

Full Length Research Paper

Comparison of the mashing and brewing potentials of crude extracts of *Abrus precatorius*, *Burnatia enneandra* and *Cadaba farinosa*

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The mashing and brewing potentials of crude extracts of three plants commonly used in Northern Cameroon to make starch gruels sweet were assessed using sorghum cultivar *Safrari* as an adjunct. Alpha-amylase activities of the extracts fluctuated between 18 and 122 U/g, while the diastatic power and the β -carboxypeptidase activities ranged between 83 and 123 WK, and 16 and 63 mg FAN/min/ μ L, respectively. A longer mashing time (180 min) was necessary to obtain higher yields of extracts. Extract obtained was 18.7 °P for malted *Safrari*, 14.6 °P for malted barley, and 9.5, 13.5 and 11.4 °P after mashing *Safrari* adjuncts with extracts from *Abrus precatorius*, *Burnatia enneandra* and *Cadaba farinosa*, respectively. Similar levels of reducing sugars ranging between 60 and 65 g/L were obtained when mashing was carried out using *A. precatorius*, *B. enneandra* and crude extracts, but about 50 g/L were obtained for *C. farinosa*. Free amino nitrogen fluctuated between 56 and 72 mg/L for *C. farinosa* and *B. enneandra*, respectively, but was as low as 17 mg/L for *A. precatorius*. Fermentability was low for the worts obtained after mashing *Safrari* adjuncts using the extracts of the three plants, as compared to the worts of malted sorghum cultivar *Safrari* and barley.

Key words: *Abrus precatorius*, *Burnatia enneandra*, *Cadaba farinosa*, α -amylase, extract, free amino nitrogen (FAN).

INTRODUCTION

The use of technical enzymes has tremendously facilitated and improved food processing for the past decades (Hudson, 1986; Delrue, 1987; Law, 1990). Mashing with high amounts of adjuncts or with poorly modified barley malts using exogenous enzymes in the brewing industry improves yields in extracts (MacFadden and Clayton, 1989; Dale et al., 1990; Bajomo and Young, 1992; Agu and Palmer, 1998; Goode et al., 2002; Goode and Arendt, 2003; Goode et al., 2003; Desobgo et al., 2010). Malted sorghum is known to have low levels of important mashing enzymes such as α -amylase and β -amylase, but

mash easily when supplemented with adequate commercial enzymes (Arri, 1990; EtokApan and Palmer, 1990; Dufour and Melotte, 1992; Nso et al., 2003; Nso et al., 2006). These enzymes and others are extracted from microorganisms and plants and commercialized as technical enzymes.

In Northern Cameroon, sorghum, maize and rice based starch gruels are traditionally made sweet using crude extracts of the plants *Abrus precatorius*, *Burnatia enneandra* and *Cadaba farinosa* (Glew et al., 2010). In this paper, we assessed the mashing and brewing

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potentials of crude extracts of these plants using unmalted *Safrari* sorghum as an adjunct.

MATERIALS AND METHODS

Biological materials

Sorghum cultivar *Safrari* was obtained from the Institute of Agronomic Research and Development (IRAD) in Maroua, Cameroon. Barley malt was obtained from "Société Anonyme des Brasseries du Cameroun" (SABC) in Garoua, Cameroun. *B. enneandra* and *C. farinosa* were obtained from Yagoua and *A. precatorius* from Ngaoundere, Cameroun.

Obtaining crude enzyme extracts

A. *precatorius* and *C. farinosa* extracts

The leaves and tender stems of these plants were harvested, chopped using a knife and air dried at 40°C for five days using a CKA 2000 AUF-type dryer Ngaoundere, Cameroun. The chips were ground into coarse particle sizes using a Fryma machine AG mill type ML-150 (CH-4310 Rheinfelden, Switzerland) and next into fine particle sizes (≤ 1 mm) using a laboratory hammer mill of model Polymix PX-MFC 90D apparatus type (VWR International S.A.S. Le Périgas 201, rue carnot, 94126 Fontenay-sous-Bois Cedex, France). Fifty milliliters of 0.2 M phosphate buffer (disodium-potassium di hydrogen phosphate)/pH 6 were added to 10 g of the powder and agitated after every 10 min for 1 h. The mix was then filtered using a Whatman paper No. 42. The filtrate was retained as the enzyme extract for liquefaction and saccharification of sorghum grits during mashing.

