



Original Research Article

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Optimization of Malting and Mashing Conditions of Sweet Sorghum Grains as a Brewing Source

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Sweet sorghum grains have high starch content which can be converted to sugars by malting and mashing processes. Three sweet sorghum cultivars 'SSV-74', 'SSV-84' and 'SSV-108' and a sorghum cultivar 'SSV-2' were screened for their amylase activity and reducing sugars. Grains were soaked at three different time intervals of 8h, 12h and 16h and germinated subsequently for 2 and 3 days. The highest amylase activity (1266.10 µg of protein/15 min/g of sample) was recorded in sweet sorghum cultivar 'SSV-84' soaked for a time interval of 16h and germinated for 3 days. Similarly, 'SSV-84' recorded the highest reducing sugars (33.85 mg/g) at a soaking time of 16h and a germination period of 3 days. Commercial α-amylase (Palkozyme) was used at different concentrations of 0.1, 0.5 and 1 per cent at different incubation temperature and incubation period for release of maximum reducing sugars. The highest reducing sugars (78.83 mg/g) was recorded in variety 'SSV-84' at an enzyme concentration of 1 per cent at 70°C for an incubation period of 24h.

Introduction

The name "Sweet sorghum" is used to identify varieties of sorghum (*Sorghum bicolor* (L.) Moench) those are sweet and juicy. These sweet stalk varieties are currently grown for sugar production, forage and silage. White sugar and jaggery was produced from it in India in 1975 (Bhise *et al.*, 1988). Sweet sorghum has become a leading contender in biomass production for energy systems because of its high yield, per cent of fermentable sugars and its adaptability to

drought areas. It could produce 50 metric tonnes of crushable stalks, 3 metric tonnes of grains and 17 metric tonnes of leaves and stem tops per hectare (Almodares *et al.*, 1994). Its rapid growth and ability to reach maturity in 3 and 5 months, when coupled with its lack of photoperiodism are favourable for its production on fallow sugarcane land, primarily because it can be grown, and harvested before the start of the sugarcane harvesting season. Sweet sorghum stalks, containing high sugars have been readily exploited for fuel ethanol. Furthermore, sweet

sorghum is rich in micronutrients and minerals (Seetharama *et al.*, 2002). The grains on the other hand are rich source of carbohydrates which can be converted into sugars by the action of endogenous enzymes activated during sprouting. The grains of sweet sorghum are rich source of carbohydrates which can be converted into sugars by the action of endogenous enzymes activated during sprouting. This could be further diverted towards beer production. Rothschild (1972) used plain sorghum, sprouted sorghum and maize grits for the production of Bantu beer in South Africa and obtained ten million litres of beer during the first month of operation. Satyanarayana and Narasimham (1975) advocated the use of 40 per cent sorghum as an adjunct in brewing and concluded that it compared very favourably with beer made from 100 per cent malted barley. Also, in the tropical countries, barley has to be imported from other temperate countries and this involves expenditure of scarce foreign exchange. Besides, sweet sorghum also requires minimum fertilizer inputs than barley. Hence, sweet sorghum could be a new potential substitute for barley and could raise economic benefits. Thus, the present investigation lies in optimising different parameters of malting and mashing of sweet sorghum grains.

Materials and Methods

The sweet sorghum varieties selected for beer production were 'SSV-74', 'SSV-84' and 'SSV-108' and a sorghum variety 'DSV-2' which were procured from AICRP on sorghum, UAS, Dharwad.

The grains, were soaked at different time interval of 8h, 12h and 15h in normal tap water and subsequently germinated at two different periods of 2 days and 3 days in clean muslin cloth. Amylase activity and reducing sugars were determined. The malt was kilned

at 50°C in hot air oven for 24h. The kilned grains were further broken into grits by running through a blender at low speed. Mashing was carried out with a grist to water ratio of 1:3 at a temperature of 45°C for 15 minutes and 60°C for one hour along with commercial α -amylase (Palkozyme, courtesy M/S maps – India Ltd. Ahmedabad). The enzyme concentrations of 0.1, 0.5 and 1 per cent were tested for saccharification of the substrate. The incubation temperature levels of 30°C, 50°C and 60°C and different incubation periods of 8, 16 and 24 h were also used for standardizing the optimum activity of commercial enzymes. The best sorghum variety and the optimised parameters for malting and mashing were used for wort preparation. The wort obtained after double filtering the mash, was boiled for an hour. For flavouring, hops in the form of pellets (acid extracted, courtesy: M/S U.B. Breweries, Mangalore) was added @ 40 ppm after 30 minutes of boiling. The pH, total soluble solids (TSS), tannin content and reducing sugars were estimated.

