Microwave assisted extraction of bound phenolicacids from sorghum and maize bran

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ABSTRACT

Sorghum and maize are increasingly being recognised as sources of phenolic acids with health-benefiting antioxidant activity. These phenolic compounds are concentrated in the bran mainly in bound form. To release the intact etherified bonds, alkaline refluxing is employed for lengthy periods. Microwave assisted extraction (MAE) offers an alternative due to its rapidity, and a combination of high temperature and pressure. Microwave assisted extraction procedure was applied to sorghum and maize bran to release bound phenolic acids and sample solubilisation was achieved after 45s in 2 M NaOH. Phenolic acids were quantified and confirmed by HPLC-MS/MS. Ferulic acid (FA) and *p*-coumaric acid (PCA) were the only monomeric phenolics found in both cereal brans. The diferulic acids (DFA) 8-*O*-4' and 8-5' benzofuran form were found in sorghum bran and only the former in maize bran. The contents of ferulic acid and diferulic acids in sorghum bran were at least 6 and 5 times less than in maize, respectively. Several phenolic acids have been reported in sorghum and maize although our findings showed few of these suggesting that MAE could have resulted in the thermal degradation of most phenolic acids. The large differences in phenolic acid contents despite similarities in structure and chemical composition of these cereals suggest variations in their biosynthesis and cross-linking, hence affecting the degree of alkaline hydrolysis and breakage of ether bonds

Keywords: microwave assisted extraction, diferulic acids, sorghum, maize

INTRODUCTION

- Phenolic compounds occur mostly in bound form, etherified to cell walls
- Alkaline hydrolysis used to release ether bonds but procedure is long and unsuitable for routine batch analyses.
- MAE alternative procedure because of its rapidity, reduced solvent consumption and high phenolic yield (Beejmohun et al., 2007).
- Furthermore, the technique combines high temperature and high pressure for optimal release of phenolic acids by cell wall breakdown.

OBJECTIVES

- To use MAE as a rapid method for the release of phenolic acids from sorghum and maize brans

MATERIALS AND METHODS

8 sorghum and 8 maize cultivars
TADD decortication, 80% extraction rate

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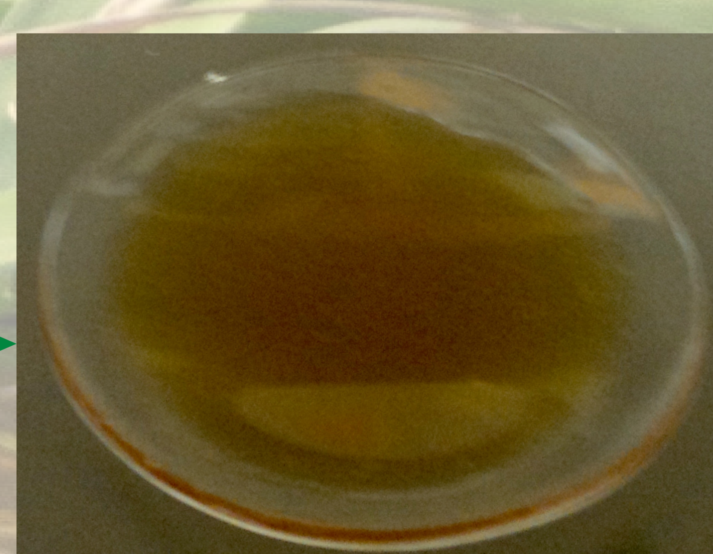
Bran fractions
Microwave assisted extraction: 2 M NaOH, 45 s, 100% power

↓ Extract →



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Analyses
70 min linear gradient, 0.7 mL/min, 0.1% acetic acid in water & 0.1% acetic acid in methanol. Mass spectra negative mode, *m/z* 100-1500

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Phenolic acid identification and quantitation

- Fluorescence microscopy of untreated sorghum and maize bran and their MAE extracts (Carl Zeiss, Oberkochem, Germany).