B. *enneandra* extract

B. enneandra nuts were washed clean repeatedly with tap water. The fibrous coating was scrapped off with a knife and the kernels obtained by cracking of the nuts. The kernels were then chopped into small chips. They were dried at 40°C for five days and enzyme extract was obtained as described above for *A. precatorius* and *C. farinosa*.

Determination of the α -amylase activity of the extracts

Alpha-amylase activity of the extracts was determined using the 3,5-dinitrosalicylic acid (DNS) method of Fischer and Stein (1969).

Determination of the diastatic power of barley and *Safrari* malts, *A. precatorius*, *C. farinosa* and *B. enneandra* powders

Diastatic power was determined according to the method described by Analytica-EBC (2006).

Determination of carboxypeptidase activity

The carboxypeptidase activities of the plant extracts, barley and *Safrari* malts, were obtained by the method described by Okolo and Ezeogu (1995).

Mashing

Mashing unmalted *Safrari* grits using crude enzyme extracts

Two hundred milliliters of distilled water were put into a 600 mL beaker and 50 g of sorghum flour (≤ 1 mm) was added with continuous stirring until a homogenous mixture was obtained. This mixture was incubated at 45°C for 30 min in a water bath with intermittent stirring at intervals of 5 min. The temperature of the mash was then raised to boiling so as to gelatinize sorghum starch during 40 min with intermittent stirring at intervals of 5 min before cooling to 65°C. Fifty milliliters of crude enzyme extracts were added to the mash and allowed to incubate for 1 h 30 min with intermittent stirring at intervals of 10 min.

Mashing of *Safrari* malt grist

Mashing was carried out as described above but 250 mL of distilled water were used. A portion of the supernatant (50 mL) was removed after 30 min decantation of the mix at 45°C. Starches in the endosperms tissue of the sorghum grits was gelatinized as described above. Decanted enzymic supernatant fractions (50 mL) were added to appropriate gelatinized sorghum mashes after they were cooled to 65°C. Mashing time for this mash mixture was 1 h, 30 min. The mashes were stirred at intervals of 10 min.

Mashing of barley malt grits

Mashing was done as described for *Safrari* malt grits with omission of boiling mash. The mashes were filtered during 1 h using Whatman paper No 42.

Determination of reducing sugars

Reducing sugars were determined according to the DNS method of Bernfeld (1986).

Determination of free amino nitrogen

Free amino nitrogen (FAN) was determined as described by Analytica-EBC (2006).

Determination of attenuation limit

Apparent fermentability of wort was determined according to the method described by Analytica-EBC (2006).

RESULTS AND DISCUSSION

The enzymatic brewing potential (α -amylase, diastatic power and β -carboxypeptidase) of malted cereals for mashing is an important criterion for accepting such raw material for beer brewing (Briggs et al., 2004). This enzymatic brewing potential of crude extracts of the plants *A. precatorius*, *B. enneandra* and *C. farinosa*, as well as the malted sorghum cultivar *Safrari* and barley, as reference malt, are compared in Table 1. Alpha-amylase

Table 1. Alpha-amylase and β -carboxypeptidase activities, and diastatic power of crude extracts of *A. precatorius*, *B. enneandra* and *C. farinosa* and malts.

Property	Extract type				
	<i>A. precatorius</i>	<i>B. enneandra</i>	<i>C. farinosa</i>	Malted <i>Safrari</i>	Malted barley
α -Amylase activity(U/g)	34.04 \pm 0.00	122.37 \pm 0.02	18.27 \pm 0.01	181.16 \pm 0.48	350,88 \pm 0.26
DP (WK)	83.43 \pm 0.10	122.47 \pm 0.25	109.52 \pm 0.33	273.32 \pm 0.18	483.84 \pm 0.13
β -Carboxy-peptidase (mg FAN/min/ μ L)	67.62 \pm 0.06	16.84 \pm 0.02	20.68 \pm 0.03	72.53 \pm 0.22	58.34 \pm 0.04

Table 2. Effect of duration of mashing unmalted *Safrari* grits using the crude enzyme extracts of *A. precatorius*, *B. enneandra* and *C. farinosa* on the wort extracts.

