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Review

Sorghum phytochemicals and their potential impact on human health

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Abstract

Sorghum is a rich source of various phytochemicals including tannins, phenolic acids, anthocyanins, phytosterols and policosanols. These phytochemicals have potential to significantly impact human health. Sorghum fractions possess high antioxidant activity in vitro relative to other cereals or fruits. These fractions may offer similar health benefits commonly associated with fruits. Available epidemiological evidence suggests that sorghum consumption reduces the risk of certain types of cancer in humans compared to other cereals. The high concentration of phytochemicals in sorghum may be partly responsible. Sorghums containing tannins are widely reported to reduce caloric availability and hence weight gain in animals. This property is potentially useful in helping reduce obesity in humans. Sorghum phytochemicals also promote cardiovascular health in animals. Such properties have not been reported in humans and require investigation, since cardiovascular disease is currently the leading killer in the developed world. This paper reviews available information on sorghum phytochemicals, how the information relates to current phytonutrient research and how it has potential to combat common nutrition-related diseases including cancer, cardiovascular disease and obesity.

Keywords: *Sorghum bicolor*; Gramineae; Phytochemicals; Tannins; Anthocyanins; Phenolic acids; Phytosterols; Policosanols; Human health; Cancer; Cardiovascular disease; Obesity

Contents

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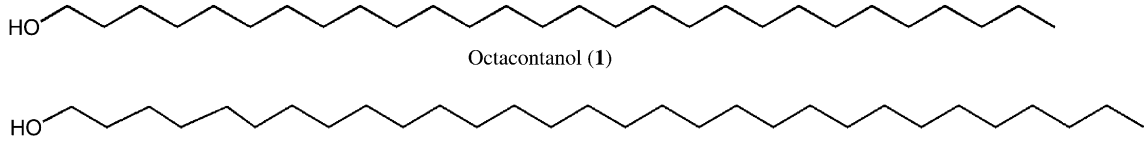
1. Introduction

Sorghum is the fifth most important cereal crop in the world after wheat, rice, corn and barley. Sorghum outperforms other cereals under various environmental stresses and is thus generally more economical to produce. More than 35% of sorghum is grown directly for human consumption. The rest is used primarily for animal feed and alcohol and industrial products. The United States is the largest producer and exporter of sorghum, accounting for 20% of world production and almost 80% of world sorghum exports in 2001–2002 (USDA-FAS, 2003). World sorghum production was 57 million metric tons during this period.

Sorghum contains various phytochemicals (including phenolic compounds, plant sterols and policosanols) that are secondary plant metabolites or integral cellular components. Phenols help in the natural defense of plants against pests and diseases, while the plant sterols and policosanols are mostly components of wax and plant oils. The phytochemicals have gained increased interest due to their antioxidant activity, cholesterol-lowering properties and other potential health benefits. The phenols in sorghums fall under two major categories; phenolic acids and flavonoids. The phenolic acids are benzoic or cinnamic acid derivatives (Hahn et al., 1983; Waniska et al., 1989), whereas the flavonoids include tannins and anthocyanins as the most important constituents isolated from sorghum to date (Gupta and Haslam, 1978; Gujer et al., 1986; Gous, 1989; Gu et al., 2002; Krueger et al., 2003). Sorghum phytosterols are similar in composition to those from corn and contain mostly free sterols or stanols and their fatty acid/ferulate esters (Avato et al., 1990; Singh et al., 2003). The sterols and stanols are structurally similar, except for the presence of a double bond at position 5 in sterols, which is lacking in stanols. The policosanols (fatty alcohols) exist mostly as free or esterified forms with C24–C34 atoms, and the general formula CH_3–$(CH_2)_n$–CH_2OH (Fig. 1). In sorghum-free forms of the C28 (octacosanol) (**1**) and C30 (triacontanol) (**2**) are the most abundant (Avato et al., 1990; Hwang et al., 2002).

Sorghums vary widely in their phenolic composition and content, with both genetics and environment affecting the kind and level of phenolic compounds. Based on extractable tannin content, sorghums have been classified as type I (no significant levels of tannins extracted by 1% acidified methanol), e.g., TX2911 (red perocarp), type II (tannins extractable in 1% acidified methanol and not methanol alone), e.g., Early Hegari and type III (tannins extractable in both acidified methanol and methanol alone), e.g., Early Sumac variety (Cummings and Axtel, 1973; Price et al., 1978). However, this classification does not account for the varying levels of other major phenolic constituents, especially anthocyanins. Another broad way to classify sorghum is based on both appearance and total extractable phenols; thus, we have white sorghums (also



Octacontanol (**1**)

Triacontanol (**2**)

Fig. 1. Common policosanols found in sorghum.

called food-type) with no detectable tannins or anthocyanins and very low total extractable phenol levels; red sorghums which have no tannins but have a red pericarp with significant levels of extractable phenols; black sorghums with a black pericarp and very high levels of anthocyanins and the brown sorghums which have a pigmented testa and contain significant levels of tannins, with varying degrees of pericarp pigmentation.

Despite the high levels and diversity of phytochemicals in sorghum, research on this crop as a source of valuable health promoting compounds lags behind other commodities (e.g., fruits and vegetables). As a result, utilization of sorghum fractions in foods to improve nutrition is very limited. Sorghum has a big potential, given its agronomic properties, as well as the emerging evidence on the biological effects of the phytochemicals present in the grain. The purpose of this paper is to demonstrate that sorghums with special characteristics exist that have potential as significant sources of condensed tannins, anthocyanins and other phytochemicals with properties that complement the phytochemicals present in fruits and vegetables.