RESULTS AND DISCUSSIONS

- Ferulic acid (t_R 42.3), PCA (t_R 38.9), 8-5' benzofuran form (44.5) and 8-*O*-4'diferulic acid (67.8) identified in sorghum and only the latter DFA in maize bran (Fig 1i-ii).
- Identity of peaks confirmed with MS/MS data (Tables 1), (Bily et al., 2004; Qiu et al., 2010).
- Peaks at t_R 21.4, 26.9 and 29.8 min in sorghum bran and at (t_R 33.7) in maize bran were unidentified, could be due to isomerisation.
- Fewer phenolics than found in sorghum and maize grains (Bily et al., 2004), PCA and FA stable at high temperature due to their chemical structures.

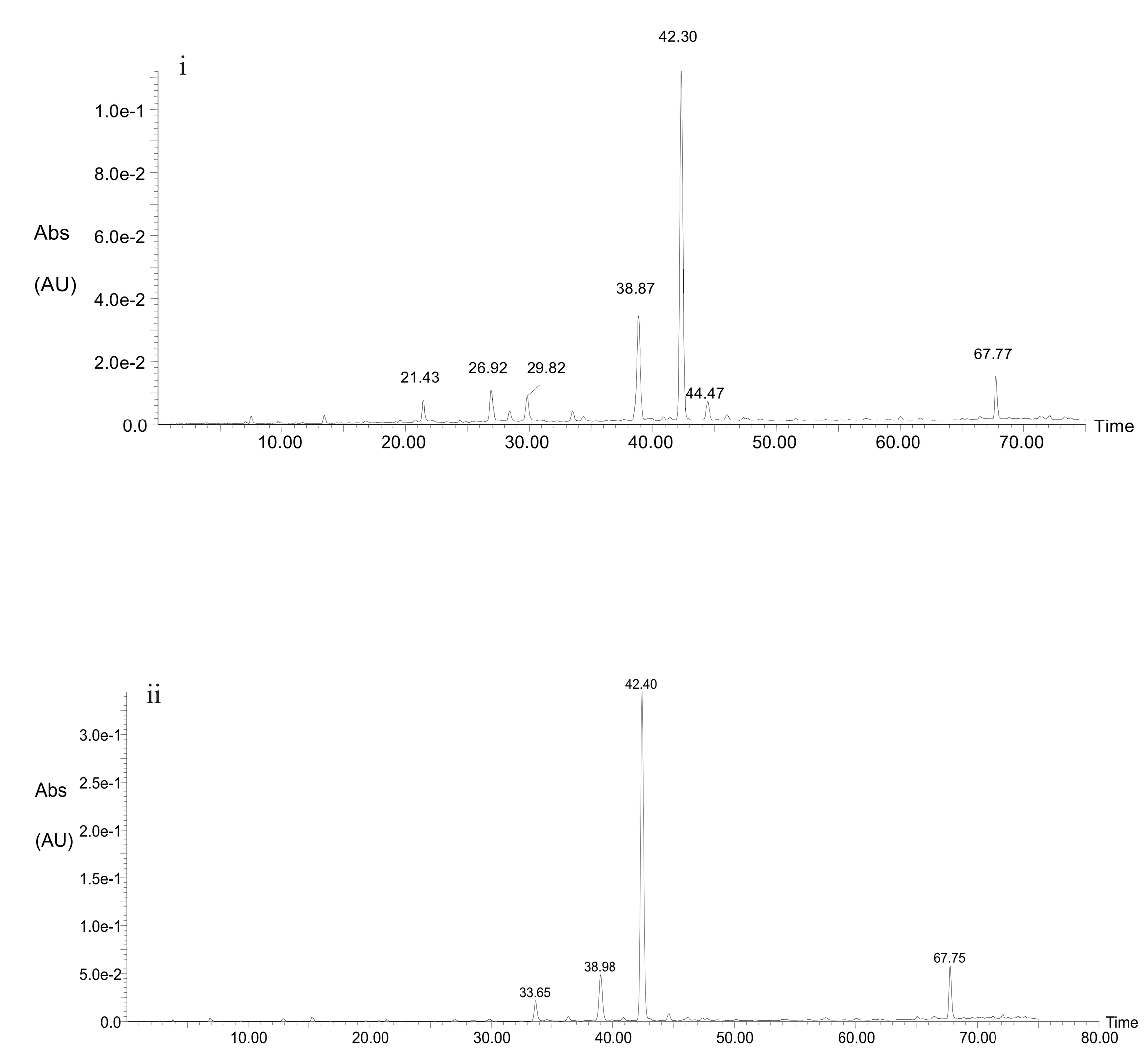

Fig 1. LC chromatogram of phenolic acids in bran of (i) sorghum cultivar PAN 8564 and (ii) maize cultivar AFG 4555 at 320 nm

Table 1. Summary of major peaks identified in sorghum and maize bran by MS/MS