Amylase activity was estimated by the method described by Bernfield (1955). Reducing sugars were determined by 3,5-dinitrosalicylic acid method (Miller, 1959). The pH meter of Analog model (Corion Research, USA) was used to record the pH of wort. Total soluble solids (TSS) was determined with the help of ERMA hand refractometer having range of 0-32°Brix at 20°C. Folin-Denis method was used for estimation of tannins (Schanderl, 1970). The experimental results were statistically analyzed as per Fisher's analysis of variance technique (Panse and Sukhatme, 1961).

Results and Discussion

The amylase activity and reducing sugars released in the grains soaked at different soaking period and germination period are

presented in Table 1. Significantly high amylase activity (1213.23 µg of protein/15 min/g sample) and reducing sugars (28.04 mg/g) was obtained on the third day of germination. Among the soaking period, 16 h of soaking recorded the highest amylase activity (1239.76 µg of protein/15 min/g sample) and reducing sugars (30.98 mg/g). Among the different varieties used, sweet sorghum 'SSV-84' recorded highest amylase (1266.10 µg of protein/15 min/g sample and reducing sugars (31.51 mg/g), while sweet sorghum SSV-74 showed the least amylase activity and reducing sugars. These results clearly indicate that the variety 'SSV-84' with a soaking period of 16 h and germination period of 3 days in optimum for malting. The difference in amylase activity and reducing sugars may be because of the soluble fraction of amylase enzyme that increases during germination upto certain extent releasing more sugars. Uvere *et al.*, (2000) determined alpha amylase activity in one red and white varieties of sorghum, steeped in water for 18 h and germinated upto 5 days and found that α -amylase activity peaked on third day of germination in white sorghum variety (550 µ/100 g).

The results of the effect of enzyme concentration, incubation temperature and incubation period of commercial α -amylase on release of reducing sugars are presented in Table 2, 3 and 4. The grains treated with 1 per cent of commercial enzymes released maximum reducing sugars (70.79 mg/g)

(Table 2). In case of incubation temperature, the highest reducing sugars was recorded at 70°C (68.52 mg/g (Table 3) which did not show any significant difference with the reducing sugars obtained at 50°C (68.42 mg/g) and were on par with each other (Table 3). Among the incubation periods, used for maximum hydrolysis of the substrate (Table 4), incubation period of 24 hours recorded high reducing sugars than other treatment (72.02 mg/g) (Table 4). Among the varieties, sweet sorghum SSV-84 recorded the highest reducing sugars in all the treatments tested for enzyme optimisation. The variation in reducing sugars among the varieties with commercial enzyme treatment could be due to the fact that the reaction of enzymes is based on the composition of the grains. High glucose content in sorghum malt wort was obtained by the addition of amyloglucosidase at a concentration of 0.59 g/L of wort and a temperature of 55°C during mashing (Pozo-Insfran *et al.*, 2004). All optimised parameters for malting and mashing were used for wort preparation. The pH of 5.20, 7.55 of 14.50° Brix, tannin content of 13.15 mg/100 ml and reducing sugars of 73.72 mg/g was recorded in the wort (Table 1A).

The present investigation clearly revealed that the grains of sweet sorghum variety 'SSV-84' soaked for 16h and germinated for a period of 3 days gave the highest amylase activity and reducing sugars compared to other treatments, thus indicating its high potential as a malt source for brewing process.

