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Quality characteristics of pearl millet malt as affected by steeping temperature and germination period

R. Dahiya, R.B. Yadav*, B.S. Yadav and R. Yadav

Department of Food Technology, Maharshi Dayanand University, 124001 Rohtak, Haryana, India; rita.by@rediffmail.com

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RESEARCH ARTICLE

Abstract

The effect of malting conditions on pearl millet malt quality was investigated. Grains were steeped for 24 h at different temperatures of 25, 30 and 35 °C and germinated at 25 °C for 24, 48 and 96 h. Generally, malt quality parameters such as malting loss, diastatic power, α- and β-amylase activity, free amino nitrogen (FAN), water extracts were significantly affected (P<0.05) by steeping temperature and germination time. Malting loss, FAN, hot water extract, diastatic power of the malt increased with increase in germination time as well as steeping temperature However germination time from 24 h onwards to 48 h exerted more pronounced enhancing effect on malting loss (140%), hot water extract (145%) and FAN (41%) of the malt. A steeping temperature of 30-35 °C and germination time of 4 days were found to be optimal for pearl millet malting as these conditions resulted in high diastatic power, α- and β-amylase activity, hot water extract, FAN and moderate malting loss.

Keywords: pearl millet malt, diastatic power, free amino nitrogen, malting conditions

1. Introduction

Pearl millet (*Pennisetum glaucum*) is the staple food and fodder crop of millions of poor rural families in the hottest and driest agricultural environments of India. Indeed in some of the hottest, driest regions of India, pearl millet is the only cereal that can be grown and so plays a critical role in food security. However, the utilisation of millets for food purpose is limited due to the presence of various anti-nutrients and poor protein/carbohydrate digestibility (Sharma and Kapoor, 1997). Various processing technologies may affect the physicochemical composition of food grain and improve their nutritive value (Hithamani and Srinivasan, 2014; Sihag *et al.*, 2015). Malting is a primary processing technology which describes the process of steeping dry grain in water until saturated, followed by germination under controlled conditions for a specific period and kilning, which is the final stage of malting (Chavan and Kadam, 1989). In African countries, pearl millet is traditionally processed by malting and fermentation. Malted pearl millet is used to make weaning foods, with reduced viscosity, for infants. It is also used for brewing traditional beer and low alcohol beverages (Pelembe *et al.*, 2002).

Malting is a biotechnological technique which involves the controlled germination of a cereal grain which aims at activating enzyme systems that catalyse the hydrolysis of polymerised reserved food materials, notably, proteins, starches and cell-wall substances, thus, extracting fermentable materials (MacLeod, 1977; Ogbonna, 2011). Important quality parameters of malting include high grain germination capacity and germinative energy, α- and β-amylase activity and free α-amino nitrogen (Evans *et al.*, 2005; Taylor, 1983). Diastatic power is a measure of the joint α- and β-amylase activity of the malt. Starch hydrolysis is carried out by the malt enzymes including α-amylase, β-amylase, limit dextrinase, and α-glucosidase (Manners, 1985). Limit dextrinase hydrolyses the (1→6)-α-glucosidic branch points in low molecular weight branched dextrins formed by the action of α- and β-amylase on starch components (Manners *et al.*, 1970). At the biochemical level, the combined action of α- and β-amylases, which develop during malting, is responsible for the breakdown of