Peak	Retention time (min)	Parention (m/z)	Fragments (*m/z*)	Identification
i	38.87	163	119	*p*-coumaric acid
ii	42.30	193	178, 134	ferulic acid
ii	44.47	385	341,326, 282, 267	8-5' benzofuran form DFA
iv	67.77	385	341, 298, 193, 178, 134	8-*O*-4' DFA

- Maize monomeric phenolic acids and DFA were 3 and 5 times higher than sorghum, respectively
- Differences in DFA content and composition of sorghum and maize could be due to variations in their biosynthesis and cross-linking, affecting the degree of alkaline hydrolysis and breakage of ether bonds (Parker et al., 2005).

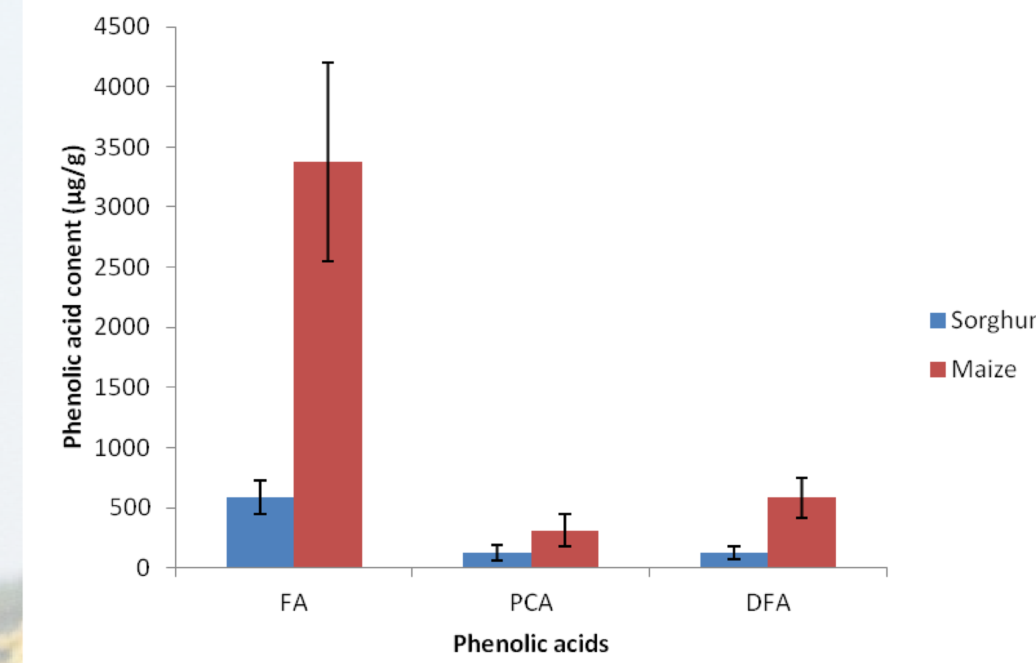

Fig 2. Bound phenolic acids of sorghum and maize bran released by MAE.

- Low FA content confirmed with fluorescence, sorghum MAE extract showed very faint regions of fluorescence than its untreated bran and that of maize bran and extract (Fig 3).

