J. Inst. Brew., November–December, 1993, Vol. 99, *pp.* 483–486

THE USE OF RAPID SCREENING TESTS TO COMPARE CHANGES DURING MALTING IN SORGHUM

By J. S. Swanston, K. Taylor

Scottish Crop Research Institute, Mylnefield, Invergowrie, Dundee DD2 5DA

N. S. Rao, V. Subramian

International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Patancheru, Andhra Pradesh 502 324, India

and D. S. Murty

ICRISAT, PMB 3491, Kano, Nigeria

Received 29 March 1993

Eight sorghum cultivars were malted and samples were taken after steeping and after each of 5 days germination. In genotypes with low levels of grain nitrogen, up to 35% of the nitrogen solubilisation during the malting process occurred in the steeping phase. After 5 days germination, extracts ranged from very low to moderate levels and correlated highly with diastase and half-grain mash results. In addition, genotypes producing higher extracts showed greater reductions in milling energy between 2 and 5 days germination. During malting, there was considerable genotype X day interaction for several quality characters, suggesting variation in rates of development as well as final quantities.

Key Words: *Sorghum, malting quality, diastase, milling energy*

Introduction

The use of malted sorghum, to replace barley malt in the brewing of lager beers, is a comparatively recent development, largely brought about by the Nigerian government's ban on imported barley, imposed in 1988[11]. However, opaque beers have been brewed, traditionally, in parts of Africa from sorghum and, in South Africa, the process has been developed commercially[12]. Some research has therefore been carried out into the physical and chemical changes occurring in the sorghum grain during malting and the effects which these have on the quality of the malt produced. It is known that, unlike barley, endosperm cell walls do not become totally degraded[6], but develop portals[14] through which enzymes can enter the cells. Starch breakdown occurs during malting, particularly in the floury endosperm[6], leading to extensive pitting of the granules, and considerable loss of potentially fermentable material to the developing rootlets and plumules[3].

There is variation between sorghum genotypes for malting quality attributes[10] and some success has been achieved in adapting rapid screening tests for barley malt to malted sorghum[21]. Recently it has been suggested that screening tests could also be combined to predict different patterns of modification in barley genotypes of varying malting quality[20]. If a similar exercise could be achieved in sorghum it could, in addition to identifying genotypes with the highest malting potential, indicate any which achieve a high level of quality in a shorter malting period. This would be of great advantage in reducing problems associate with mold development and malting loss[8], which increase as malting progresses.

Materials and Methods

Eight genotypes of sorghum, provided by the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Kano, Nigeria, were assessed for milling energy by means of a comparamill[1]. Duplicate samples were malted by the method of Swanston *et al.*[22], and sampled at completion of steeping and after each of 5 days germination. These malted sorghum samples were assessed for hot water extract, diastase activity and malt milling energy as previously described[22]. Soluble nitrogen was measured on the extracts, after dilution, by a spectrophotometric method[5], which is based on differences of absorption at 2 wavelengths in the UV spectrum. Data currently being prepared for publication has shown that, in sorghum, the correlation between soluble nitrogen determinations by this method and an established auto-analyzer technique[9] is extremely high ($r = 0.9$, $0.001 > p$).

In addition, 25 grains of malt were halved longitudinally and extracted in hot water in a half-grain mashing technique[13]. The modifications proposed by Gothard[7] were used to quantify the results. To allow for the higher gelatinisation temperature of sorghum, compared with barley[16], a temperature of 70°C was used.

As the techniques used here were primarily derived for rapid screening of large populations, some lack sufficient sensitivity to measure accurately very low values or distinguish between genotypes when differences are small. For that reason, this paper will, for the most part, concentrate on differences observed from the second day of germination onwards.

Results and Discussion

The genotypes studied showed a considerable range in both grain nitrogen contents and milling energies (Table I). Although the malt samples were subjected to prolonged extraction, i.e. 1 h at 45°C followed by 3 hr at 70°C, extracts, even after 5 days germination, ranged from very poor to moderate levels (Table II). These were lower than extracts observed in a previous population[22], or in a study by other researchers[10]. Diastase activities were also generally lower than those observed previously[21,22], when a close association between diastase and extract was detected. While a wide range of values was obtained for all malting parameters (Table II), even the highest diastase activities observed may have been inadequate to produce high levels of extract. Analysis of variance (Table III) indicated highly significant differences between genotypes and between days of germination for malting parameters, while good agreements between replicate values were obtained throughout. There was, however, also highly significant genotype x day interac-

*Submitted as J.A. # 1469 by the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT).