starch to fermentable sugars during the process of malting (Palmer, 1989). Free amino nitrogen (FAN), which consists of free amino acids and small peptides and malt hot water extract, which gives an estimate of how much of the malt will solubilise during the brewing process (Briggs *et al.*, 1981) are other particularly important malt characteristics in lager beer brewing. Steeping is generally acknowledged as the most critical step in the malting process (Brigs *et al.*, 1981; Taylor and Dewar, 2001). Unlike barley and sorghum, little is known about the technology of millet malting especially regarding studies on the steeping and germination conditions for pearl millet malting. Steeping is generally considered as the most critical stage of the malting process as it activates the metabolism in embryonic and aleuronic tissues, which in turn leads to development of hydrolytic enzymes that initiate the biochemical changes during the process of malting (Bamforth and Barclay, 1993). Dewar *et al.* (1997) have reported that steeping time and temperature are critical in affecting the diastatic power, FAN and hot water extract of malt. However in contrast to barley malting, the steeping of the millet grains has been considered to be relatively less critical and therefore, limited research studies underlining the effect of steeping conditions particularly steeping temperature on the pearl millet malt quality are available (Pelembe *et al.*, 2002). Because of the small size of the pearl millet grains, the optimum steeping time is relatively short in contrast to 48-72 h for barley and a steeping time ranging from 6 and 16 h has been reported for pearl millet grains (Gomez *et al.*, 1997; Muoria and Bechtel, 1998). Researchers have reported optimum germination period of 3 to 5 days at a temperature ranging from 22 to 25 °C for malting of pearl millet (Gomez *et al.*, 1997; Muoria and Bechtel, 1998).

For commercial brewing, much of the malt used in India is from barley because of its outstanding malting qualities. However, the high cost of barley malt and limited barley production in many tropical countries and the high cost of importing conventional brewing ingredient into these countries especially African countries led to the adaptation of locally available cheaper food commodities like pearl millet. Therefore, pearl millet could be an alternate for increasing malt availability for both traditional and industrial use at low cost. The present study was undertaken with an objective to study the effect of steeping and germination conditions on the quality characteristics of pearl millet malt.

2. Materials and methods

Pearl millet grains (3.0 kg) of standard variety were purchased from local grain market in Rohtak (India). The grains were manually cleaned by removing broken and damaged kernels and foreign materials and washed three times with tap water before malting.

Quality characteristics of pearl millet grains

The various parameters measured were thousand kernels weight, moisture content, germinative energy, germinative capacity, and germination count and water sensitivity.

Thousand kernel weight

Three lots of 1000 kernels were taken and weighed to determine the weight of 1000 kernels on dry basis.

Moisture content

Moisture content of pearl millet grain was determined by drying 5 g of sample in a hot air oven at a temperature of 130±5 °C till a constant weight was obtained (AOAC, 1995).

Germinative energy

One hundred kernels grains were placed on two filter papers (Whatman No. 1; Whatman plc., Maidstone, UK) wetted with 4 ml of distilled water placed at the bottom of a petri dish, taking care to ensure that all the kernels were in good contact with the moist filter papers. The petri dish was then covered and incubated at an average temperature of 30 °C for 48 h. The kernels that sprouted at the end of the incubation were counted and germinative energy (GE) was expressed as suggested by Agbo (1996):

$$GE\ (\%) = 100 - N$$

where N = number of ungerminated grains.

Germinative capacity

Pearl millet grains (100) were placed in a 100 ml glass beaker containing 7.5% hydrogen peroxide solution and steeped at 25 °C for 48 h. The steep water was strained off and the sprouted grains separated from the unsprouted grains were then transferred on to a petri dish lined with moist filter paper and its lid also replaced with another moist filter paper. The petri dish was then wrapped in a jute bag and allowed to germinate at ambient temperature (32±2 °C) for about 24 h with distilled water being sprinkled at regular intervals. Newly germinated grains were counted and the result was added to the first (Agbo, 1996; Badau, 2004). The germinative capacity (GC) was calculated as:

$$GC\%\ (H_2O_2) = (200 - N)/2$$

where N = number of ungerminated grains.

Water sensitivity

Two lots of 100 grains were grown on filter papers in petri dishes; one moistened with 4 ml and the other with 8 ml water. The difference in the number of grains that germinated after 48 h at 28 °C in the two petri dishes was noted as the water sensitivity value (Briggs *et al.*, 1981).