Table.1A Chemical analysis of wort prepared from sweet sorghum variety (SSV-84) for beer production

Parameters	Values
pH	5.20
Total Soluble Solids (TSS)	14.50 °Brix
Tannin content	13.15 mg/100 ml
Reducing sugars	73.72 mg/g

Table.1 Amylase activity and reducing sugars of 2 and 3 days germinated grains at different soaking time

Varieties	AMYLASE ACTIVITY (µg of protein/15 min/g of sample)								REDUCING SUGARS (mg/g)							
	2 days of germination				3 days of germination				2 days of germination				3 days of germination			
	8 h	12 h	16 h	Mean	8 h	12 h	16 h	Mean	8 h	12 h	16 h	Mean	8 h	12 h	16 h	Mean
SSV-74	1125.03	1163.36	1200.66	1163.02	1168.40	1186.30	1234.00	1196.23	21.70	24.45	25.55	23.90	23.26	25.55	28.43	25.75
SSV-84	1225.83	1240.66	1260.43	1242.31	1240.10	1251.26	1266.10	1252.48	25.38	30.60	31.51	29.16	28.80	31.46	33.85	31.37
SSV-108	1154.20	1177.40	1212.80	1181.46	1156.86	1182.76	1219.70	1186.44	23.10	25.16	27.98	25.41	23.83	27.03	29.70	26.85
DSV-2	1195.16	1206.46	1235.60	1212.41	1202.40	1211.63	1239.26	1217.76	23.13	27.35	30.73	27.07	24.16	28.48	31.96	28.20
Mean	1175.05	1196.97	1227.37	1199.80	1191.94	1207.99	1239.76	1213.23	23.32	26.89	28.94	26.38	25.01	28.13	30.98	28.04
Source	SEm±				CD at 1%				SEm±				CD at 1%			
Varieties (V)	6.059				22.989				0.274				1.039			
Soaking time (S)	5.247				19.908				0.237				0.899			
Germination period (G)	4.284				16.254				0.194				0.736			
VS	10.495				NS				0.475				1.802			
VG	8.569				NS				0.388				NS			
SG	7.421				NS				0.336				NS			
VSG	14.843				NS				0.672				NS			

NS – Non significant

Table.2 Optimization of enzyme concentration for saccharification by commercial α -amylase

Sl. No.	Varieties	Reducing sugars (mg/g)				
		Concentration of α -amylase (%)				
		0.1	0.5	1.0	Mean	Control (without amylase enzyme)
1.	SSV-74	48.00	57.74	65.92	49.77	27.44
2.	SSV-84	63.32	67.32	73.42	59.06	32.20
3.	SSV-108	49.36	62.21	71.64	53.33	30.14
4.	DSV-2	50.18	65.60	72.18	54.67	30.71
	Mean	52.71	63.22	70.79	54.21	30.12
	Source	S.Em \pm		CD at 1%		
	Varieties (A)	0.435		1.771		
	Concentration (B)	0.376		1.533		
	Interaction (A x B)	0.753		3.067		

Table.3 Optimization of incubation temperature for commercial α -amylase

Sl. No.	Varieties	Reducing sugars (mg/g)				
		Incubation temperature ($^{\circ}$ C)				
		30	50	70	Mean	Control (without amylase enzyme at ambient temperature)
1.	SSV-74	56.64	63.74	63.81	53.01	27.86
2.	SSV-84	64.14	72.35	72.38	60.45	32.92
3.	SSV-108	59.32	67.10	67.17	55.74	29.36
4.	DSV-2	62.73	70.51	70.71	58.60	30.46
	Mean	60.70	68.42	68.52	56.95	30.15
	Source	S.Em \pm		CD at 1%		
	Varieties (A)	0.572		2.331		
	Concentration (B)	0.495		2.018		
	Interaction (A x B)	0.991		NS		

Ambient temperature = Minimum = 13.8° C
 Maximum = 28.2° C

Table.4 Optimization of incubation period for commercial α -amylase

Sl. No.	Varieties	Reducing sugars (mg/g)				
		Incubation period (h)				
		8	16	24	Mean	Control (without amylase enzyme)
1.	SSV-74	51.18	55.29	66.62	50.30	28.10
2.	SSV-84	60.38	65.04	78.72	59.15	32.47
3.	SSV-108	50.06	57.82	69.31	51.73	29.74
4.	DSV-2	56.26	61.17	73.43	55.71	31.99
	Mean	54.47	59.83	72.023	54.22	30.58
	Source	S.Em \pm		CD at 1%		
	Varieties (A)	0.276		1.126		
	Concentration (B)	0.239		0.975		
	Interaction (A x B)	0.479		1.951		

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