100% by day 3 in the Germinative Energy test (Dewar, Taylor & Joustra, 1995). It is also very important that such grain has a good potential for the production of enzymes under the correct malting conditions, particularly in sorghum beer brewing, where the malt generally has to act on at least twice its own weight of starchy adjunct (Novellie, 1966; Dewar, 1997).

Maltsters use the growth of the rootlets and acrospires (shoots) as indicators of the progress of malting. The root and shoot growth reported in this research, 10.4% (of total weight), is higher than that of finger millet, 5.1%, germinated for 5 days at 25 °C, found by Nout & Davies (1982) and lower than that found by Dewar, Taylor & Berjak (1997b), working with sorghum. These latter authors found roots and shoots values around 12 % in 4 days germination at 25 °C. The lower root and shoot growth observed with pearl millet malts in this research compared with that of sorghum, by Dewar, Taylor & Berjak (1997b), may be a reflection of the relatively lower DP levels found in pearl millet, compared to that of sorghum. Dewar, Taylor & Berjak

(1997b) found a DP of 45 SDU/g dry malt, for sorghum; while in this research the highest DP found at 30 °C, high watering, was 34.6 PMDU/g dry malt.

Reports on pearl millet and sorghum root and shoot growth in the literature are presented in the way that they cannot be directly compared with the results of this research, since authors Malleshi & Desikachar (1986c) only reported how the growth of root and shoot influenced the sorghum, finger and pearl millets malting loss. And in some cases the results of root and shoots of sorghum malt are reported in terms of length, as by Okolo & Ezeogu (1996).

In barley, rootlets are reported to contain on a dry weight basis non-protein extract 35-40 %, proteinaceous compounds 20-35 %, fatty materials and also vitamins A, B, D and E depending on the malt (Moll & Blauwe, 1991). The roots must be removed since they contribute undesirable substances to the wort (bitterness, too intense coloration of the wort, etc.) (Moll & Blauwe, 1991). However, in sorghum beer brewing roots and shoots are not removed, particularly because they are a good source of FAN in the wort (Dewar, Taylor & Berjak, 1997a).

Unlike malted pearl millet, ungerminated pearl millet grains of both varieties did not exhibited any DP. Diastatic Power, which is a measure of the joint activity of α- and β-amylase, is the single most important indicator of malt quality for sorghum beer brewing (Novellie, 1962a; Dufour, Mélotte & Srebrnik, 1992; Dewar, Taylor & Berjak, 1997b).

The rate of increase in malt DP, which was observed at 25 °C, high watering treatment, declined over longer periods of germination. This negative effect of the high watering treatment on DP of pearl millet malts during the last days of germination observed in this investigation was also observed for sorghum by Novellie (1962a; 1962b); Morrall, Boyd, Taylor & Van der Walt (1986) and Dewar, Taylor & Berjak (1997b). The reason for the decline in the rate of

increase of DP under high watering treatments can be the fact that higher moisture possibly promotes high amylase activity and hence rapid denaturation. This is supported by the fact that contrarily high moisture continued to have a beneficial effect on free α-amino nitrogen (FAN), the products of protease activity (Dewar, Taylor & Berjak, 1997b), late in germination. Dewar, Joustra & Taylor (1993) stated that for sorghum beer brewing a minimum DP of 28 SDU/g is required for sorghum malt. Hence, with respect to DP pearl millet malt, germinated for 4 and 5 days for variety SDMV 89004 and SDMV 91018, respectively, are suitable for sorghum beer brewing. The DP values for pearl millet malt found in this research are similar to those found for sorghum and millets by other authors (Novellie, 1959, 1962a, 1962b, working with sorghum; Morrall, Boyd, Taylor & Van der Walt, 1986, working with sorghum; Gomez, Obilana, Martin & Madzvamuse, 1997, working with sorghum and pearl millet), but lower than those found by Dewar, Taylor & Berjak (1997b) also working with sorghum.

The fact that pearl millet malts of SDMV 89004 variety, germinated with medium watering treatment, gave consistently higher α- (measured by inactivation of β-amylase) and β-amylase activity (measured by inactivation of α-amylase) compared with SDMV 91018 could be related to the former's higher germinability, which is a reflection of higher metabolic activity. Although these α- and β-amylase activities are similar to sorghum malt, they are lower than that of barley malt reported by Taylor & Robbins (1993) in a direct comparative study with sorghum malt.

The optimum temperature for maximum α-amylase activity found in pearl millet, between 25-30 °C, for both varieties is in agreement with the findings of other authors working with sorghum (Novellie, 1959, 1962a, 1962b; Nout & Davies, 1982; Pathirana, Shivayogasundaram & Jayatissa, 1983; Morrall, Boyd, Taylor & Van der Walt, 1986; Nzelibe & Nwasike, 1995; Dewar, Taylor & Berjak, 1997b), with finger millet and sorghum (Nout & Davies, 1982) and

with sorghum, fonio (*Digitaria exilis*) and pearl millets (Nzelibe & Nwasike, 1995). On this basis and that of their own data Muoria & Bechtel (1998) suggested that a germination temperature > 22 °C would be more desirable for sorghum and pearl millet, than barley, in order to obtain higher values of α-amylase. However, the results of this research showed that, in order to produce pearl millet malt of good quality, the temperature of germination should be ≥ 25 °C. Germination temperature of 22 °C is probably slightly too low for pearl millet malting.

A difference between the tropical (sorghum and millets) and temperate climate (barley) cereals is the fact that cereals indigenous to the tropical and sub-tropical areas of the world have no more than traces of β-amylase (Novellie & De Schaepdrijver, 1986; Dufour, Mélotte & Srebrnik, 1992; Taylor & Robbins, 1993; reviewed by Zeigler, 1999). However, both types of cereals have little α-amylase in the ungerminated grains. Germination of tropical cereals leads to the production of both amylases with α-amylase predominating (Dyer & Novellie, 1966; Novellie & De Schaepdrijver, 1986; Dufour, Mélotte & Srebrnik, 1992; Lewis & Young, 1995; Palmer, 1986). Ungerminated cereals from the more temperate zones have moderate amounts of β-amylase, but little α-amylase (Novellie & De Schaepdrijver, 1986; Dufour, Mélotte & Srebrnik, 1992). On germination, α-amylase is formed and β-amylase is synthesized during temperate cereal (barley) development but is rendered fully active during germination (MacGregor, Gordon, Meredith & Lacroix, 1972; MacGregor & Lenoir, 1987; reviewed by Palmer, 1989; MacGregor, 1996).

Cereal β-amylase plays a crucial role during brewing. As a contributor to the diastatic power of malt, its activity is essential for the generation of maltose, the most abundant fermentable carbohydrate in wort, and other easily fermentable sugars from cereal grain starch in the mashing process to fuel the production of alcohol by yeast (MacGregor, 1996; Zeigler, 1999).

In this research, soluble β-amylase activity (Betamyl assay) of pearl millet malts and the sorghum malt standard showed an effect of the reducing agent (cysteine) used in the assay. These results differ from that for sorghum found by Taylor & Robbins (1993), where there was no reducing agent effect. In the assay used by Taylor & Robbins (1993), the enzyme extraction was carried out using the reducing agent mercaptoethanol. The difference in results may have been due to the fact that the use of cysteine, as a reducing agent in the measurement of both soluble and total β-amylase in cereal grains ensures effective extraction and maximum stability of the extracted enzyme (McCleary & Codd, 1989).

Free α-amino nitrogen (FAN) is the second most important indicator of malt quality for sorghum beer brewing, because it is a source of nitrogen for yeast metabolism during fermentation (Daiber & Novellie, 1968; Pickerell, 1986, 1987). Low malt FAN content therefore would result in decreased fermentation, and hence low alcohol beer. During malting, some modification of the proteinaceous matrix is also necessary to make starch more easily degraded in mashing, as well as to form low-molecular weight nitrogenous compounds, especially amino acids (Lewis & Young, 1995). The latter will support the growth of the embryo and also of the yeast during fermentation.

The highest level 199 mg/100 g malt FAN was observed with pearl millet malt of SDMV 91018 variety, germinated at 35 °C for 5 days, medium watering treatment. This may be related to the fact that this variety had the higher percentage of roots and shoots 10.4%, which are a good source of malt FAN. Dewar, Taylor & Berjak (1997b), working with sorghum, reported that although roots and shoots represent only a relatively small proportion of the total weight of sorghum malt, their contribution to the total malt FAN was as high as 62%. During the germination process the increase in the amount of FAN in roots and shoots is a result of translocation of the products of storage protein breakdown from the kernel (Taylor, 1983).

A typical FAN specification for sorghum malt for sorghum beer brewing would be a minimum of 110 mg/100 g malt (Dewar, Joustra & Taylor, 1993). Hence, pearl millet malt is suitable for sorghum beer brewing with respect to FAN.

In cereals, FAN development may vary among varieties, probably because of differences in major enzyme characteristics and rate of protein metabolism during malting, as well as variations in grain protein structure and degradability, amino acid and peptide transport processes (reviewed by Owuama, 1999). It would be expected that the two pearl millet varieties investigated would show differences in malt FAN, since the endosperm texture as well as the protein content and Nitrogen Solubility Index of the two varieties were different. Variety SDMV 89004 had softer endosperm, higher protein content and higher Nitrogen Solubility Index compared to SDMV 91018.

Since the levels of nitrogenous substances in malt are not always consistent with the proteolytic activities, this may suggest the involvement of other factors than proteolysis, which influence protein modification during cereal germination. High solubility of nitrogenous substances may lead to low proteolytic activity or vice-versa (Okolo & Ezeogu, 1996).