Malting of pearl millet grains

The cleaned pearl millet grains (300 g) were placed in a 1 L jar to which 0.1% formaldehyde (600 ml) was added (Taylor and Dewar, 1992). The grains were steeped for 24 h at different temperatures of 25, 30 and 35 °C, respectively. The water was drained, and the grains were steeped with a fresh solution of formaldehyde for 18 h, drained, and allowed for a 2 h resting period in air. The jar was shaken gently to mix the grains and laid on its side to allow the grains to spread uniformly. The degree of steeping was calculated as follows:

$$\text{The degree of steeping (\%)} = \frac{(\text{weight of steeped grains} - \text{weight of unsteeped grains})}{\text{weight of unsteeped grains}} \times 100$$

Soaked grains were germinated in an incubator maintained at the 25 °C for 24, 48 and 96 h, respectively. Water was sprayed at an interval of 12 h. The germinated grains were turned occasionally to avoid meshing of the roots and shoots. At the end of each germination treatment, the germinated grains were removed from the vessel and dried in a forced air draft drying oven at 50 °C for 24 h. The dried malt (about 7.8% moisture content) was stored in the air tight polyethylene bags until further analysis.

Sprout length and roots /shoots weight of malted grains

Sprout length was measured by taking the average of length of 10 sprouts. The rootlets and acrospires were separated from the germinated pearl millet kernels by rubbing the grains in a nylon bag of coarse mesh size, which allowed the rootlets and acrospires to escape while retaining the kernels (Morrall *et al.*, 1986). The weight of rootlets and acrospires was expressed as the percentage of the total weight.

Malting loss

Total malt loss was calculated according to the easy and simple method described by Gomez *et al.* (1997):

$$\text{Malting loss (\%)} = \frac{\text{initial grain dry weight} - \text{dry malt weight}}{\text{initial grain dry weight}} \times 100$$

Moisture content of malt

The moisture content of pearl millet grain was determined by drying 5 g of sample in a hot air oven at a temperature of 105±5 °C till to constant weight was obtained (AOAC, 1995).

Cold water extract (CWE)

Cold water extract was determined by the method of Gothard *et al.* (1980). 3.0 g of sample was extracted with 30 ml solution (27.5 ml distilled water + 2.5 ml 0.1 M NH_4OH solution) at 25 °C. The extract was filtered through Whatman No. 1 filter paper and the percentage of dissolved sugar read at 20 °C with an abbe refractometer. The percentage of CWE was calculated as given below:

$$\% \text{ CWE} = P(M + 1000) / (100 - P)$$

where P is the percentage sugar by refractometer reading, M is moisture content of malt.

Hot water extract

The method as described by Gothard *et al.* (1980) and modified by Demuyakor and Ohta (1992) was used for determination of hot water extract (HWE). Malt grist (3 g) was placed into a 100 ml Erlenmeyer flask and extracted with 25 ml distilled water in a water bath at 45 °C for 30 min. The wort was then decanted from the mesh and kept separately while the mesh was brought to 100 °C in a boiling water bath. It was maintained at 100 °C for 2 min and then cooled to 65 °C. The mash and the wort were recombined and then further extraction was carried out at 65 °C for 1 h. At the end of the extraction, the solution was cooled and the volume was made up to 35 ml with distilled water. It was mixed and allowed to settle for 20 min and it was decanted into a fluted filter (Whatman No. 1). The percentage sugar content of the clarified wort was read with an abbe refractometer and the percentage HWE was calculated by using the following equation:

$$\%\text{HWE} = P(M + 1000) / (100 - M)$$

where P is the percentage sugar by refractometer reading, M is the moisture content of malt.

Free α-amino nitrogen content (FAN)

One gram of milled malt was added to 40 ml of 5% trichloroacetic acid at 30 °C. Extraction was carried out for 1 h at 30 °C. At intervals of 15 min, the extraction tubes were swirled to suspend the contents. 10 ml of extract was centrifuged at 4,500 rpm for 10 min and 1.0 ml of clear supernatant was diluted to 25 ml with distilled water. These samples were then subjected to the European Brewery

Convention ninhydrin assay (Lie, 1973). The absorbance of the sample as well as standard was measured using UV-spectrophotometer (UB-3200; Labindia, Mumbai, India). The results were expressed as mg free α-amino nitrogen content (FAN)/100 g dry weight malt:

$$FAN = \frac{\text{absorbance of the sample} \times 400}{\text{absorbance of the mean of standard}} \times \frac{100}{100 - M}$$

where M is the moisture content of malt.