Sample type	Extract ($^{\circ}$ P)	
	Mashing time (min)	
	90	180
Unmalted <i>Safrari/A. precatorius</i>	7.8 \pm 0.11	9.5 \pm 0.05
Unmalted <i>Safrari/B. enneandra</i>	8.8 \pm 0.13	13.5 \pm 0.21
Unmalted <i>Safrari/C. farinosa</i>	8.0 \pm 0.30	11.4 \pm 0.15
Malted <i>Safrari</i>	16.6 \pm 0.11	18.7 \pm 0.11
Malted Barley	13 \pm 0.09	14.6 \pm 0.04

Table 3. Reducing sugars, FAN and fermentability after mashing *Safrari* and barley malts, and unmalted *Safrari* using crude extracts of *A. precatorius*, *B. enneandra* and *C. farinosa*.

Wort characteristics	Wort type				
	<i>Safrari</i> malt	Barley malt	Unmalted <i>Safrari/A. precatorius</i>	Unmalted <i>Safrari/B. enneandra</i>	Unmalted <i>Safrari/C. farinosa</i>
Reducing sugars (g/L)	34.29 \pm 0.18	83.40 \pm 0.09	64.31 \pm 0.03	60.34 \pm 0.15	49.56 \pm 0.33
FAN (mg/L)	139.14 \pm 0.01	173.27 \pm 0.13	17.18 \pm 0.02	71.60 \pm 0.01	55.85 \pm 0.03
Fermentability (%)	58.2	58.2	41.6	45.8	37.5

activity of the plant extracts was highest for *B. enneandra*, followed by *A. precatorius* and then *C. farinosa*. The importance of this activity is in the ratio of 100:28:15%, respectively. This activity was however highest for barley malt extract, about two times that of malted *Safrari* extract, and three times that of *B. enneandra* extract. Similarly, diastatic power of the extracts was in the ratio of 100:89:68% for *B. enneandra*, *C. farinosa* and *A. precatorius*, respectively. It was however highest for barley extract, about two times that of malted *Safrari* extract and four times that of *B. enneandra*. The ratio for β -carboxypeptidase activity is, 100:30:25% for *A. precatorius*, *C. farinosa* and *B. enneandra*, respectively. The activity of this enzyme for *A. precatorius* extract is comparable to that of malted *Safrari* and barley malt. Though the enzymatic potential of these crude extracts are lower than that for malted barley and *Safrari*, longer mashing periods or highly concentrated purified extracts will help obtain acceptable mashing performances.

The importance of time (90 and 180 min) on yields of extracts when mashing unmalted *Safrari* grits using the crude enzyme extracts of *A. precatorius*, *B. enneandra* and *C. farinosa* was also compared. The results obtained (Table 2) show that yields of extracts were higher after mashing for 180 min as compared to mashing for 90 min. The greatest impact of mashing time on yields of extracts was obtained for unmalted *Safrari/B. enneandra* sample type, a difference of about 5 $^{\circ}$ P. A difference of about 2 $^{\circ}$ P was obtained for unmalted *Safrari/A. precatorius*, *Safrari* and barley malts was obtained and about 3 $^{\circ}$ P for unmalted *Safrari/C. farinosa* sample type. A mashing time of 180 min was therefore generally considered appropriate if higher yields of extract are to be obtained particularly when mashing with *B. enneandra* extracts.

Sugars and free amino nitrogen (FAN) are decisive in the fermentability of worts during beer brewing (Hough et al., 1982; Briggs et al., 2004; Edney et al., 2007). Three major brewing properties of wort compared were: the reducing sugars, FAN and fermentability (Table 3).