Unlike DP, germination at the high watering treatment gave continuously higher malt FAN for both pearl millet varieties investigated. High moisture treatment promotes the protease activity of the germinating grain. The increase in malt FAN with germination time was also found in sorghum (Nout & Davies, 1982; Morrall, Boyd, Taylor & Van der Walt, 1986; Evans & Taylor, 1990; Dewar, Taylor & Berjak, 1997b) and in finger millet (Nout & Davies, 1982). The increase in malt FAN with germination time has to do with the fact that during germination, cereal proteins, mainly insoluble storage proteins in the endosperm, are converted into soluble proteins, peptides and amino acids, by the process of transamination (change of one amino acid into another one), which may have to supply nutrients to the developing embryo,

by the action of proteolytic enzymes (MacGregor, 1996). The FAN content of malt and subsequently of the wort depends on the proteolytic activity of the peptidases and proteinases (proteases) of the malt. The FAN content of wort would also depend on the mashing conditions applied (Taylor & Boyd, 1986).

Pearl millet malts had higher FAN in the grains germinated at high watering treatment, while medium and low watering treatment gave progressively lower malt FAN. In sorghum, Morrall, Boyd, Taylor & Van der Walt (1986) and Dewar, Taylor & Berjak (1997b) also found similar results. This could be due to the fact that high watering treatments favour root and shoot growth, hence the increase in FAN levels.

Agu & Palmer (1996), working with sorghum, also reported that the quantities of nitrogen transferred from endosperm to embryos increase with time and temperature of germination. The fact that DP, α- and β-amylases and FAN increased with an increase of germination time and temperature in malts of both pearl millet varieties investigated suggests that the effect was due to increased amylase and protease activity in the malt.

Malt extract is particularly important in lager beer brewing since it gives an estimation of how much of the malt will solubilise during mashing in the brewing process. In fact, the essence of mashing is to physically and enzymatically solubilise the malt. Hot water extract gives also an indication of the modification of the malt during the malting process. Fermentable sugars dominate the composition of malt wort. The maximum fermentability of wort that can be produced by malt enzymes is 75-78 %. This is mostly made up of maltose and maltotriose, which are result of α- and β-amylase acting together. The non-fermentable fraction of wort is at least 20% of the total extract. Most wort dextrins are branched molecules and contain the α-1-6 link of starch which malt amylases cannot attack (Lewis & Young, 1995).

The highest malt extract of about 70 % was found after 5 days of germination at high watering treatment. The increase in pearl millet malt hot water extract with germination time observed in this research is an indication of the progress of modification (breakdown of the endosperm reserves by amylase and protease activity) of the malt during germination. High watering treatments may have facilitated the solubilisation of the solids of pearl millet malts. An increase in malt extract with germination time was also found, in pearl millet, by Nzelibe & Nwasike (1995); in finger millet, by Malleshi & Desikachar (1986c) and Nout & Davies (1982) and in sorghum, by Morrall, Boyd, Taylor & Van der Walt (1986). These last workers also found that extract increased with watering treatment level.

The difference in the level of malt extract between the two pearl millet varieties investigated as well as with the sorghum and barley standards used are related to the degree of modification of the malts of the cereals used during the malting process, since the action of α- and β-amylase enzymes in the degradation of starch is very important during mashing. Pearl millet malts had higher malt extracts than the sorghum malt standard, but generally lower than the barley malt standard. The difference in malt extract is also related to the degree of solubility in water of the resulting products of the hydrolysed malts.

It would also appears that although it is heat (gelatinisation) that renders starch soluble in mashing to form extract, it is often assumed that α-amylase action helps starch solubilisation. This implies in practical terms that α-amylase is the extract-producing enzyme (Lewis & Young, 1995).

Efficient starch conversion does not occur during mashing until the gelatinisation temperatures of the starch granules which are being mashed are reached (Palmer, 1986). Therefore, the temperature (60 °C) at which the hot water extract assay is performed may be another reason for the

differences in hot water extract of the three cereals. Barley starch gelatinises and is completely solubilised in well-made malts during mashing for determination of hot water extract. The temperature of gelatinisation of cereals depends on the source of starch but for barley's is mostly between 65 and 75 °C (Lewis & Young, 1995). However, the starch of pearl millet and sorghum does not begin to gelatinise until substantially higher temperatures are reached (Palmer, 1989; Briggs, 1998). In pearl millet and sorghum, if the temperatures of mashes are raised much over 65 °C, enzyme destruction is rapid and so starch conversion is incomplete and recovered extracts are low (Briggs, 1998). At elevated mash temperatures the fermentability of the wort is low because the β-amylase is rapidly denatured. Beta-amylase works best at 55-60 °C and α-amylase about 10-15 °C higher (Taylor, 1992; Lewis & Young, 1995).

Mashing below 65 °C increases the fermentability of the wort; mashing above 65 °C reduces fermentability and increases molecular sizes of wort dextrins. This alteration to the sugar spectra of wort is understandable because the starch extract-releasing and liquefying enzyme α-amylase is more heat stable than the sugar (maltose) releasing enzyme β-amylase (Palmer, 1989). Beer filtration problems reported in sorghum brewing by Aisien & Muts (1987) may reflect the possibility that during malting localised "breakdown" of the endosperm cell walls may expose portals through which proteases and amylases migrate. This is substantiated by the observation that cell walls do not degrade, like in barley, during malting (reviewed by Palmer, 1989). Incomplete starch conversion is desired for the production of opaque beers. In contrast, total starch conversion is highly desirable for the production of conventional larger beers.

Although the highest hot water extract 69.0 % found is lower than 75-80%; a value considered suitable for conventional beer brewing (Bamforth & Barclay, 1993; Briggs, 1998); the fact that it was, in some cases higher than that of barley malt standard, 59.4 %, and also higher than some reported values for

extracts for barley malt (Briggs, Hough, Stevens & Young, 1981; Bamforth & Barclay, 1993; Briggs, 1998), together with the fact that malting considerably reduced the pearl millet fat content, is *per se* a good indication for the potential use of pearl millet malt in conventional beer brewing. Pearl millet is very rich in fat, which could lead to rancidity problems as well as poor foam head retention during the brewing process, hence affecting negatively the organoleptic properties of the beer produced (Zeurcher, 1971). In this research, the levels to which pearl millet fat content was reduced by malting are almost within the range (2-3 %) as it occurs in barley (Briggs, 1998). Moll & Blauwe (1991) suggested that in order to reduce the lipid content of pearl millet malt in brewing it is recommend to add sulphuric acid during mashing. However, no scientific explanation was given concerning the mechanism by which sulphuric acid reduces the lipid content of pearl millet malts during the mashing process of brewing.

A possible method to improve the extract content was proposed by Nout & Davies (1982) working with finger millet. These authors suggested that a small addition of barley malt could be used. This could simultaneously increase the β-amylase activity in the mash. Later, Taylor & Daiber (1988) suggested that the increased α-amylase stability afforded by calcium conditions, could improve extract in sorghum mashes.

One of the objectives of the malting industry is to operate as economically as possible, using a rapid process with a minimal malting loss (Moll & Blauwe, 1991). The increase in malting loss with the germination time, temperature and watering treatment observed in this research can be attributed to the respiratory activity which takes place on germination. A direct relation between germination time and malting loss was also found in sorghum (Novellie, 1962a; Pathirana, Shivayogasundaram & Jayatissa, 1983; Morrall, Boyd, Taylor & Van de Walt, 1986; reviewed by Owuama, 1999) and in barley (reviewed by Briggs, Hough, Stevens & Young, 1981; reviewed by Bamforth & Barclay, 1993; reviewed by Briggs, 1998).

Potassium bromate has been used to reduce malting loss (Nout & Davies, 1982; Agu & Okeke, 1991; 1992; Agu & Ezeanalue, 1993; Nzelibe & Nswasike, 1995). However, the safety of the use of potassium bromate has not been clarified.

In this investigation, the highest malting loss, around 12 %, was recorded with variety SDMV 91018, at 5 days germination, medium watering treatment. Similar results were reported for sorghum (Novellie, 1962a, 1962b) and finger millet (Nout & Davies, 1982). However, Morrall, Boyd, Taylor & Van de Walt (1986) reported relatively higher losses in sorghum. The high malting losses reported by these authors may be related to the high respiratory activity observed in sorghum and the long steeping time used. These authors steeped sorghum for 16 h, while in this research pearl millet grains were steeped for 8 h only.

In barley, the total malting losses during malting are usually in the range 6-12% of the original dry weight (Briggs, 1998). However, barley malting loss results presented in the literature are not directly comparable to pearl millet results of this investigation since they do not include the roots and shoots.

From now on, in this discussion, changes brought about by germination which have a broader influence on pearl millet's food value than just brewing will be considered. These changes are mainly in nutritional and functional properties.

Carbohydrates are the single most important source of food energy in the world. They comprise some 40 to 80% of total food energy intake, depending on locale, cultural considerations or economic status (FAO/WHO, 1997). The reduction in the total carbohydrate content of the pearl millet varieties, from 78.4 to 53.8% and from 75.3 to 54.5% for the variety SDMV 89004 and SDMV 91018, respectively, and increase in the percentage of the TCES, from 10.6 to 13.5% for the variety SDMV 89004 and from 10.1 to 11.0% for the variety

SDMV 91018, during germination, can be attributed to the fact that some of the endosperm starch is consumed during germination to provide energy.