Diastatic power

Diastatic power (DP)was determined following ferricyanide method as used by Gomez *et al.* (1997). 0.5 g of milled malt sample was weighed into centrifuge tube and added with 10 ml of peptone solution. The sample tube was placed in a water bath at 30 °C for 2 h and 15 min to extract the diastatic enzymes. At the end of extraction, the suspensions were centrifuged for 2 min at 3,000 rpm. Supernatant aliquot (0.5 ml) obtained from centrifuged suspensions was dispensed from centrifuged tube in to a 250 ml Erlenmeyer flask containing 100 ml buffered starch solution maintained at 30 °C in a water bath. After 1 h thorough mixing, 5 ml portion of digested starch solution was mixed with 4 ml of 0.05 N alkaline ferricyanide solution and left to stand in boiling water bath for 20 min. On cooling to 30 °C, 10 ml acetic acid salt and 0.4 ml potassium iodide solutions were added and the solution titrated with 0.05 N sodium thiosulphate solution until the complete disappearance of the blue colour of the starch-iodine complex. A blank was prepared of the unfiltered malt infusion and 2% buffered starch solution. Results were expressed as pearl millet diastatic units (PMDU) per g dry weight malt.

The diastatic power (DP) was calculated as follows:

$$DP = B - A\ (23 \times 200/250 \times 1/C)$$

where A = volume of sodium thiosulphate used for direct titration, B = volume of sodium thiosulphate used for blank determination, C = volume of unfiltered malt extract used for the digestion.

α- and β-amylase activity

The supernatant aliquot containing the extracted enzymes was taken in two separate Erlenmeyer flasks. α-amylase activity was determined according to the method of Preece (1947), as modified by Novellie (1960) by inactivating β-amylase by adding 0.316 g of calcium acetate to one flask. To inactivate the α-amylase, 0.284 g ammonium oxalate was added to another flask (Taylor and Von Benecke, 1990). α- and β-amylase activities were calculated as described in method for the determination of diastatic power.

Statistical analysis

Three replicates were taken for the determination of each parameter and the results were reported as means ± standard deviation. Two-way analysis of variance was performed using OPSTAT statistical software (Haryana Agricultural University, Hisar, India) at a significance level of 5%.

3. Results and discussion

Quality characteristics of pearl millet grains

The results on various quality characteristics of pearl millet grains as studied are shown in Table 1. Thousand kernel weight was recorded to be 9.23 g. Higher thousand kernel weight tends to have higher potential extract yields (Nso *et al.*, 2003). The moisture content of the grains was 11.60%. GE and GC of pearl millet grains were high (96.83 and 99.32%, respectively), compared with the recommended value i.e. >90% for malting (O'Rourke, 2004). The higher germination count of pearl millet grains (above 98%) was indicative of the good viability of the grains for malting.

Pearl millet malt quality characteristics as affected by malting conditions

Degree of steeping

Degree of steeping is described as the percentage increase in the weight of the grains after steeping due to increase in the moisture content of the grains. The degree of steeping increased as the steeping temperature increased and the highest degree of steeping (57.90%) was recorded after steeping of the grains for 24 h at 35 °C as shown in Table 2. Dewar *et al. (*1997) reported that degree of steeping of pearl millet grains is affected by both steeping time and temperature. According to Novellie (1962), the steep moisture increases as steeping temperature rises from 10 to 30 °C. The steeping of cereal grains in water is widely acknowledged as the most critical stage of the malting

Table 1. Quality characteristics of pearl millet grains used for malting.

Parameter	Value
1000 kernel weight (g)	9.23±0.03
Moisture content (%)	11.60±0.23
Germination capacity (%)	99.32±0.16
Germination energy (%)	96.83±0.16
Germination count	98.80±0.21
Water sensitivity	2.00±0.00

Table 2. Degree of steeping (%), moisture content (%) of dried and green malt as affected by varied conditions of steeping temperature and germination period.