TABLE I. Grain nitrogen and grain milling energies of 8 sorghum cultivars

Cultivar No.	Designation	Grain Nitrogen (%)	Milling Energy (Joules)
1	ICSV 111	1.67	330.1
2	ICSV 400	1.94	286.4
3	ICSH 89001	1.46	264.0
4	ICSH 89002	1.44	284.6
5	CS 61	1.66	355.8
6	ISIAP Dora	1.61	311.8
7	Kaura	1.20	236.4
8	Fara-Fara	1.45	248.6

TABLE II. Results of tests for malting parameters on 8 sorghum cultivars between 2 and 5 days germination. Genotypes 1–8 are as given in Table I

Days Germ.	Genotype No. 1	2	3	4	5	6	7	8
				Extract (%)				
2	35.3	30.3	36.3	38.4	36.2	32.4	25.5	25.2
3	38.6	40.3	34.8	36.2	40.5	41.3	26.7	21.8
4	52.7	49.8	45.4	43.1	51.3	50.9	39.6	23.8
5	57.4	53.4	51.1	45.6	54.5	47.6	45.0	28.6
				Solule Nitrogen (mg/L)				
2	232.2	290.4	205.7	249.7	209.2	202.2	173.2	242.8
3	288.1	355.4	239.3	282.3	256.7	318.2	206.8	271.8
4	357.7	455.2	293.9	346.1	336.8	387.9	285.8	328.7
5	451.7	604.9	393.7	353.1	389.0	466.8	340.3	416.9
				Half-Grain Mash (%)				
2	10.8	12.1	12.6	10.2	9.3	10.5	7.1	8.8
3	17.5	22.4	14.5	18.8	19.8	18.2	12.7	12.2
4	22.6	23.1	17.5	18.2	21.9	21.0	14.1	12.9
5	29.9	24.6	23.6	21.1	22.2	21.2	15.2	13.8
				Milling Energy (Joules)				
2	260.9	212.6	201.6	200.5	271.1	227.4	169.3	164.5
3	214.6	158.1	166.0	169.2	218.1	181.7	149.8	153.8
4	178.1	142.0	139.5	161.0	193.8	160.3	133.9	146.6
5	164.0	130.9	146.3	153.0	175.2	144.9	124.6	142.3
				Diastase Activity (Enzyme Units)				
2	0.96	0.83	0.83	1.35	1.61	1.15	0.91	0.87
3	3.08	4.19	2.13	3.08	2.61	3.16	2.21	1.74
4	4.66	5.85	4.43	4.19	4.27	4.42	3.56	2.06
5	5.69	5.93	4.43	4.09	4.87	4.11	4.19	3.08

tion for all characters, as would be expected from the data in Table II, where the ranking order of genotypes did not remain constant. This is in contrast with the situation previously observed in barley, where, in general, the ranking order of cultivars, for factors such as hot water extract and malt milling energy, remains the same throughout malting[19]. On this basis it was proposed that milling energy scores, after 1 or 2 days germination, could be used to predict barley malting potential[18]. Such a prediction is unlikely in sorghum, so breeders will probably have to rely on actual measurements made after the required malting period. However, the results here also suggest that there may be variation in the rate at which malting parameters reach a given level, so selection for genotypes which malt more rapidly may be possible.

Correlations between parameters measured at the completion of malting (Table IV) indicate a close relationship between extract and diastase activity, which confirms observations on a previous population[21]. A highly significant correlation was also observed between extract and half-grain mash results. In barley, it was suggested that breakdown of both the cell walls and protein matrix facilitated the removal of cell contents by hot water[13] and led to high scores in the half-grain mash test. In sorghum, cell walls are not removed[6] so half-grain mash results are presumably associated with other structural changes. Results here showed no significant correlation between half-grain mash and soluble nitrogen (Table IV), although half-grain mash results did correlate significantly with diastase activity.

Sorghum has distinct areas of vitreous and floury endosperm, which differ in physical structure[15]. In the floury endosperm, the starch granules are contained fairly loosely and would require comparatively little protein degradation to release them. By contrast, in the vitreous endosperm,

TABLE IV. Correlation matrix showing relationships between malting parameters in 8 genotypes of sorghum after 5 days germination

	Extract	Soluble Nitrogen	Malt Milling Energy	Diastase Activity
Soluble Nit.	0.278 NS			
Malt Mill. En.	0.386 NS	−0.209 NS		
Diastase	0.873**	0.601 NS	0.181 NS	
Half-Grain Mash	0.858**	0.431 NS	0.486 NS	0.831*

* = $0.05 > p > 0.01$
** = $0.01 > p > 0.001$
NS = Not significant at the 5% level

TABLE III. Analysis of variance for malting parameters measured on 8 sorghum cultivars

	df	Extract MS	Soluble Nitrogen MS	Half-Grain Mash MS	Milling Energy MS	Diastase Activity MS
Between:						
Reps	1	4.7 NS	760 NS	2.8 NS	15.8 NS	0.2 NS
Cult.	7	412.1***	21432***	82.8***	4858.0***	3.4***
Days	3	909.3***	122911***	373.2***	13609.0***	41.4***
C × D	21	29.3***	1787**	10.6***	310.7***	0.6**
Res.	31	8.2	521	2.6	49.5	0.2
Total	63					