In this research, pearl millet carbohydrates decreased about 10 % in only 48 h. Opoku, Ohenhen & Ejiofor (1981) found lower decrease in the level of carbohydrates, which decreased about 8% after malting pearl millet at 25 °C for 48 h. Working with finger millet, Malleshi & Desikachar (1986c) reported that germination for 48 h resulted in about 5% loss in starch content and that the continuation of germination up to 96 h resulted in about 10% loss in starch content. Sripriya, Antony & Chandra (1997) found that starch content of finger millet decreased by about 12 % on germination for 24 h at 30 °C. In contrast, the starch content of barley is reduced by only about 10% during malting (Bathgate & Palmer, 1972; Lewis & Young, 1995). The difference between the results of this investigation and that of other authors can be attributed to the differences in the amylase activity of barley, pearl and finger millets varieties used.

The fact that the reduction in carbohydrate content was higher in variety SDMV 89004 than SDMV 91018 could be related to its higher germinability. Variety SDMV 89004 had higher DP and higher carbohydrate content. This also indicates that the carbohydrate content of pearl millet was affected by the grain variety and as well as by the respiratory activity of the grains.

The reduction of carbohydrate during germination observed in this research would contribute to a decrease in the energy value of food products prepared from germinated flours. In adults, it is important that the amount of energy ingested be matched to the amount of energy expended. However, this decrease is compensated by the fact that carbohydrates of malted pearl millet are more soluble than that of non-germinated grains. In infants the amount of energy ingested is more than they expend since they use the rest of the energy to build up their bodies (Mosha, 1985). Carbohydrates exert a protein-

sparing effect. When carbohydrates are depleted in the animal body and the animal needs additional energy, it gets this energy by oxidizing fats and proteins (Potter & Hotchkiss, 1995). However, if carbohydrates are supplied, the body oxidizes them for energy in preference to protein and, thus, the protein spared. It is crucial that the reduction of the level of carbohydrates (starch) should not be very high if the malts are meant for the preparation of traditional southern African food products, such as opaque beers, porridges, and traditional unleavened pancakes, called *makati* in Mozambique, as well as weaning foods for infants called *nthlatu* in the south of Mozambique, where minimum carbohydrate reduction may be advantageous.

The increase in the TCES with germination time observed in this investigation was also found by Khetarpaul & Chauhan (1990b). These authors found that the *in vitro* starch digestibility increased by more than three-fold when pearl millet grains were germinated for 24 h. Improvements in starch digestibility of pearl millet through germination has also been reported by Khetarpaul & Chauhan (1990a); Chaturvedi & Sarojini (1996) and Pawar & Pawar (1997).

The changes in the susceptibility of starch to enzyme attack which took place during the germination of pearl millet are advantageous in respect of producing a product with improved nutritional quality, which can be used as an ingredient in various food products, particularly weaning foods for infants.

The non-significant difference between the percentage of the TCES of the two pearl millet varieties before they were germinated can be explained by the fact that the ratio of amylose/amylopectin in both varieties was also not significantly different. A difference may have been expected if the ratio had been different. Amylopectin is a highly branched chain biopolymer which is more susceptible to enzyme attack than amylose. Its branched chains give water the ability to penetrate the structure more easily than that of amylose and to gelatinise the starch (Rebar, Fishbach, Apostolopoulos & Kokini, 1984).

The lower phytic acid content in pearl millet of variety SDMV 89004 may be the reason for the higher percentage of the TCES observed in the pearl millet malts of variety SDMV 89004 compared to SDMV 91018. Phytic acid may decrease starch digestibility by binding with calcium which is known to be necessary for α-amylase activity (Yoon, Thompson & Jenkins, 1983).

The Water Absorption Index (WAI), which is a reflection of the amount of pearl millet starch dispersed in excess water, was not significantly different between the two pearl millet varieties. This may be attributed to the fact that both pearl millet varieties had similar starch amylose contents. The decrease in the Water Absorption Index and the increase in the Water Solubility Index (WSI) with germination time for both varieties may have to do with the fact that during the germination process the carbohydrate content decreased as a result of hydrolysis by the amylase enzymes. The increase in WSI with germination is of significance since it gives an indication that germination can be used to increase the amount of soluble materials, such as starch and amino acids, which can be easily digestible.

Fat content was reduced from 6.4% to 3.4% and to 3.1% for the varieties SDMV 89004 and SDMV 91018, respectively, after only 5 days of germination. During germination, lipids are not metabolised as fast as other food reserves like carbohydrates and proteins (Opoku, Ohenhen & Ejiofor, 1981). Other authors also reported that there is a reduction in lipid (fat) content in finger and pearl millets during germination (Opoku, Osagie & Ekperigin, 1983; Mtebe, Ndabikunze, Bangu & Mwenezi, 1993; Pawar & Pawar, 1997). Since fat provides twice as much energy as carbohydrates, the reduction in fat content observed during germination implies a reduction in the energy value of pearl millet malt compared to grain. However, in the case of pearl millet, this reduction may bring an increase in palatability of pearl millet food products. The development of fatty acids, which occur mainly due to the action of lipase, cause bitterness and can make pearl millet meals unacceptable (Lai & Varriano-Marston, 1980a). Generally, pearl millet

varieties are characterised by high lipid and high protein contents compared to other species of millets and sorghum (Hoseney, Varriano-Marston & Dendy, 1981). This is due to a high ratio of germ to endosperm of the pearl millet grain, which is responsible not only for the high protein content but also the high lipid content of the grain.

In both pearl millet varieties, malts had lower protein content than the grains. The slight decrease in protein content of pearl millet germinated grains with germination time can be attributed to the loss of low molecular weight nitrogenous compounds during the steeping process and rinsing of the grains during germination.

The results of this research are similar to those of Bhise, Chavan & Kadam (1988), who found that the crude protein of pearl millet decreased from 11.6 to 11.2% after 72 h of germination at 30 °C. Mtebe, Ndabikunze, Bangu & Mwenezi (1993) revealed that germination did not induce significant variation in protein content of pearl millet and other cereal grains investigated. However, Opoku, Ohenhen & Ejiofor (1981) found an increase in protein content of pearl millet, from 8.6% to 11.8 %, after germination for 3 days at 25 °C. The increase in protein content during germination was also observed in wheat, triticale, barley and rye by Dalby & Tsai (1976). In sorghum, Subramanian, Sambasiva, Rao, Jambunathan, Murty & Reddy (1995) reported that, malts show lower protein than non-malted grains. The difference of the results of this research and that of other workers can be explained by the fact that the reported increase in protein content during germination, which is attributed to loss in dry weight, particularly carbohydrates, through respiration, is not true, but apparent, since the absolute amount of protein per kernel does not change significantly during germination.

Nitrogen Solubility Index or Modification Index, or Kolbach index, as is known by brewers (Lewis & Young, 1995; Nzelibe & Nwasike, 1995), gives an indication of the amount of water-soluble nitrogen expected in the wort. The increase in Nitrogen Solubility Index and soluble nitrogen with germination time for pearl millet of SDMV 89004 variety can be due to gradual degradation of reserve protein into amino acids and short peptides caused by rising the levels of protease enzymes during germination. Working with pearl millet, Nzelibe & Nwasike (1995) also found that soluble nitrogen of wort and Modification Index increased with germination time. The fact that in variety SDMV 91018, ungerminated pearl millet grains had higher Nitrogen Solubility Index and soluble nitrogen than germinated samples may have been due to the loss of amino acids through leaching during the steeping process and during the watering treatments. Nitrogenous substances are lost by leaching during steeping, but there are no gains or appreciable losses from the whole grain during the other stages (Briggs, 1998). This decrease was unexpected since, as from the malt FAN results, the two pearl millet varieties used apparently had similar grain protein structure and degradability. Additionally, variety SDMV 89004, which had softer endosperm, had lower NSI and soluble nitrogen than SDMV 91018.

Nitrogen Solubility Index is important for food use since soluble nitrogen is regarded as digestible nitrogen. Hence, the increase in Nitrogen Solubility Index due to malting is a compliment of the increase in *in vitro* protein digestibility observed in both varieties of pearl millet malts investigated.

The increase in *in vitro* protein digestibility with time of germination, from 69% to 95% and from 58% to 94% for the varieties SDMV 89004 and SDMV 91018, respectively, can be attributed to an increase in soluble proteins, due to partial hydrolysis of storage proteins by endogenous proteases produced during the germination process. Such partially hydrolysed storage proteins may be more easily available for pepsin attack (Wu & Wall, 1980; Bhise,

Chavan & Kadam, 1988). Pepsin is used to hydrolyse the insoluble protein into soluble amino acids and peptides in the *in vitro* protein digestibility assay.

The decrease in antinutrients (phytic acid) may have also contributed to the high levels of protein digestibility observed. In this research, ungerminated pearl millets had 0.09% and 0.11% of total polyphenols in the variety SDMV 89004 and SDMV 91018, respectively. These values are similar to that reported in pearl millet by other authors (reviewed by Serna-Saldivar & Rooney, 1995). Antinutrients, such as tannin-polyphenols and phytic acid can bind to proteins including enzymes, and are therefore likely to inactivate enzymes involved in hydrolysis of endosperm materials (Chavan, Kadam & Salunkhe, 1981).

Other authors have also reported a significant increase in *in vitro* protein digestibility when finger and pearl millets are germinated (Bhise, Chavan & Kadam, 1988; Khetarpaul & Chauhan, 1990b; Pawar & Pawar, 1997). The increase in *in vitro* protein digestibility due to malting observed in pearl millet is of great nutritional significance for people living in the Semi-Arid Tropics (SAT) of the world since it will mean better utilisation of the protein of pearl millet.