Steeping temperature (°C)	Degree of steeping	Moisture content of dried malt				Moisture content of green malt			
		Germination period (h)			Mean A	Germination period (h)			Mean A
		24 h	48 h	96 h		24 h	48 h	96 h	
25 °C	49.50±0.65	7.89±0.0	7.97±0.0	8.13±0.0	7.99	34.4±0.26	42.7±8.8	42.0±8.5	39.7
30 °C	52.71±2.51	8.15±0.0	8.30±0.1	7.81±0.1	8.09	42.0±8.50	38.0±0.2	40.2±5.1	40.0
35 °C	57.90±1.02	7.41±0.0	7.23±0.1	7.40±0.0	7.35	50.5±13.8	60.6±15	29.4±0.2	46.8
Mean B		7.81	7.83	7.78		42.3	47.1	37.2	

process (French and McRuer, 1990). Moisture content both at the end of steeping and of the green malt have been reported to be positively correlated with malt quality in terms of DP, FAN and hot water extract (Dewar *et al.*, 1997).

Moisture content of green and dried malts

The moisture content of green malt was significantly affected by germination time and steeping temperature as well as their interactive effect (P<0.05) (Table 2). Moisture content ranging from 34 to about 60% was recorded for green malt at various steeping temperatures and germination time combinations. Regardless of the steeping temperature green malt moisture increased with increasing germination period from 24 to 48 h, after which it decreased. However, the moisture content of green malt increased very marginally as the steep temperature was increased from 25 to 30 °C. However, a further increase in steeping temperature to 35 °C resulted in a significant increase in moisture content. Green malt steeped at 35 °C and germinated for 48 h exhibited the highest moisture content (60.6%). However, from the results of the present study it is evident that germination of pearl millet may be possible at the low green malt moisture level in contrast to barley where 42-46% moisture is required. Comparatively low moisture level of green malt pearl millet in contrast to barley is possibly because of about one-third size with a proportionally larger germ and pericarp and hence, higher oil and insoluble fibre content, respectively (Serna-Saldivar and Rooney, 1995). Moisture content ranging from 33 to 60% has been reported by Pelembe *et al.* (2004) in green malt produced from different varieties of pearl millet after a germination period of 5 days. The moisture content of the green malt has been reported to be affected by the watering regime during the steeping (Pelembe *et al.*, 2004). It has been reported by Palmer (2006) that increasing germination moisture improved sorghum, pearl millet and barley malt quality in terms of diastatic power, FAN, hot water extract and malting loss. The moisture content of dried malt appeared to be affected non-significantly (P<0.05) by germination time but significantly by steeping temperature. However, the interactive effect of the two factors had significant (P<0.05) influence on the moisture content of dried malted grains.

Sprout length and roots and shoots weight

Sprout length was greatly affected by steeping temperature and germination time. The sprout length increased with increase in steeping temperature and germination period as shown in Table 3. Germination involves the outgrowth of the plumule and radical of the seedling until suitable enzymes (e.g. starch degrading enzymes and proteases) have been produced for the malt (Palmer, 1989). The highest sprout length of 4.25 cm was recorded when grains were steeped at 35 °C and germinated for 96 h. The percentage of roots and shoots in pearl millet malts were significantly (P<0.05) affected by germination time and steeping temperature as shown in Table 3. The percentage roots and shoots increased with germination time. The highest percentage of roots and shoots (13.63%) was recorded after steeping at 35 °C and germination for 96 h. The observed percentage roots and shoots values were comparable to those reported by Dewar *et al.* (1997).

Malting loss

An important aspect of malting with regard to the potential of pearl millet is malting loss. The malting loss was significantly (P<0.05) affected by germination time and steeping temperature. The malting loss (%) increased with increased germination period irrespective of steeping temperature as shown in Table 3. Regardless of the steeping temperature, highest malting loss of 21.9% was recorded after a germination period of 96 h. Ogundiwin *et al.* (1991) and Pelembe *et al.* (2004) also reported increased malting loss with increasing germination period. With regard to the effect of germination time on malting loss, the present findings are also in agreement with Dewar *et al.* (1997) and Nout and Davies (1982). Irrespective of

Table 3. Sprout length, root and shoot weight (%) and malting loss (%) of malted pearl millet grains prepared at different steeping temperature and germination periods.