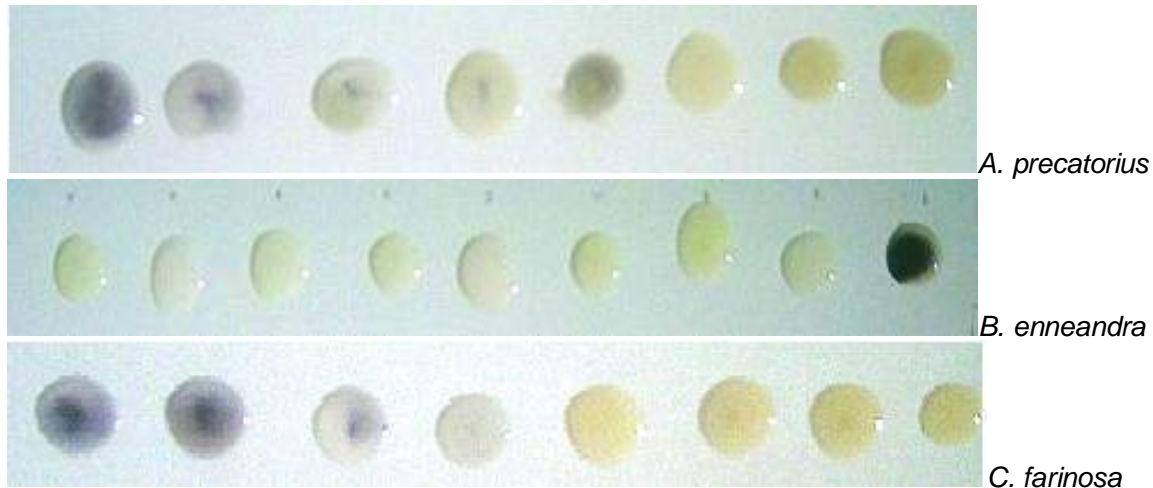


Figure 1. Iodine test during mashing of unmalted *Safrari* sorghum using crude extracts of *A. precatorius*, *B. enneandra* and *C. farinosa*.

Reducing sugars are about the same level after mashing unmalted *Safrari* with *A. precatorius* and *B. enneandra*, but slightly lower with *C. farinosa*. The level was two times on the average as low after mashing *Safrari* malt, as compared to those obtained for mashing unmalted *Safrari* using *A. precatorius* and *B. enneandra* extracts, but about three times as high after mashing barley malt. FAN was highest after mashing unmalted *Safrari* with *B. enneandra*, followed by *C. farinosa* and then *A. precatorius* extracts and this is in the ratio of 100:78:24%. The free amino nitrogen content of malted barley wort was about 120% that of malted *Safrari* worts. The fermentability levels of *Safrari* and barley worts were similar but were much higher than those of the extracts and adjuncts. Worts obtained after mashing unmalted *Safrari* using the crude enzyme extracts of *A. precatorius*, *B. enneandra* and *C. farinosa* showed fermentabilities fluctuating between 37 and 46%, with *C. farinosa* giving the lowest results and *B. enneandra* the highest. The fermentation performance of the yeast strain used in this study was however lower than expected (Briggs et al., 2004). The results obtained after carrying out the iodine test during mashing are shown in Figure 1.

Conclusion

The crude extracts of *A. precatorius*, *B. enneandra*, and *C. farinosa* used in northern Cameroon to make sweet starch gruel for local consumption do have amylase and proteolytic activities. These extracts can be used for mashing and brewing of sorghum and other cereals. Further fractionation and purification of the crude extracts in order to concentrate their various enzyme components, would be of great use not only to the people of this part of Cameroon but also for industrial purposes.