** = $0.01 > p > 0.001$
*** = $0.001 > p$
NS = Not significant at the 5% level

starch granules and protein bodies are bound much more tightly into the protein matrix and extensive proteolysis may be necessary to disrupt the structure[15]. Consequently, the relationship between half-grain mash results and soluble nitrogen content may be greatly influenced by the endosperm texture of the cultivars studied. However, in sorghum as in barley, the largest single component of the endosperm is starch and this will account for most of the material removed in half-grain mashing. Damage to the granules greatly enhances solubility[4], so granules partially degraded by diastase activity may be preferentially extracted. Thus, high levels of diastase may contribute to higher half-grain mash results.

In a recent report it was shown that clear distinctions between sorghum cultivars, for diastase activity, could not be made before 48 h germination[17]. A similar result was obtained here, although this could have been due to the fact that all samples were extracted in the same manner and a more concentrated extract could perhaps have detected smaller differences. Substantial increases in diastase activity were therefore observed between 2 and 5 days germination. During that period there was also a large reduction in milling energy with considerable variation between genotypes. When the milling energy after 2 days germination is presented as a percentage of the milling energy at 5 days (Fig. 1), it is seen that the smallest reductions are associated with the genotypes 4, 7 and 8 which gave lowest levels of extract. These also had low levels of diastase activity (Table II).

A proportion of the grain nitrogen was solubilised during malting. Of that proportion between 40 and 70% had been solubilised by the end of the second day of germination, depending on the cultivar (Figure 1). Results for half-grain mashing indicated an approximately similar range of values for results at day 2 presented as a percentage of those at day 5, but there was little evidence of a relationship between the two parameters in the genotypes studied. Genotypes 4 and 8, which gave the lowest proportion of nitrogen solubilisation between 2 and 5 days, however, also showed low levels of milling energy reduction during that period.

Asien[2] reported that β-glucanase activity was present in the resting grain of sorghum and, unlike other enzymes, did not demonstrate a large increase during malting. It is therefore likely that the limited cell wall breakdown will begin very early in the malting process, permitting access of other enzymes and disappearance of the protein matrix is reported to begin during steeping[6]. This appeared to be confirmed by results here (Table 5), which indicated that up to 35% of the protein solubilisation during malting occurred in the steeping phase. Considerable variation excited within the eight cultivars for this character and could, in theory, be due to differences in cell wall modification affecting the rate of access of proteases. However, it was clear that those samples in which large proportions became solubilised during steeping, i.e. genotypes 3, 4 and 7 and 8, had the lowest levels of grain nitrogen (Table I). This may indicate that, as grain nitrogen levels increase, a greater proportion is associated with the matrix and protein bodies in the vitreous endosperm and is less likely to be solubilised early in the malting process.

Glennie *et al.*[16] reported that, within the vitreous endosperm, protein bodies retained their closely packed arrangement until a large amount of matrix was extensively broken down. Degradation of starch granules and protein bodies then became simultaneous, with starch granules retaining their shape until extensive pitting led to collapse. As the cell walls remained substantially intact throughout, it is likely to be this disruption of the internal cell structure which leads to a reduction in milling energy. The relationship between milling energy reduction, nitrogen solubilisation and diastase activity, especially during the latter stages of malting, is likely, therefore, to be important in determining patterns of endosperm modification in sorghum. Future research, with larger populations, will concentrate on these areas.

TABLE V. Proportions of nitrogen solubilised during malting occurring in 3 stages of the process, in 8 sorghum genotypes

Genotype Number	Proportion of Nitrogen		
	Steeping	Germination Days 1–2	Germination Days 3–5
1	17.5	33.6	48.8
2	13.9	34.1	52.0
3	35.8	16.5	47.7
4	33.4	37.3	29.3
5	24.7	29.1	46.2
6	18.2	25.1	56.7
7	29.6	21.3	49.1
8	30.9	27.3	41.8