Steeping temperature (°C)	Sprout length (cm)				Root and shoot weight (%)				Malting loss (%)			
	Germination period (h)			Mean A	Germination period (h)			Mean A	Germination period (h)			Mean A
	24 h	48 h	96 h		24 h	48 h	96 h		24 h	48 h	96 h	
25 °C	0.12±0.01	0.30±0.02	2.10±0.06	0.84	0.75±0.05	2.24±0.06	9.02±0.04	4.00	6.12±0.31	16.73±0.72	18.62±1.81	13.82
30 °C	0.31±0.03	1.95±0.05	2.90±0.05	1.72	1.43±0.07	3.43±0.18	8.61±0.19	4.49	8.00±0.80	16.20±0.10	19.70±1.16	14.63
35 °C	1.04±0.05	1.12±0.03	4.25±0.07	2.13	2.62±0.12	11.14±0.07	13.63±0.17	9.13	9.32±0.90	23.34±1.20	27.65±1.52	20.10
Mean B	0.49	1.12	3.08		1.60	5.60	10.42		7.81	18.75	21.99	

germination period malting loss increased with increase in steep temperature and maximum malt loss of 20.0% occurred at a steeping temperature of 35 °C. Dewar *et al.* (1997) reported a temperature of 25-30 °C to give relatively moderate malting loss. A significant interactive effect of steeping temperature and germination period was observed (P<0.05) and malt lost appeared to be highest (27.6%) at a steeping temperature of 35 °C and germination period of 96 h. According to Novellie (1962) and Pathirana *et al.* (1983), malting loss could be between 10.9 and 35% depending upon the malting conditions.

The malting loss increased with germination time due to the degradation of cell wall contents, mobilisation of endosperm matter and loss of dry matter as sprouts and volatiles (Briggs *et al.*, 1981).The studies have also correlated dry matter losses to the amylase activity and diastatic activity (Palmer, 1989). The growth of roots and shoots in the grain is indicator of the metabolic activities inside the grain. Higher roots and shoots growth implies a translocation of nutrients from the endosperm to them and hence responsible for more malt losses. The respiration losses are even more important where roots and shoots are not removed as in case of sorghum malt for the production of opaque beer (Beta *et al.*, 1995). In case of barley the roots and shoots are essentially removed from the malt and hence, accounts for great proportion in malting loss. Minimising malting loss is essential if pearl millet malting is to be economically viable (Pelembe *et al.*, 2004; Taylor *et al.*, 2006). In a study by Muller and Methner (2015), malting loss was measured in terms of monetary losses by comparing the differences in the dry weights of the malts between different samples. The monetary loss was observed to be € 2.64/t of malt considering a malt price of € 360/t.

Cold water extract

CWE (%) is used as criteria for evaluating grain malt quality. Indirectly, CWE indicates the extent of modification of the grain. CWE measures only water extract by preventing the enzyme action with ammonia. CWE was significantly affected by germination period, steeping temperature and their interactive effect as shown in Table 4. CWE increased with increasing germination time and steeping temperature up to 35 °C. Highest CWE (24.2%) was obtained at a steeping temperature of 35 °C and germination period of 96 h. CWE gives an indication of the amount of starch that has been hydrolysed during malting and is again an indication of modification. Extracting in cold water does not activate the enzymes, only the starch hydrolysed during

Table 4. Cold water extract, hot water extract (%) and free α-amino nitrogen of pearl millet malt as affected by varied conditions of steeping temperature and germination period.

Steeping temperature (°C)	Cold water extract (%)				Hot water extract (%)				Free α-amino nitrogen (100 mg/g)			
	Germination period (h)			Mean A	Germination period (h)			Mean A	Germination period (h)			Mean A
	24 h	48 h	96 h		24 h	48 h	96 h		24 h	48 h	96 h	
25 °C	3.53±0.3	9.15±0.6	17.40±0.6	10.02	2.35±0.9	34.45±1.2	43.54±0.3	26.78	51.22±5.6	86.67±3.0	118.92±2.2	85.60
30 °C	8.12±0.6	12.72±0.3	17.45±0.6	12.76	18.53±1.0	24.83±0.6	32.25±0.6	25.20	89.73±6.0	91.92±1.5	80.66±3.7	87.43
35 °C	6.59±0.3	15.31±0.6	24.23±0.2	15.37	19.91±0.9	40.80±0.6	45.20±0.6	35.30	74.15±5.3	124.11±3.1	116.04±4.5	104.7
Mean B	6.08	12.39	19.69		13.59	33.36	40.33		71.7	100.9	105.2	