REFERENCES

- Agu RC, Palmer GH (1998). Effect of mashing with commercial enzymes on the properties of sorghum worts. *World J. Microbiol. Biotechnol.* 14:43-48.
- Analytica-EBC (2006). European Brewery Convention. Fachverlag Hans Carl: Nürnberg, Germany.
- Arri BK (1990). Problems associated with the use of sorghum for lager beer production. *Industrial utilization of sorghum: Symposium on the Current Status and Potential of Industrial Uses of Sorghum*. ICRISAT: Kano, Nigeria.
- Bajomo MF, Young TW (1992). Development of profile for the use of microbial enzymes in brewing with raw sorghum (80%) and malted barley of sorghum malt (20%). *J. Inst. Brew.* 98:515-523.
- Bernfeld P (1986). Amylases, alpha and beta. This week's citation classic n° 40. Tufts University School of Medicine. Boston, MA. *Meth. In Enzymol.* 1:149-158.
- Briggs DE, Boulton CA, Brookes PA, Stevens R (2004). *Brewing science and practice*. Cambridge: Woodhead Publishing Limited and CRC Press LLC.
- Dale CJ, Young TW, Omole T (1990). Small scale mashing experiments with grist containing high proportions of raw sorghum. *J. Inst. Brew.* 96:403-409.
- Delrue RM (1987). A review of glucose and fructose syrups. In: A. Turner, Ed., *Food Technology, International Europe*. Sterling Publications International. London: 171-174.
- Desobgo ZSC, Nso EJ, Tenin D, Kayem GJ (2010). Modelling and optimizing of mashing enzymes - effect on yield of filtrate of unmalted sorghum by use of response surface methodology. *J. Inst. Brew.* 116 (1):62-69.
- Dufour JP, Melotte L (1992). Sorghum malts for the production of a lager beer. *J. Am. Soc. Brew. Chem.* 50 (3):110-119.
- Edney MJ, Eglinton JK, Collins HM, Barr AR, Legge WG, Rosnagel BG (2007). Importance of Endosperm Modification for Malt Wort Fermentability. *J. Inst. Brew.* 113(2):228-238.
- EtokApan OU, Palmer GH (1990). Comparative studies of the development of endosperm-degrading enzymes in malting sorghum and barley. *World J. Microbiol. Biotechnol.* 6:408-417.
- Fisher EH, Stein EA (1969). *Enzymes 2nd Ed. Vol 4*, Academic Press, New York: 343.
- Glew RH, Kramer JKG, Hernandez M, Pastuszyn A, Ernst J, Ngouya DN, VanderJagt DJ (2010). The amino acid, mineral and fatty acid content of three species of human plant foods in Cameroon. *J. Phytochem.* 62(4):21-30.
- Goode LD, Arendt EK (2003). Pilot scale production of lager beer from

- grist containing 50% unmalted sorghum. *J. Inst. Brew.* 109(3):208-217.
- Goode LD, Halbert C, Arendt EK (2002). Mashing studies with unmalted sorghum and malted barley. *J. Inst. Brew.* 108(4):465-473.
- Goode LD, Halbert C, Arendt EK (2003). Optimisation of mashing conditions when mashing with unmalted sorghum and commercial enzymes. *J. Am. Soc. Brew. Chem.* 61(2):69-78.
- Hough JS, Briggs DE, Stevens R, Young TW (1982). *Malting and brewing science*. University Press ed. London: Chapman and Hall.
- Hudson J (1986). What's new in brewing technology? *Food Manuf.* 60:64-65.
- Law BA (1990). Tailoring enzymes for food users. In: A. Turner, Ed., *Food Technology International Europe*. Sterling Publications International, London. 161-165.
- MacFadden DP, Clayton M (1989). Brewing with sorghum-use of exogenous enzymes. *Brewing and Beverage Industry International.* 1:71-81.
- Nso EJ, Ajebesone PE, Mbofung CM, Palmer GH (2003). Properties of three sorghum cultivars used in the production of Bili-Bili beverage in Northern Cameroon. *J. Inst. Brew.* 109(3):245-450.
- Nso EJ, Nanadum M, Palmer GH (2006). The effect of formaldehyde on enzyme development in sorghum malts. *Tech. Q. Master Brew. Assoc. Am.* 43(3):177-182.
- Okolo BN, Ezeogu LI (1995). Effects of air rest period on the mobilization of sorghum reserve proteins. *J. Inst. Brew.* 101:463-468.