Protein quality is critically important in developing countries where human diets consist mainly of cereals. This is especially valid for weanling children and infants who have a high essential amino acid requirement per kilogram of body weight (Serna-Saldivar & Rooney, 1995). However, a diet of cereals (with 8-10% protein content) is able to meet the protein requirements of adults, provided enough is eaten to supply the energy requirements (Cheftel, Cuq & Lorient, 1985).

Amino acid composition of food is important in evaluating the nutritive value of the protein, while protein digestibility is a primary determinant of the availability of its amino acids (FAO/WHO, 1990). A complete protein is one

that contains all the essential amino acids in the amounts and proportions to maintain life and support growth when used as the sole source of protein. Such a protein is said to have high biological value (Potter & Hotchkiss, 1995). In general, the levels of essential amino acids showed little changes with germination time. Although a change in the amino acid profile was observed due to germination, the total amino acid content remained the same since the protein content did not change.

Like in other tropical cereals, the most limiting amino acid in pearl millet is lysine. Lysine content of the variety SDMV 89004 showed a non-significant decrease. However, the lysine content of the pearl millet of the variety SDMV 91018 increased throughout germination. The difference in lysine content between the two varieties could be due to slight differences in the proportion of germ in both pearl millet varieties investigated. The increase in the lysine content of the protein of germinated pearl millet of variety SDMV 91018 is related to the transamination (change of one amino acid into another one), which may have occurred during germination affecting the amino acid profile of pearl millet. This transamination was also reported in sorghum by Taylor (1983).

In this research, the level of leucine in pearl millet malts was generally higher than the FAO Scoring Pattern (Serna-Saldivar, McDonough & Rooney, 1990; Hoseney, 1994). Generally, the essential amino acid contents found in this investigation are between the range published by other authors (reviewed by Lásztity, 1984; Chung & Pomeranz, 1985; Ejeta, Hassan & Mertz, 1987; Serna-Saldivar, McDonough & Rooney, 1990; reviewed by Serna-Saldivar & Rooney, 1995) for pearl millet grains. With the exception of the leucine, they were lower than that of the FAO Scoring Pattern. However, the lysine content of the malt of one of the pearl millet variety investigated was about 75% of the FAO Scoring Pattern for lysine. Taylor (1983) found an increase of nearly 4-fold in lysine content of sorghum during germination. Almeida-Dominguez, Serna-Saldivar, Gomez-Machado & Rooney (1993) reported an

increase in lysine from 2.2 to 3.2 and 3.0 to 7.8 g/100 g of protein when normal and high lysine-sorghums were germinated. The difference between the results of pearl millet with that of sorghum may be related to the fact that in pearl millet grains, lysine content is already relatively high compared to sorghum.

The nutritional and health significance of high leucine content cereals has been a controversial subject. Reports from Bender (1983) and Magboul & Bender (1983) indicate that diets in leucine can precipitate niacin deficiency. Niacin deficiency disease, pellagra, is endemic, and signs of it are sometimes referred as the four successive "D": dermatitis, diarrhea, dementia and death (Hulse, Laing & Pearson, 1980). However, more recent studies showed that leucine is not involved in the etiology of pellagra (Cook & Carpenter, 1987; Young & Fukagawa, 1988).

The fact that phytic acid content was reduced from 0.24% and 0.27% to 0.024% and 0.037% for the varieties SDMV 89004 and SDMV 91018, respectively, with germination time is presumably due to phytase activity. Phytase is an enzyme which can hydrolyse phytic acid to inositol and free orthophosphate (Thompson & Serraino, 1985). Like other cereals, pearl millet contains a considerable amount of phytic acid, representing more than 70% of the total phosphorus in the grain (Chauhan, Suneja & Bhat, 1986). Opoku, Ohenhen & Ejiofor (1981) also found a decrease in pearl millet phytic acid from 0.26 % in grain to 0.04 % in malt. Hulse, Laing & Pearson (1980) reported phytic acid values from 0.21 to 0.25 % in finger millet, whereas phytate content values for whole meal grain of proso millet ranged from 0.17 to 0.47%.

As stated, phytic acid interacts with minerals and other nutrients such as proteins making them unavailable to the organism (Kumar & Kapoor, 1984). The decrease in pearl millet phytic acid observed in this research will improve the nutritional quality of pearl millet malt food products by increasing the

bioavalability of proteins and minerals. Pearl millet is known as a good source of the essential minerals, calcium, iron, zinc, copper and manganese (Kumar & Kapoor, 1984). However, the bioavailability of these minerals may be affected by the presence of antinutrients such as phytic acid and polyphenols.

The malting process drastically reduced the pasting peak viscosity of pearl millet malts in both varieties investigated. Malleshi & Desikachar (1986b) and Mbithi-Mwikya, Van Camp, Yiru & Huyghebaert (2000) working with finger millet, have also reported this. The reduction in peak viscosity may be attributed to the high α-amylase activity of malts.

Alpha-amylase is known to rapidly solubilise starch into dextrins, thereby reducing the viscosity of starch solutions (Bamforth & Barclay, 1993). Additionally, the decrease in starch content observed in both pearl millet varieties investigated may have also contributed somewhat to the reduction in flour paste viscosity. When α-amylase acts on starch molecules a few cleavages of α (1 → 4) glucosidic bonds by this enzyme causes rapid decrease in the size of starch molecules, and there is an accompanying dramatic decrease in the viscosity of a starch paste. For this reason, the enzyme is commonly called the liquefying enzyme (Lewis & Young, 1995).

Germination of pearl millet for more than 1 day produced malts of free-flowing slurry with minimum viscosity. The rate of lowering of viscosity in pearl millet malt flours was greatest up to 3 days germination, beyond which the changes were slight, in fact could not be any further reduction since it was like water. This observation agrees with the findings of Mtebe, Ndabikunze, Bangu & Mwemezi (1993), working with finger and pearl millets.

The reduction of viscosity of the variety SDMV 89004, which had higher α-amylase activity than SDMV 91018, was greater than in SDMV 91018. This clearly shows the role of α-amylase in malt in reducing the flour viscosity. The peak viscosity of samples germinated for longer periods was lower than that

of samples germinated for shorter periods, presumably due to the partial hydrolysis of starch by α-amylase during the germination process and the higher level of α-amylase in the longer germinated malts.

In the SADC region and most other African countries where porridge is a staple food, the pasting properties (i.e. viscosity of the flour) is an important parameter. Porridges of high viscosity are more preferable for adults, because porridge is generally eaten with the fingers. Low viscosity porridges are suitable for consumption by infants as weaning foods due to their limited stomach capacity and the ability to chew (Pelembe, Erasmus & Taylor, in press). The period of a human being's development from the neonatal stage to the preschool stage is critical to growth. During this period, adequate food intake (including that of weaning foods) is vital to good nutritional status (Mosha & Lorri, 1987). The low viscosities observed after germinating both pearl millet varieties is a good indication that malted pearl millet is suitable as a diastatic adjunct to reduce the viscosity of cereal-based weaning porridges.

Mosha & Lorri (1987) found that three times as much germinated flour could be used, while maintaining the same consistency of the gruel using equal values of porridge. The addition of 5% germinated low-tannin sorghum flour (enzyme rich) to thick ungerminated sorghum and maize gruels reduced the viscosity to acceptable weaning food consistency. Food intake by preschool children 12 - 48 months of age was found to be significantly higher for bulk-reduced, low viscosity gruel with 20 % solids, than with ungerminated gruel (Mosha & Lorri, 1987). In ungerminated gruel, the viscosity of 1000 - 3000 cP (spoonable) is only obtained with gruels containing about 10 % flour (Mosha & Svanberg, 1983). Hence, the low viscosity malts found in this research could be suitable as bulk-reduced weaning foods of high nutrient density, which could eventually improve the nutritional status of young children and raise the nutrient intake of the infirm.

The fact that germination successfully reduced the mousy odour of pearl millets is important for the rural communities of Africa and India, where pearl millet is a staple food, since it will increase the palatability and could increase the consumption of pearl millet food products.

Since the exact compound, which causes the mousy odour, as well as the mechanism in which the mousy odour is promoted, are not known for certain, one could speculate that the phenolic pigments, which are responsible for the mousy odour, may have been leached out during the germination process. In fact the reduction of the C-glycosylflavone (Reddy, Faubion & Hoseney, 1986), the major flavone known to be present in pearl millet grains and mainly concentrated in the germ, could be noticed by the changes in the colour from the natural grey colour characteristic of pearl millet grain to the light brown (tan) of the pearl millet malts. The decrease in mousy odour observed during the pearl millet malting process can also be attributed to the decrease in pH due to the growth of lactic acid bacteria. The colour of phenolic pigments can be changed as a function of pH (Davídek, Velísek & Pororný, 1990).

CHAPTER 6

CONCLUSIONS AND RECOMMENDATIONS

The finding that modification of pearl millet grain structure during malting started at the germ floury endosperm interface and moved in the direction of the peripheral endosperm together with the finding that there was not much modification of the cell walls, aleurone layer or the horny endosperm shows that the pattern of modification in pearl millet is indeed similar to that of sorghum and different from that of barley.

The similarity of pearl millet with sorghum is also in the fact that non-germinated grains of pearl millet do not exhibit DP, α- or β-amylase activity.

The optimum malting conditions for high DP, α- and β-amylase activity, good FAN, and moderate malting loss are 25-30°C and 3-5 days germination, medium watering treatment. However, if the malting process is to be conducted in a short period of time, i.e. 1-3 days, the germination temperature and watering regime should be higher, 30-35 °C, high watering treatment. The levels of DP, FAN, α-amylase activity and malting loss of pearl millet malts, which are similar to sorghum malts, represent an excellent potential for utilisation of pearl millet for opaque beer brewing purposes. Additionally, pearl millet malt could be better than sorghum as an alternative for lager beer brewing due to the fact that it has higher β-amylase activity.