malting leaches out. It was reported by Owuama and Ashino (1994) that increase in steeping moisture with steeping time around 24 h at steeping temperature 30 °C is accompanied by a corresponding increase in reducing sugar content in cold and hot water extracts.

Hot water extract

Malt HWE (%) is particularly important in lager beer brewing (Pelembe *et al.*, 2004). It determines the amount of beer that can be produced from a known quantity of malt. The percentage HWE was significantly (P<0.05) influenced by germination period, steeping temperature and their interactive effect (Table 4). HWE increased with increased germination period from 24 to 96 h regardless of the steeping temperature and it also increased with increased steeping temperature up to 35 °C. Similar results have been reported by Bekele *et al.* (2012) and Dewar *et al.* (1997). Hot water extract was highest (45.2%) at a steeping temperature of 35 °C and germination period of 96 h. The increase in HWE with increasing germination time is an indication of the progress of modification (breakdown of the endosperm reserves, predominantly by amylase and protease activity) of the malt during germination (Briggs, 1998). Genetics, biochemical components of the grain and malting conditions are all known to affect malt extract (Dewar *et al.*, 1997). HWE of millet is lower than the hot water extract of barley due to low diastatic power (Nzeribe and Nwasike, 1995).

Free α-amino nitrogen

FAN (mg/100 g) consists of free amino acids and small peptides, produced by proteases and peptidases activity of the malt. Malt FAN was significantly affected by steeping temperature, germination period and their interactive effect as shown in Table 4. FAN of the malt increased as the germination time increased. Similar observations have been reported by earlier researchers in case of pearl millet malting (Pelembe *et al.*, 2004) and sorghum malting (Bekele *et al.*, 2012). Evans and Taylor (1990) also reported increased proteolytic activity of sorghum malt with increased germination time. The highest level of FAN (124.1 mg/100 g) was recorded with a steeping temperature of 35 °C and germination period of 48 h. FAN levels of 130-150 mg/l were considered adequate to support yeast growth for higher fermentation output (Ogu *et al.*, 2006; Olatunji *et al.*, 1993). However, a typical FAN specification for sorghum malt for sorghum beer brewing has been reported to be a minimum of 110 mg FAN/100 g malt (Dewar *et al.*, 1997).

Diastatic power

The most important characteristics of good malt are high enzyme levels to degrade starch and obtain high extract yield. The DP (PMDU/g dry weight malt) is a measure of the joint α- and β-amylase activity and indicates the degree of starch conversion to fermentable sugar that can be extracted from the malt. Malts produced from the grains steeped at 30 and 35 °C gave the highest DP, which was significantly (P<0.05) affected by germination time, steeping temperature and their interactive effect as well (Table 5). DP of the pearl millet malt increased as the germination period increased from 24 to 96 h. The increase in steep temperature from 25 to 35 °C irrespective of germination period increased the DP of malt. However, this increase was non-significant when the temperature was increased from 30 to 35 °C. Dewar *et al.* (1997) also reported an increase in DP with an increase in steeping temperature up to 30 °C. The highest DP (39.8 PMDU/g) was obtained at a steeping temperature of 35 °C and the germination period of 96 h. According to Dewar *et al.* (1995), a minimum specification of the diastatic power of 28 SDU/g for malt derived from sorghum or millet is widely recommended for commercial beer brewing. Hence, with respect to DP pearl millet is suitable for sorghum beer brewing.