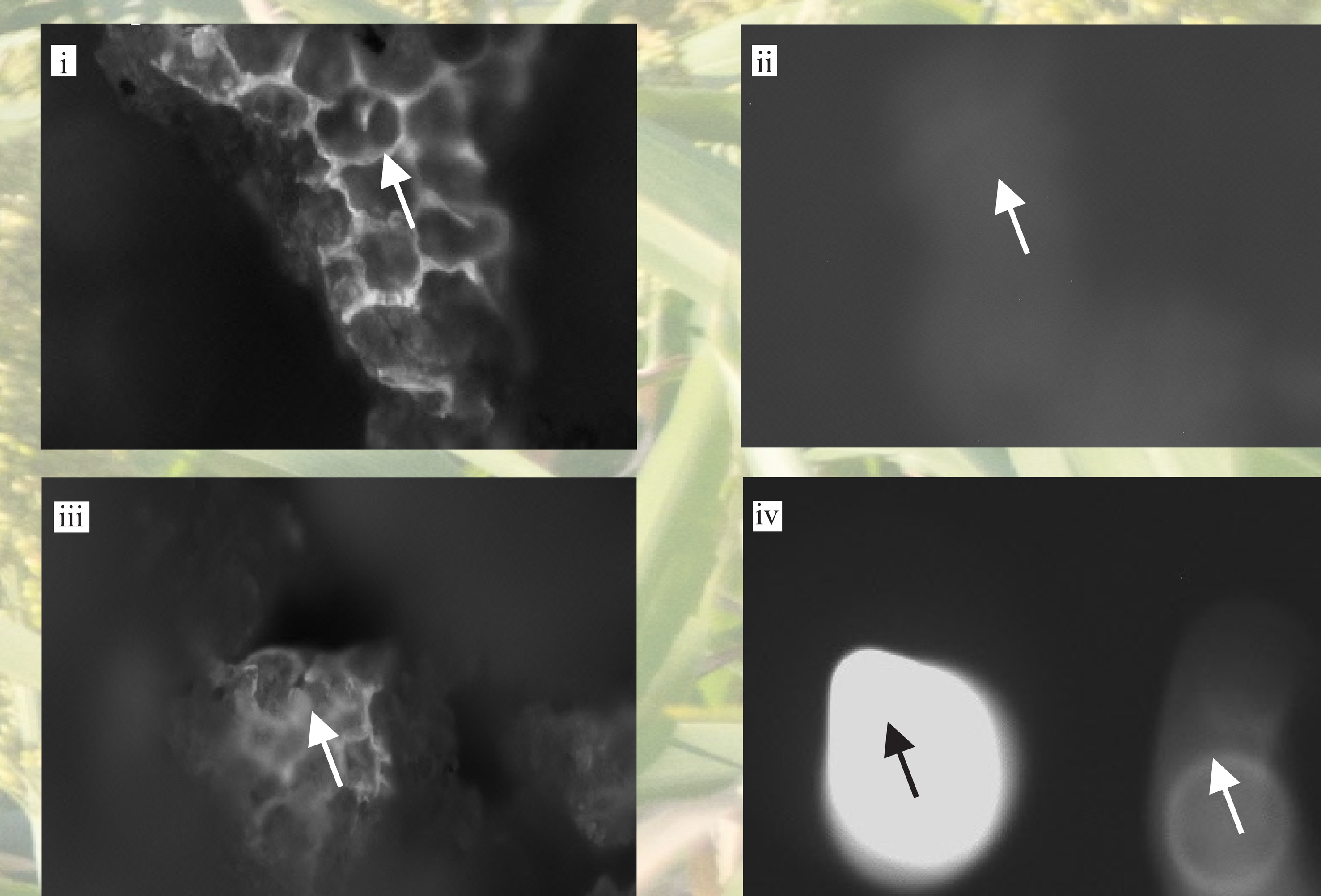

Fig 3: FA fluorescence of (i) Untreated sorghum bran, (ii) sorghum bran MAE extract, (iii) untreated maize bran and (iv) maize bran MAE extract. Arrows point to FA fluorescing regions.

CONCLUSIONS

- MAE has potential for rapid release of higher quantities of FA and DFA from maize bran, under the conditions used.
- More work is required to optimise microwave conditions for sorghum extraction

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