The high level of malt extract found in pearl millet malt, around 70%, together with the fact that malting considerably reduced the fat content to the levels which occur in barley, is an indication that pearl millet malt can be used in lager beer brewing at least as a barley malt extender. However, brewing studies are recommended to find out to what extent pearl millet malt can

substitute barley malt in the production of lager beer. At the above mentioned optimum germination conditions, the malting losses were moderate. This is an important finding since minimum malting loss and maximum achievable extract per unit weight of malt can increase the profitability of the brewing operation by lowering the production costs.

Variety affected most malt quality parameters, with variety SDMV 89004 producing better malt quality than variety SDMV 91018, possibly on account of its higher germinability.

Malting increases the percentage of the total carbohydrates which are enzyme susceptible, but reduces the total carbohydrate in pearl millet. The increase in the percentage of the total carbohydrates which are enzyme susceptible is due to starch hydrolysis into short chains of glucose (dextrins) and fermentable sugar maltose by endogenous amylases. The reduction in the total carbohydrate, is due to utilisation of some carbohydrates in malting to provide energy for germination changes, should not be very accentuated if malts are meant for the preparation of traditional southern African food products, such as porridges, weaning foods for infants and unleavened bread, where comparatively less carbohydrate reduction may be advantageous, to prevent the decrease of energy and nutrient density of the food products.

The decrease in fat during the germination process brings an increase in palatability of pearl millet food products since it reduces the possibility of development of free fatty acids, which occurs mainly due to the action of lipase, causing bitterness and makes the meals unacceptable within a few days.

The increase in the Water Solubility Index and soluble nitrogen with germination is of significant importance, since it gives an indication that germination can be used to increase the amount of soluble materials, carbohydrates and amino acids, which can be easily digestible, in pearl millet

food products. The increase in soluble proteins and consequently in Nitrogen Solubility Index, due to partial hydrolysis of storage proteins by endogenous proteases produced during the germination process, which makes the partially hydrolysed storage proteins more available for pepsin attack, is probably responsible for the increase in protein digestibility observed in pearl millet malts.

Although there were small changes in the amino acid profile, due to transamination, during germination, generally, germination imparted little change to the amino acid composition of the two pearl millet varieties investigated. The lysine content of the protein of germinated pearl millet of SDMV 91018 variety accounted for about 75% of the FAO Scoring Pattern.

Of particular nutritional importance is the decrease in phytic acid content of pearl millet during malting. This is probably due to phytase activity. The reduction in phytic acid can improve the bioavailability of both protein and essential minerals in pearl millet.

The reduction of pearl millet flour viscosity by malting is of functional importance in the SADC region and most other African countries, where porridge is a staple food. Porridges of high viscosity are more preferable for adults, because they are often eaten with fingers. Porridges of low viscosity are suitable for consumption by infants as weaning foods due to their limited stomach capacity and inability to chew. The reduction in pasting peak viscosity may be attributed to the greatly increased α-amylase activity in the malt. Such reduction in porridge viscosity is nutritionally important since it leads to acceptable weaning food consistency.

As with brewing malt quality parameters, variety SDMV 89004 produced malts of higher nutritional and better functional properties than SDMV 91018. The higher Germinative Vigour and Germinative Energy could be the reason for

the better nutritional and functional properties exhibited by the variety SDMV 89004.

Germination successfully almost eliminated the mousy odour, characteristic of pearl millet meals after short periods of storage. The water-soluble phenolics responsible for the generation of the mousy odour may have been inactivated or reduced to very low levels due to decrease in pH (by the growth in lactic acid bacteria) and by leaching out the phenolic pigments or as a result of the metabolic changes which took place in the germ during germination.

The findings that the mousy odour is almost eliminated by germination and the fact that malting can be carried out at potentially low cost without sophisticated and expensive equipment is very important to the rural communities of Africa and India who rely on pearl millet meals for their energy and other nutritional requirements. Additionally, the fact that the malting conditions and subsequent malt quality of pearl millet are similar to those of sorghum makes pearl millet malts suitable for the production of sorghum type beer widely consumed in the rural areas of Semi-Arid Tropics (SAT).

CHAPTER 7

REFERENCES

ABD ALLAH, M.A., MAHMOUD, R.M., EL-KALYOUBI, M.H. & ABOU ARAB, A.A. 1987. Physical properties of starches isolated from pearl millet, yellow corn, sorghum, sordan and pearl millet. *Starch/Staerke* **39**: 9-12.

ABDELRAHMAN, A., HOSENEY, R.C. & VARRIANO-MARSTON, E. 1984. The proportions and chemical compositions of hand-dissected anatomical parts of pearl millet. *Journal of Cereal Science* **2**: 127-133.

AGU, R.C. & EZEANALUE, J.C. 1993. Combined mashing of millet (*Pennisetum maiwa*) malts prepared with potassium bromate and gibberellic acid (GA_3) as additives. *Process Biochemistry* **28**: 475-479.

AGU, R.C. & OKEKE, B.C. 1991. Studies on the effect of potassium bromate on some malting properties of Nigerian millet. *Process Biochemistry* **26**: 89-92.

AGU, R.C. & OKEKE, B.C. 1992. Effect of potassium bromate on diastase, cellulase and hemicellulase development in Nigerian malted millet (*Pennisetum maiwa*). *Process Biochemistry* **27**: 335-338.

AGU, R.C. & PALMER, G.H. 1996. Enzymatic breakdown of endosperm proteins of sorghum at different malting temperatures. *Journal of the Institute of Brewing* **102**: 415-418.

BENDER, D.A. 1983. Effects of a dietary excess of leucine on the metabolic of tryptphan in the rat: A mechanism for pellagragenic action of leucine. *British Journal of Nutrition* **50**: 25-32.

BHISE, V.J., CHAVAN, J.K. & KADAM, S.S. 1988. Effects of malting on proximate composition and *in vitro* protein and starch digestibilities of grain sorghum. *Journal of Food Science and Technology - India* **25**: 327-329.

BIDLINGMEYER, B.A. COHEN, S.A. & TARVIN, T.L. 1984. Rapid analysis of amino acids using pre-column derivatization. *Journal of Chromatography* **336**: 93-104.

BIRZER, D.M. & KLOPFENSTEIN, C.F. 1988. The pearl millet goitrogens. *Cereal Foods World* **33**: 229-231.

BRIGGS, D.E. 1972. Enzyme formation, cellular breakdown and the distribution of gibberllins in the endosperm of barley. *Planta* **108**: 351-354.

BRIGGS, D.E. 1998. *Malts and Malting*. Blackie Academic & Professional. London. Pages 579-736.

BRIGGS, D.E. & MACDONALD, J. 1983. Patterns of modification in malting barley. *Journal of the Institute of Brewing* **89**: 260-263.

BRIGGS, D.E., HOUGH, J.S., STEVENS, R. & YOUNG, T.W. 1981. *Malting and Brewing Science*. Vol. 1. Chapman and Hall. London. Pages 281-389.

CHANDRASEKHER, G. & PATTABIRAMAN, T. 1981. Natural plant enzyme inhibitors: Isolation and characterization of two trypsin inhibitors from bajra (*Pennisetum typhoideum*). *Indian Journal of Biochemistry and Biophysics* **19**: 1-5.

DUFFUS, C.M. 1987. Physiological aspects of enzymes during grain development and germination. In: KRUGER, J.E., LINEBACK, D. & STAUFFER, C.E. (Eds.). *Enzymes and Their Role in Cereal Technology.* American Association of Cereal Chemists, St. Paul, MN. Pages 83-115.

DUFOUR, J.P., MÉLOTTE, L. & SREBRNIK, S. 1992. Sorghum malts for the production of a lager beer. *Journal of the American Society of Brewing Chemists* **50**: 110-119.

DYER, T.A. & NOVELLIE, L. 1966. Kaffircorn malting and brewing studies. *Journal of Food Technology* **4**: 219 - 225.

EJETA, G., HASSAN, M.M. & MERTZ, E.T. 1987. *In vitro* digestibilities and amino acid composition of pearl millet (*Pennisetum typhoides*) and other cereals. *Applied Biology* **84**: 6016-6019.

EUROPEAN BREWERY CONVENCTION. 1987. Method 36. Germinative Energy. *Analytica* - EBC, 4th Ed. Brauerei - und Getranke - Rundschau. Page E 39.

EVANS, D.J. & TAYLOR, J.R.N. 1990. Influence of cultivar and germination conditions on proteolytic activities in sorghum malt. *Journal of the Institute of Brewing* **96**: 399-402.

EZEOGU, L.I. & OKOLO, B.N. 1995. Effects of air rest periods on malting of sorghum response to final warm water steep. *Journal of the Institute of Brewing* **101**: 39-45.

FOOD AND AGRICULTURE ORGANISATION & WORLD HEALTH ORGANISATION (FAO/WHO). 1990. *Carbohydrates in Human Nutrition. Interim Report of Joint FAO/WHO Expert Consultation.* Rome, Italy.

FOOD AND AGRICULTURE ORGANISATION. 1998. *Production Yearbook.* FAO. Rome, Italy.

FAULKS, R.M. & BAILEY, A.L. 1990. Digestion of cooked starches from different food sources by porcine α-amylase. *Food Chemistry* **36**: 191-203.