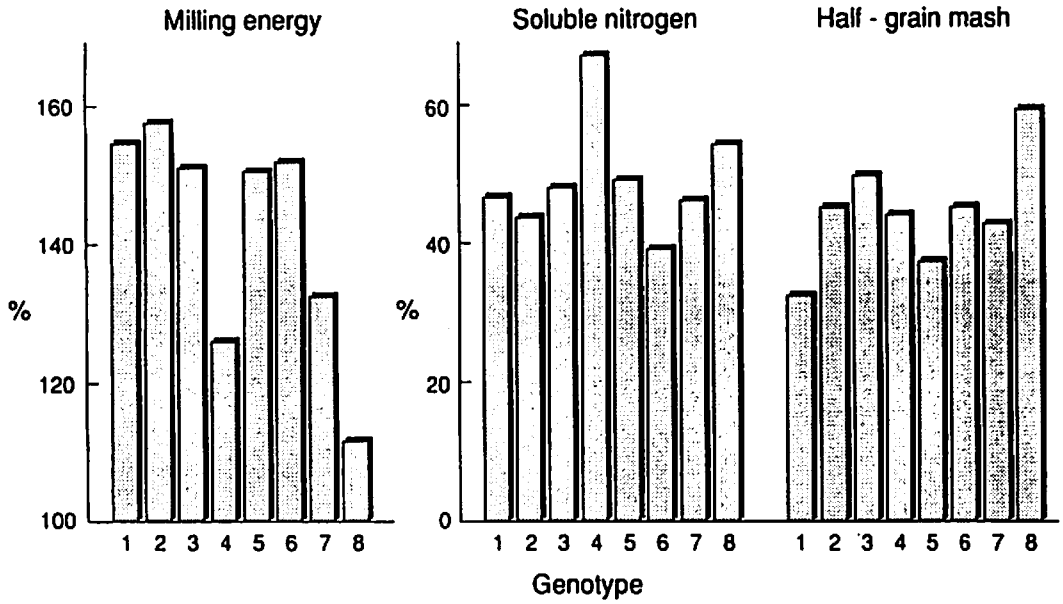

FIG. 1. Milling Energy, soluble nitrogen and half-grain mash results for 8 sorghum cultivars, after 2 days germination, as a percentage of the results for the same parameters after 5 days germination.

References

1. Allison, M. J., Cowe, I. A., Borzucki, R., Bruce, F. M. & McHale, R., *Journal of the Institute of Brewing*, 1979, **85**, 262.
2. Asien, A. O., *Journal of the Science of Food and Agriculture*, 1982, **33**, 754.
3. Asien, A. O. & Muts, G. C. J., *Journal of the Institute of Brewing*, 1987, **93**, 328.
4. Craig, S. A. S. & Stark, J. R., *Starke*, 1984, **36**, 127.
5. Franken-Luykx, J. M. M., *Journal of the Institute of Brewing*, 1967, **73**, 187.
6. Glennie, C. W., Harris, J. & Liebenberg, N. V. D., *Cereal Chemistry*, 1983, **60**, 27.
7. Gothard, P. G., in Barley Genetics IV, *Proceedings of the Fourth International Barley Genetics Symposium, Edinburgh*, 1981, 242.
8. Ikedobi, C. O., in Industrial Utilisation of Sorghum, *Summary Proceedings of the Symposium on the Current Status and Potential of Industrial Uses of Sorghum in Nigeria*, 1990, 32.
9. Jambunathan, R., Rao, N. S. & Gurtu, S., *Cereal Chemistry*, 1983, **60**, 192.
10. Jayatissa, P. M., Pathirana, R. A. & Sivayogasunderam, K., *Journal of the Institute of Brewing*, 1980, **86**, 18.
11. Koleoso, O. A. & Olatunji, O., in *Utilisation of Sorghum and Millets*, 1992, 41.
12. Novellie, L., *Wallerstein Laboratory Communications*, 1968, **31**, 17.
13. Palmer, G. H., *Journal of the Institute of Brewing*, 1975, **81**, 408.
14. Palmer, G. H., *Options Mediterraneennes, Series A No. 20, New Trends in Barley Quality for Malting and Feeding*, 1991, 19.
15. Rooney, L. W. & Miller, F. R., *Proceedings of the International Symposium on Sorghum Grain Quality, ICRISAT Centre, Patancheru, India*, 1981, 143.
16. Stark, J. R., Asien, A. O. & Palmer, G. H., *Starke*, 1980, **35**, 73.
17. Subramanian, V., Murty, D. S., Rao, N. S. & Jambunathan, R., *Journal of the Science of Food and Agriculture*, 1992, **58**, 35.
18. Swanston, J. S., *Journal of the Institute of Brewing*, 1990, **96**, 209.
19. Swanston, J. S. & Taylor, K., *Journal of the Institute of Brewing*, 1988, **94**, 311.
20. Swanston, J. S., Taylor, K., Camm, J-P. & Ellis, R. P., *Journal of the Institute of Brewing*, 1992, **98**, 493.
21. Swanston, J. S., Taylor, K. & Murty, D. S., *Proceedings of the EUCARPIA (Cereal Section) Meeting, Schwerin*, 1991, 334.
22. Swanston, J. S., Taylor, K. & Murty, D. S., *Journal of the Institute of Brewing*, 1992, **98**, 129.