α-amylase activity

As with DP, α-amylase activity (PMDU/g dry weight malt) was also significantly affected by germination time and steeping temperature. The α-amylase activity increased significantly (P<0.05) up to 96 h of germination period as shown in Table 5. Similar to DP, the α-amylase activity increased significantly (P<0.05) when the steeping temperature was increased from 25 to 30 °C. The highest α-amylase activity (29.90 PMDU/g) was obtained when the grains were steeped at 30 °C and germinated for 96 h. The results of the present study are reasonably in agreement with the previous researchers (Muoria and Bechtel, 1998; Pelembe *et al.*, 2002). Muoria and Bechtel (1998) suggested that a germination temperature >22 °C would be more desirable for sorghum and pearl millet, than for barley, in order to obtain higher values of α-amylase. Pelembe *et al.* (2002) reported the highest amylase activity of 33 PMDU/g after 5 days of germination at 25 °C with a pearl millet variety. A temperature of 18 °C and possibly lower than this has been reported as optimal for barley (Briggs *et al.*, 1981). β-amylase plays crucial role during brewing and its production is influenced by the germination time and temperature. Pearl millet is tropical crop while barley is a temperate crop. Tropical cereals like pearl millet have little α-amylase but no β-amylase in ungerminated form and germination of tropical cereals leads to production of both amylases with α-amylase predominating (Dufour *et al.*, 1992; Palmer, 1989). Ungerminated temperate cereals like barley have moderate amount of β-amylase in ungerminated form and during germination α-amylase are synthesised and β-amylase, which is already synthesised during the development of grain, is rendered fully active during germination (MacGregor *et al.*, 1971).

Table 5. Diastatic power, α-amylase and β-amylase activity of pearl millet malt as affected by varied conditions of steeping temperature and germination period.

Steeping temperature (°C)	Diastatic power (PMDU/g)				α-amylase activity (PMDU/g)				β-amylase activity (PMDU/g)			
	Germination period (h)			Mean A	Germination period (h)			Mean A	Germination period (h)			Mean A
	24 h	48 h	96 h		24 h	48 h	96 h		24 h	48 h	96 h	
25 °C	7.72±0.7	17.81±1.3	39.04±0.6	21.52	5.65±0.6	15.67±1.3	30.15±0.6	17.15	3.32±0.6	7.80±0.6	12.28±0.6	7.80
30 °C	14.81±1.0	17.80±1.3	41.12±8.4	24.57	12.36±1.9	17.81±1.3	29.90±0.6	20.02	5.64±0.6	10.03±0.6	16.73±0.6	10.80
35 °C	13.32±1.3	21.06±2.0	39.80±1.3	24.72	13.30±1.3	15.53±1.3	21.05±0.6	16.62	3.37±0.6	12.10±0.6	12.15± 0.6	9.21
Mean B	11.95	18.89	39.98		10.43	16.33	27.03		4.11	9.97	13.72	

β-amylase activity

Both α- and β-amylases are required for hydrolysis of starch that has been gelatinised during malting. As with diastatic power and α-amylase, β-amylase activity (PMDU/g dry weight malt) was also significantly influenced (P<0.05) by germination period, steeping temperature and their interactive effect (Table 5). According to Morrall *et al.* (1986) steeping of sorghum grains for 18 to 22 h at 30 °C is optimal for enzymatic activity. The results of the present study also revealed the maximum activity of α- and β-amylase regardless of germination period at a steeping temperature of 30 °C. However, interaction effect showed the highest β-amylase activity (16.7 PMDU/g) after steeping at 30 °C followed by germination for 96 h. The starch-liquefying and dextrinising power is referred to α-amylase activity while the starch saccharifying or saccharolytic power is referred to as β-amylase activity (Dicko *et al.*, 2006). The β-amylase is usually bound to proteins and is released by proteolytic enzymes during germination (Guerin *et al.*, 1992).

4. Conclusions

This study revealed significant effects of steeping temperature and germination period on the quality characteristics of pearl millet malt. Based upon the gradient tests for various malting conditions, this study revealed high DP, α- and β-amylase activity, good FAN, and moderate malting loss at steeping temperature of 30-35 °C and germination for 4 days. The pearl millet malt perhaps even can have better potential than sorghum malt in lager beer brewing, at least as a barley malt extender, especially in areas where these grains are cultivated and cultivation of barley is limited.

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