FINCHER, G.B. & STONE, B.A. 1993. Physiology and biochemistry of germination of barley. In: MacGREGOR, A.W. & BHATTY, R.S. (Eds.). *Barley: Chemistry and Technology.* American Association of Cereal Chemists, St. Paul, MN. Pages 247-295.

FRETZDORFF, B. POMERANZ, Y. & BECHTEL, D.B. 1982. Malt modification assessed by histo-chemistry, light microscopy, and transmission and scanning electron microscopy. *Journal of Food Science* **47**: 786-789.

FUSSEL, L.K. & DWARTE, D.M. 1980. Structural changes of the grain associated with black region formation in *Pennisetum americanum*. *Journal of Experimental Botany* **31**: 645-654.

GARCIA-VILLA NOVA, R., GARCIA-VILLA NOVA, R.J. RUIZ DE LOPE, C. 1982. Determination of phytic acid by complexometric titration of excess of iron (III). *The Analyst* **107**: 1503-1506.

GAZZAZ, S.S., RASCO, B.A., DONG, F.M. & BORHAN, M. 1989. Effects of processing on the thiamin, riboflavin, and vitamin B12 content of fermented whole grain cereal products. *Journal of Food Processing and Preservation* **13**: 321-334.

GIBBONS, G.C. 1981. On the relative role of the scutellum and aleurone in the production of hydrolases during germination of barley. *Carlsberg Research Communications* **46**: 215-225.

GLENNIE, C.W. 1984. Endosperm cell wall modification in sorghum grain during germination. *Cereal Chemistry* **61**: 285-289.

GLENNIE, C.W., HARRIS, J. & LIEBENBERG, N.V.D.W. 1983. Endosperm modification in germinating sorghum grain. *Cereal Chemistry* **60**: 27-31.

GOMEZ, M.I., OBILANA, A.B., MARTIN, D.F., MADZVAMUSE, M. & MONYO, E.S. 1997. *Manual of Laboratory Procedures for Quality Evaluation of Sorghum and Millet.* ICRISAT, Patancheru, India. Pages 37-45.

HAMAD, A.M. & FIELDS, M.L. 1979. Evaluation of the protein quality and available lysine of germinated and fermented cereals. *Journal of Food Science* **44**: 456-459.

HAMAKER, B.R., KIRLEIS, A.W., BUTLER, L.G., AXTELL, J.D. & MERTZ, E.T. 1987. Improving *in vitro* digestibility of sorghum with reducing agents. *Proceedings of the National Academy of Sciences*- USA **84**: 626-628.

HOSENEY, R.C. 1994. *Principles of Cereal Science and Technology.* 2nd ed. American Association of Cereal Chemists, St. Paul, MN. Pages 15-24.

HOSENEY, R.C., VARRIANO-MARSTON, E. & DENDY, D.A.V. 1981. Sorghum and millets. In: POMERANZ, Y. (Ed.). *Advances in Cereal Science and Technology.* Vol. 4. American Association of Cereal Chemists, St. Paul, MN. Pages 71-144.

HOSENEY, R.C., ANDREWS, D.U. & CLARK, H. 1987. Sorghum and pearl millet. In: OLSON, R.A. & FREY, K. (Eds.). *Nutritional Quality of Cereal Grains: Genetic and Agronomic Improvement.* American Society of Agronomy. Madison, WS. Pages 397-456.

HULSE, J.H., LAING, E.M. & PEARSON, O.E. 1980. *Sorghum and the Millets: Their Composition and Nutritive Value*. Academic Press, London. Pages 374-469.

HWANG, P. & BUSHUK, G.S. 1973. Some changes in the endosperm proteins during sprouting of wheat. *Cereal Chemistry* **50**: 147-150.

INTERNATIONAL STANDARDISATION ORGANISATION. 1988. Sorghum. Determination of tannin content. International Organisation for Standardisation. *ISO* 9648. ISO Properties. Jersey City, NJ.

JAIN, A.K. & DATE, W.B. 1975. Relative amylase activity of some malted cereal grains. *Journal of Food Science and Technology - India* **12**: 131-132.

JAYATISSA, P.M., PATHIRANA, R.A. & SIVAYOGASUNDERAM, K. 1980. Malting quality of Sri Lankan varieties of sorghum. *Journal of the Institute of Brewing* **86**: 18-20.

KACED, I., HOSENEY, R.C. & VARRIANO-MARSTON, E. 1984. Factors affecting rancidity in ground pearl millet (Pennisetum americanum (L.) Leeke). *Cereal Chemistry* **61**: 187-192.

KHETARPAUL, N. & CHAUHAN, B.M. 1989. Effect of germination and pure culture fermentation on HCl-extractability of minerals of pearl millet (*Pennisetum typhoideum*). *International Journal of Food Science and Technology* **24**: 327-331.

KHETARPAUL, N. & CHAUHAN, B.M. 1990a. Effect of germination and fermentation on available carbohydrate content of pearl millet. *Food Chemistry* **38**: 21-26.

KHETARPAUL, N. & CHAUHAN, B.M. 1990b. Effect of germination and fermentation on *in vitro* starch and protein digestibility of pearl millet. *Journal of Food Science* **55**: 883-884.

KLOPFENSTEIN, C.F. & HOSENEY, R.C. 1995. Nutritional properties of sorghum and the millets. In: DENDY, D.A.V. (Ed.). *Sorghum and Millets: Chemistry and Technology*. American Association of Cereal Chemists, St. Paul, MN. Pages 125-168.

KLOPFENSTEIN, C.F., HOSENEY, R.C. & LEIPOLD, H.W. 1983a. Effects of ascorbic acid in sorghum-, high leucine-, and casein-fed guinea pigs. *Nutritional Reports International* **27**: 121-129.

KLOPFENSTEIN, C.F., HOSENEY, R.C. & LEIPOLD, H.W. 1983b. Goitrogenic effects of pearl millet diets. *Nutritional Reports International* **27**: 1039-1047.

KNUTSON, C.A. 1986. A simplified colorimetric procedure for the determination of amylose in maize starches. *Cereal Chemistry* **63**: 89-92.

KOEHER, D.E. 1981. Hydrolytic enzyme production during sorghum germination. *Plant Physiology* **67**: 218.

KUMAR, V. & KAPOOR, A.C. 1984. Trace mineral composition of different varieties of cereals and legumes. *Indian Journal of Nutrition and Dietetics* **21**: 137-143.

LAI, C.C. & VARRIANO-MARSTON, E. 1980. Changes in pearl millet meal during storage. *Cereal Chemistry* **57**: 275-277.

LÁSZTITY, R. 1984. *The Chemistry of Cereal Proteins.* CRC Press. Boca Raton, FL. Pages 185-189.

LORENZ, K. 1980. Cereal sprouts: Composition, nutritive value and food applications. *Critical Reviews in Food Science and Nutrition* **13**: 353-385.

LEWIS, M.J. & YOUNG, T.W. 1995. *Brewing*. Chapman & Hall. London. Pages 84-120.

MACEY, A. 1977. Malting in the seventies. *Brewers Guardian* **106**: 81-85.

MACGREGOR, A.W. 1996. Malting and brewing science: Challenges and opportunities. *Journal of the Institute of Brewing* **102**: 97-102.

MACGREGOR, A.W. & MATSUO, R.R. 1982. Starch degradation in endosperm of barley and wheat kernels during initial stages of germination. *Cereal Chemistry* **59**: 210-212.

MACGREGOR, A.W. & LENOIR, C. 1987. Studies on alpha-glucosidase in barley and malt. *Journal of the Institute of Brewing* **93**: 334-337.

MACGREGOR, A.W., GORDON, A.G., MEREDITH, W.O.S. & LACROIX, L. 1972. Site of alpha-amylase in developing barley kernels. *Journal of the Institute of Brewing* **78**: 174-179.

MAGBOUL, B.I. & BENDER, D.A. 1983. The effects of dietary excess leucine on the synthesis of nicotinamide nucleotides in the rat. *British Journal of Nutrition* **49**: 321-329.

MALLESHI, N.G. & DESIKACHAR, H.S.R. 1986a. Nutritive value of malted millet flours. *Qualitas Plantarum Plant Foods for Human Nutrition* **36**: 191-196.

MALLESHI, N.G. & DESIKACHAR, H.S.R. 1986b. Influence of malting conditions on quality of finger millet malt. *Journal of The Institute of Brewing* **92**: 81-83.

MALLESHI, N.G. & DESIKACHAR, H.S.R. 1986c. Studies on comparative characteristics of some tropical cereals and millets. *Journal of The Institute of Brewing* **92**: 174-176.

MATHEWSON, P.R. & SEABOURN, B.W. 1983. A new procedure for specific determination of β-amylase in cereals. *Journal of Agricultural and Food Chemistry* **31**: 1322-1326.

MAYER, A.M. & POLJAKOFF-MAYBER, A. 1989. *The Germination of Seeds*, 4th ed. Pergamon Press. Oxford.

MBITHI-NWIKYA, S., VAN CAMP, J. YIRU, Y. & HUGHEBAERT, A. 2000. Nutrient and antinutrient changes in finger millet (*Eleusine coracana*) during sprouting. *Lebensmittel-Wissenschaft und - Technologie* **33**: 9-14.

McCLEARY, B.V. & CODD, R. 1989. Measurement of β-amylase in cereal flours and commercial enzyme preparations. *Journal of Cereal Science* **9**: 17-33.

McDONOUGH, C.M. & ROONEY, L.W. 1985. Structure and phenol content of six species of millets using fluorescence microscopy and HPLC. *Cereal Foods World* **30**: 550.

McFADDEN, G.I. AHLUWAILIA, B., CLARKE, A.E. & FINCHER, G.B. 1988. Expression sites and development regulation of genes encoding (1→3, 1→4)-β-glucanases in germinated barley. *Planta* **173**: 500-508.

McGRATH, R.M., KALUZA, W.Z., DAIBER, K.H., VAN DER RIET, & GLENNIE, C.W. 1982. Polyphenols of sorghum grain, their changes during malting and their inhibition nature. *Journal of Agricultural and Food Chemistry* **30**: 450-456.

MERTZ, E.T., HASSEN, M.M., CAIRNS-WHITTERN, C. KIRLEIS, A.W., TU, L. & AXTELL, J.D. 1984. Pepsin digestibility of proteins in sorghum and other major cereals. *Proceeding of National Academy of Sciences*- USA **81**: 1-2

MOLL, M. & DE BLAUWE, J.J. 1991. *Beers & Coolers*. Intercept. Andover, U.K. Page 29.

MORGAN, A.M., MILLER, F.R. & QUINBY, J.R. 1977. Manipulation of sorghum growth and development with gibberellic acid. *Agronomy* **69**: 789-793.

MORRALL, P., BOYD, H.K., TAYLOR, J.R.N. & VAN DER WALT, W.H. 1986. Effect of germination time, temperature and moisture on malting of sorghum. *Journal of the Institute of Brewing* **92**: 439-445.

MOSHA, A.C. 1985. Weaning foods with particular emphasis on dietary bulk. Keynote address to an International Workshop on Dietary Bulk. Iringa, Tanzania. UNICEF, New York, USA and Swedish International Development Authority, Stockholm, Sweden. Pages 28-32.

MOSHA, A.C. & LORRI, W.S.M. 1987. High-nutrient density weaning foods from germinated cereals. In: ALNWICK, D., MOSES, S. & SCHMIDT, O.G. (Eds.). *Improving Young Child Feeding in Eastern and Southern Africa. Household-level Food Technology*. Proceedings of a workshop held in Nairobi, Kenya. 12-16 October 1987. International Development Research Centre, Ottawa, Canada. Pages 288-298.

MOSHA, A.C. & SVANBERG, U. 1983. Preparation of weaning foods with high nutrient density using flour of germinated cereals. *Food Nutrition Bulletin* **5**: 10-14.

MTEBE, K., NDABIKUNZE, B.K., BANGU, N.T.A. & MWENEZI, E. 1993. Effect of cereal germination on the energy density of togwa. *International Journal of Food Science and Nutrition* **44**: 175-180.

MUORIA, J.K. & BECHTEL, P.J. 1998. Diastatic power and α-amylase activity in millet, sorghum, and barley grains and malts. *Journal of the American Society of Brewing Chemists* **56**: 131-135.

MUTS, G.C.J. 1991. The use of sorghum in the brewing of lager beer. In: AXCELL, B. (Ed.). Proceedings of the Third Scientific and Technical Convention. Institute of Brewing. Central and Southern Africa Section. Sandton, South Africa. Pages 51-54.

NOUT, M.J.R. & DAVIES, B.J. 1982. Malting characteristics of finger millet, sorghum and barley. *Journal of the Institute of Brewing* **88**: 157-163.

NOVELLIE, L. 1959. Kaffircorn malting and brewing studies. III. Determination of amylases in kaffircorn malts. *Journal of the Science of Food and Agriculture* **10**: 441-449.

NOVELLIE, L. 1960. Kaffircorn malting and brewing studies. Part V. Occurrence of beta-amylase in kaffircorn malts. *Journal of the Science of Food and Agriculture* **11**: 457-463.

NOVELLIE, L. 1962a. Kaffircorn malting and brewing studies. XI. Effect of malting conditions on the diastatic power of kaffircorn malt. *Journal of the Science of Food and Agriculture* **13**: 115-120.

NOVELLIE, L. 1962b. Kaffircorn malting and brewing studies. XII. Effect of malting conditions on malting loss and total amylase activity. *Journal of the Science of Food and Agriculture* **13**: 121-123.

NOVELLIE, L. 1966. Kaffircorn malting and brewing studies. XIV. Mashing with kaffircorn malt: Factors affecting sugar production. *Journal of the Science of Food and Agriculture* **17**: 354-361.

NOVELLIE, L. & DE SCHAEPDRIJVER, P. 1986. Modern developments in traditional African beers. *Progress in Industrial Microbiology* **23:** 73-157.

NZELIBE, H. & NWASIKE, C.C. 1995. The brewing potential of "*Acha*" (*Digitaria exilis*) malt compared with pearl millet (*Pennisetum typhoides*) malts and sorghum (*Sorghum bicolor*) malts. *Journal of the Institute of Brewing* **101**: 345-350.

OKAMOTO, K., KITANO, H. & AKAZAWA, T. 1980. Biosynthesis and excretation of hydrolysates in germinating cereal seeds. *Plant Cell Physiology* **21**: 201-203.

OKOLO, B.N. & EZEOGU, L.I. 1995. Effects of air rest period on the mobilisation of sorghum reserve proteins. *Journal of the Institute of Brewing* **101**: 463-468.

OKOLO, B.N. & EZEOGU, L.I. 1996. Enhancement of amylolytic potential of sorghum malts by alkaline steeping treatment. *Journal of the Institute of Brewing* **102**: 79-85.

OKON, E.U. & UWAIFO, A.O. 1985. Evaluation of malting sorghums. *The Brewers Digest* **60**: 24-29.

OPOKU, A.R., OHENHEN, S.O. & EJIOFOR, N. 1981. Nutrient composition of millet (Pennisetum typhoides) grains and malt. *Journal of the Science of Food and Agriculture* **29**: 1247-1248.

OPOKU, A.R., OSAGIE, A.U. & EKPERIGIN, E.R. 1983. Changes in the minor constituents of millet (*Pennisetum americanum)* during germination. *Journal of Agricultural and Food Chemistry* **31**: 507-509.

OSAGIE, A.U. & KATES, M. 1984. The lipid composition of millet (*Pennisetum americanum*) seeds. *Lipids* **19**: 958-965.

OWUAMA, C.I. 1999. Brewing beer with sorghum. *Journal of the Institute of Brewing* **105**: 23-34.

PAL, A., WAGLE, D.S. & SHEORAIN, V.S. 1976. Some enzymatic studies on bajra (*Pennisetum typhoides*) and barley (*Hordeum vulgare*). *Journal of Food Science and Technology - India* **13**: 75-78.

PALMER, G.H. 1980. The morphology and physiology of malting barleys. In: INGLETT, G. & MUNCK, L. (Eds.). *Cereals for Food and Beverages.* Academic Press, New York. Pages 301-307.

PALMER, G.H. 1986. Adjuncts in brewing and distilling. In: CAMPBELL, I. & PRIEST, F.G. (Eds.). *Proceedings of the Second Aviemore Conference on Malting, Brewing and Distilling.* Aberdeen University Press. Aberdeen, U.K. Pages 24-45.

PALMER, G.H. 1989. Cereals in malting and brewing. In: PALMER, G.H. (Ed.). *Cereal Science and Technology.* Aberdeen University Press. Aberdeen, U.K. Pages 61-278.

PALMER, G.H. & MACKENZIE, C.I. 1986. Levels of alkali-soluble β-D-glucans in cereal grains. *Journal of the Institute of Brewing* **92**: 461-474.

PATHIRANA, R.A., SHIVAYOGASUNDARAM, K. & JAYATISSA, P.M. 1983. Optimisation of conditions for malting sorghum. *Journal of Food Science and Technology* - India **20**: 108-111.

PAWAR, V.S. & PAWAR, V.D. 1997. Malting characteristics and biochemical changes of foxtail millet. *Journal of Food Science and Technology – India* **34**: 416-418.

PELEMBE, L.A.M., ERASMUS, C. & TAYLOR, J.R.N. Development of protein-rich composite sorghum-cowpea instant porridge by extrusion cooking process. *Lebensmittel-Wissenschaft und - Technologie* (in press).

PICKERELL, A.T.W. 1986. The influence of free alpha-amino nitrogen in sorghum beer fermentation. *Journal of the Institute of Brewing* **92**: 568-571.

PICKERELL, A.T.W. 1987. *Factors Affecting Fermentation of Sorghum Beer.* MSc. Thesis, University of the Witwatersrand, Johannesburg, South Africa.

POTTER, N.N. & HOTCHKISS, J.H. 1995. *Food Science.* 5th ed. Chapman & Hall, New York. Pages 49-51.

RANKI, H. 1990. Secretion of α-amylase by the epithelium of barley scutellum. *Journal of the Institute of Brewing* **96**: 307-309.

REBAR, V., FISHBACH, E.R., APOSTOLOPOULOS, D. & KOKINI, J.L. 1984. Thermodynamics of water and ethanol adsorption on four starches as model biomass systems. *Biotechnology and Bioengineering* **26**: 331-335.

REDDY, V.P., FAUBION, J.M. & HOSENEY, R.C. 1986. Odor generation in ground, stored pearl millet. *Cereal Chemistry* **63**: 403-406.

ROOD, S.B. 1995. Heterosis and the metabolism of gibberellin A_{20} in sorghum. *Plant Growth Regulation* **16**: 271-278.

ROONEY, L.W. & MILLER, F.R. 1982. Variation in the structure and kernel characteristics of sorghum. In: MERTIN, J.V. (Ed.). Proceedings of The International Symposium on Sorghum Grain Quality. ICRISAT, Patancheru, India. Pages 143-162.

SANKARA RAO, D.S. & DEOSTHALE, Y.G. 1983. Mineral composition, ionisable iron and soluble zinc in malted grains of pearl millet and ragi. *Food Chemistry* **11**: 217-223.

SEITZ, L.M., WRIGHT, R.L., WANISKA, R.D. & ROONEY, L.W. 1993. Contribution of 2-acetyl-1-pyrroline to odors from wetted ground pearl millet. *Journal of Agricultural and Food Chemistry* **41**: 955-958.

SERNA-SALDIVAR, S. & ROONEY, L.W. 1995. Structure and chemistry of sorghum and millets. In: DENDY, D.A.V. (Ed.). *Sorghum and Millets: Chemistry and Technology*. American Association of Cereal Chemists, St. Paul, MN. Pages 69-124.

SERNA-SALDIVAR, S., McDONOUGH, C.M. & ROONEY, L.W. 1990. The millets. In: LORENZ, K.J. & KULP, K. (Eds.). *Handbook of Cereal Science & Technology*. Marcel Dekker, New York. Pages 271-301.

SHARMA, A. & KAPOOR, A.C. 1997. Effect of processing on the nutritional quality of pearl millet. *Journal of Food Science and Technology - India* **34**: 50-53.

SHEORAIN, B.S. & WAGLE, D.S. 1973. β-amylase activity in germinated bajra and barley varieties. *Journal of Food Science and Technology - India* **10**: 184-186.

SHUKLA, S.S., GUPTA, O.P., SWARKAR, N.J., TOMAR, A.K. & SHARMA, Y.K. 1986. Malting quality of ragi varieties: Nutrient and mineral composition of their malts. *Journal of Food Science and Technology - India* **23**: 235-237.

SLACK, P.T., BAXTER, E.D. & WAINWRIGHT, T. 1979. Inhibition by hordein of starch degradation. *Journal of the Institute of Brewing* **85**: 112-114.

SOUTH AFRICAN BUREAU OF STANDARDS. 1970. *Standard Test Method for the Determination of Diastatic Power of Malts prepared from Kaffircorn (Sorghum) including Bird-proof varieties and from Millet.* ***SABS Method 235.*** South Africa Bureau of Standards. Pretoria.

SRIPRIYA, G., ANTONY, U. & CHANDRA, T.S. 1997. Changes in carbohydrate, free amino acids, organic acids, phytate and HCl extractability of minerals during germination and fermentation of finger millet (*Eleusine coracana*). *Food Chemistry* **58**: 345-350.

STUART, I.M., LOI, L. & FINCHER, G.B. 1986. Development of (1→3), (1→4)-β-D glucan endohydrolase isoenzymes in isolated scutella and aleuorne layers of barley (*Hordeum vulgare*). *Plant Physiology* **80**: 310-314.

SUBRAMANIAN, V., SAMBASIVA, N., RAO, N.S., JAMBUNATHAN, R., MURTY, D.S & REDDY, B.V.S. 1995. The effect of malting on the extractability of proteins and its relationship to diastatic activity in sorghum. *Journal of Cereal Science* **21**: 283-289.

TAYLOR, J.R.N. 1983. Effect of malting on the protein and free amino nitrogen composition of sorghum. *Journal of the Science of Food and Agriculture* **34**: 885-892.

TAYLOR, J.R.N. 1992. Mashing with malted grain sorghum. *Journal of the American Society of Brewing Chemists* **50**: 13-18.

TAYLOR, J.R.N. & BOYD, H.K. 1986. Free alpha-amino nitrogen production in sorghum beer mashing. *Journal of the Science of Food and Agriculture* **37**: 1109-1117.

TAYLOR, J.R.N. & DAIBER, K.H. 1988. Effect of calcium ions in sorghum beer mashing. *Journal of the Institute of Brewing* **94**: 68-70.

TAYLOR, J.R.N. & DEWAR, J. 1992. Sorghum malting Technology. In: DENDY, D.A.V. (Ed.). *5th Quadrennial Symposium on Sorghum and Millets.* International Association of Cereal Science and Technology. Paris. Pages 55-72.

TAYLOR, J.R.N. & DEWAR, J. 2001. Developments in sorghum food technologies. *Advances in Food and Nutrition Research* **43**: 217-264.

TAYLOR, J.R.N. & ROBBINS, D.J. 1993. Factors influencing beta-amylase activity in sorghum malt. *Journal of the Institute of Brewing* **99**: 413-416.

TAYLOR, J.R.N. & VON BENECKE, R. 1990. Development of a simple assay for the direct determination of β-amylase in sorghum malt. In: CAMPBELL, I. (Ed.). *Proceedings of the Third Aviemore Conference on Malting, Brewing and Distilling* Institute of Brewing. London. Pages 344-347.

THOMPSON, L.U. & SERRAINO, M.R. 1985. Effect of germination on phytic acid, protein and fat content of rapeseed. *Journal of Food Science* **50**: 1200-1203.

UDAYASEKHARA RAO, P. 1994. Evaluation of protein quality of brown and white ragi (*Eleusine coracana*) before and after malting. *Food Chemistry* **51**: 433-436.

VERBUGGEN, M.A., BELDMAN, G. & VORAGEN, A.G.J. 1995. The selective extraction of glucuroarabinoxylans from sorghum endosperm cell walls using barium and potassium hydroxide solutions. *Journal of Cereal Science* **21**: 271-282.

WANISKA, R.D., HUGO, L.F. & ROONEY, L.W. 1992. Practical methods to determine presence of tannins in sorghum. *Journal of Applied Poultry Research* **1**: 122-128.

WHITNEY, E.N., CATALDO, C.B. & ROLFES, S.R. 1987. *Understanding Normal and Clinical Nutrition.* 2nd ed. West Publishing, New York. Pages 34-37.

WU, Y.V. & WALL, J.S. 1980. Lysine content of protein increased by germination of normal and high lysine sorghum. *Journal of Agricultural and Food Chemistry* **28**: 455-458.

YOON, J.H., THOMPSON, L.U. & JENKINS, D.J.A. 1983. The effect of phytic acid on *in vitro* rate of starch digestibility and blood glucose response. *American Journal of Clinical Nutrition* **38**: 835-837.

YOUNG, V.R. & FUKAGAWA, N.K. 1988. Amino acid interactions: A selective review. In: BODWELL, C.E. & ERDMAN, J.W. (Eds.). *Nutrient Interactions*. Marcel Dekker, New York. Pages 27-71.

ZELEZNAK, K. & VARRIANO-MARSTON, E. 1982. Pearl millet (*Pennisetum americanum* (L.) Leeke) and grain sorghum (*Sorghum bicolor* (L.) Moench) ultrastructure. *American Journal of Botany* **69**: 1306-1313.

ZEIGLER, P. 1999. Cereal beta-amylases. *Journal of Cereal Science* **29**: 195-204.

ZEURCHER, C. 1971. Isolierung einiger lipide aus dem malz und ihre quantitative bestimmung in würze und bier. *Monatsschrift für Brauerei* **24**: 276-277.

APPENDIX A: SCORE SHEET USED IN THE SENSORY EVALUATION OF ODOUR GENERATED BY PEARL MILLET

DESCRIPTIVE TEST: PEARL MILLET FLOUR

NAME:_____ SET:____ DATE:_____

You have been provided with 5 samples of pearl millet flour. Please carefully open the polytop while holding it close to your nose. Smell sample "C" first, then smell the other 4 coded samples and evaluate them for the intensity of the ***Mousy odour*** in comparison with sample "C", which is the sample with a rating of ***9*** for the ***Mousy odour***. Rate the aroma intensity on the scale below, with ***1*** being the ***least intense*** and ***9*** the ***most intense***. Please close the polytops in between the evaluations to prevent the volatilise from escaping.

SAMPLE:___

Mousy odour

1 2 3 4 5 6 7 8 9

SAMPLE:___

Mousy odour

1 2 3 4 5 6 7 8 9

SAMPLE:___

Mousy odour

1 2 3 4 5 6 7 8 9

SAMPLE:___

Mousy odour

1 2 3 4 5 6 7 8 9

APPENDIX B: LIST OF PUBLICATIONS AND PRESENTATIONS

PUBLICATIONS:

PELEMBE, L.A.M., DEWAR, J. & TAYLOR, J.R.N. 2002. Effect of malting conditions on pearl millet malt quality. *Journal of the Institute of Brewing* **108**: 7-12.

PELEMBE, L.A.M., HALL, A.N., DEWAR, J. & TAYLOR, J.R.N. 2001. Modification of pearl millet grain structure during malting. *Microscopy Society of Southern Africa - Proceedings* **31**: 54.

ORAL PRESENTATION:

PELEMBE, L.A.M., DEWAR, J. & TAYLOR, J.R.N. 2001. Effect of malting conditions on functional and nutritional properties of pearl millet. *16th Biennial Congress of the South African Association of Food Science and* Technology, Durban, South Africa, September 12-14, 2001.

PELEMBE, L.A.M., HALL, A.N., DEWAR, J. & TAYLOR, J.R.N. 2001. Modification of pearl millet grain structure during malting. *Microscopy Society of Southern Africa Annual Conference.* University of the Witwatersrand, Johannesburg, South Africa, December 5-7, 2001.

POSTER PRESENTATION:

PELEMBE, L.A.M., DEWAR, J. & TAYLOR, J.R.N. 2001. Pearl millet (*Pennisetum glaucum* (L.) R. Br.) malting. The 9th *International Symposium on Pre-Harvest Sprouting in Cereals.* Berg-en-Dal Camp, Kuger National Park, South Africa; June 24-28, 2001.