2. Traditional sorghum use for food

Sorghum is used in a variety of foods. The white food sorghums are processed into flour and other products, including expanded snacks, cookies and ethnic foods, and are gaining popularity in areas like Japan (United States Grains Council, 2001; Rooney, 2001). In the US, the white sorghum products are used to a small extent to substitute for wheat in products for people allergic to wheat gluten (Fenster, 2003).

Other varieties of sorghum are also used for food in various parts of the world, including parts of Africa, Central and South America, China, and India. In Eastern and southern Africa, for example, traditional sorghum varieties of moderate tannin content are widely grown and used for staple food and alcoholic beverages. Their agronomic advantages outweigh any negatives such as reduced nutrient availability or astringency. In Southern Africa, small-scale farmers intercrop tannin and tannin-free sorghums in areas prone to high bird predation in order to reduce grain losses in the field. When harvested the grain is mixed and used in porridge making. In some African cultures, the tannin sorghums are actually preferred because porridge from the tannin sorghums 'remains in the stomach longer' and the farmer feels full for most of the day doing field work. Other pigmented sorghums are also preferred in some African cultures because of the characteristic color they produce in certain foods, and also due to traditional belief that they promote the health of unborn babies and are therapeutic against diseases of the digestive system. In the western world, however, pigmented sorghum use for food is almost non-existent. Innovative ways of incorporating these sorghums into the mainstream diet are necessary to realize the benefits they may offer.

3. Tannins in sorghum

Tannins are the most uniquely important phytochemical components of sorghum since they possess properties that produce obvious and significant effects in animals, and have also been associated with various positive and negative impacts on human health. These aspects and their relevance are discussed in later sections of the review.

3.1. Production and genetics of tannin sorghums

Even though tannins are commonly associated with sorghums, more than 99% of sorghum currently produced in the US is tannin-free. Decades of breeding efforts to eliminate tannins from sorghum were motivated mostly by the reduced feed value of the tannin sorghums. Tannins bind to and reduce digestibility of various food/feed nutrients, thus negatively affecting productivity of livestock. Current non-tannin sorghums grown for livestock feed in the US have virtually the same energy profile as corn. The limited quantities of tannin sorghums grown in the US are mostly identity preserved seed stock lines. However, in many other parts of the world where pests and diseases are common, tannin sorghums are still grown in significant quantities since they are more tolerant of such conditions than the non-tannin varieties (Tipton et al., 1970; Ellis, 1972; Rooney and Sullins, 1977; Hahn et al., 1983; Waniska et al., 1989).

Tannins are present in sorghums with a pigmented testa (classified as type II and III sorghums) (Fig. 2). These sorghums have dominant B_1_B_2_ genes. The B_1 and B_2 genes control the presence or absence of the pigmented testa layer (Hahn and Rooney, 1986). Both genes must be dominant for a pigmented testa to develop. When the S gene (spreader gene) is dominant concurrently with the dominant B_1 and B_2 genes, pericarp color becomes phenotypically brown (Earp et al., 1983). The sorghums with the dominant S_ gene generally contain tannins that are more easily extractable than the ones with the recessive gene (Hahn and Rooney, 1986). Such sorghums (with dominant S_ gene) also produce greater anti-nutritional effects in animals (Cousins et al., 1981). Since the pericarp color and secondary plant color of sorghum is genetically controlled, it is possible to develop different combinations of pericarp and plant color with and without the pigmented testa and spreader genes, which opens the possibility of significantly different levels and combinations of phenolic compounds.

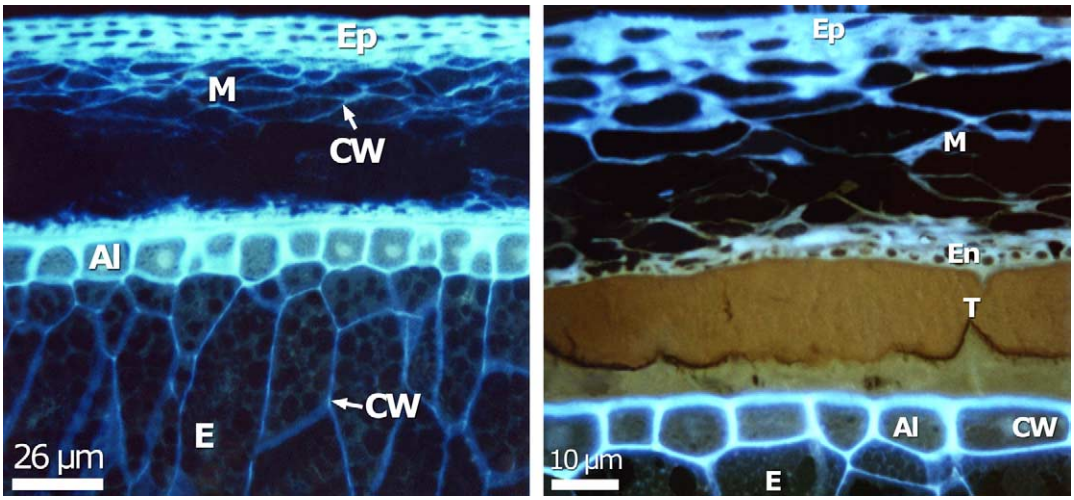


Fig. 2. Fluorescence photomicrograph of sorghum bran cross-section, showing structural differences between a non-tannin sorghum without a testa (left) and a tannin sorghum with a pigmented testa (right). Al, aleurone layer; CW, cell wall; E, endosperm; En, endocarp; Ep, epicarp; M, mesocarp; T, pigmented testa (courtesy of Cheryl Earp, RiceTech, Alvin, TX).

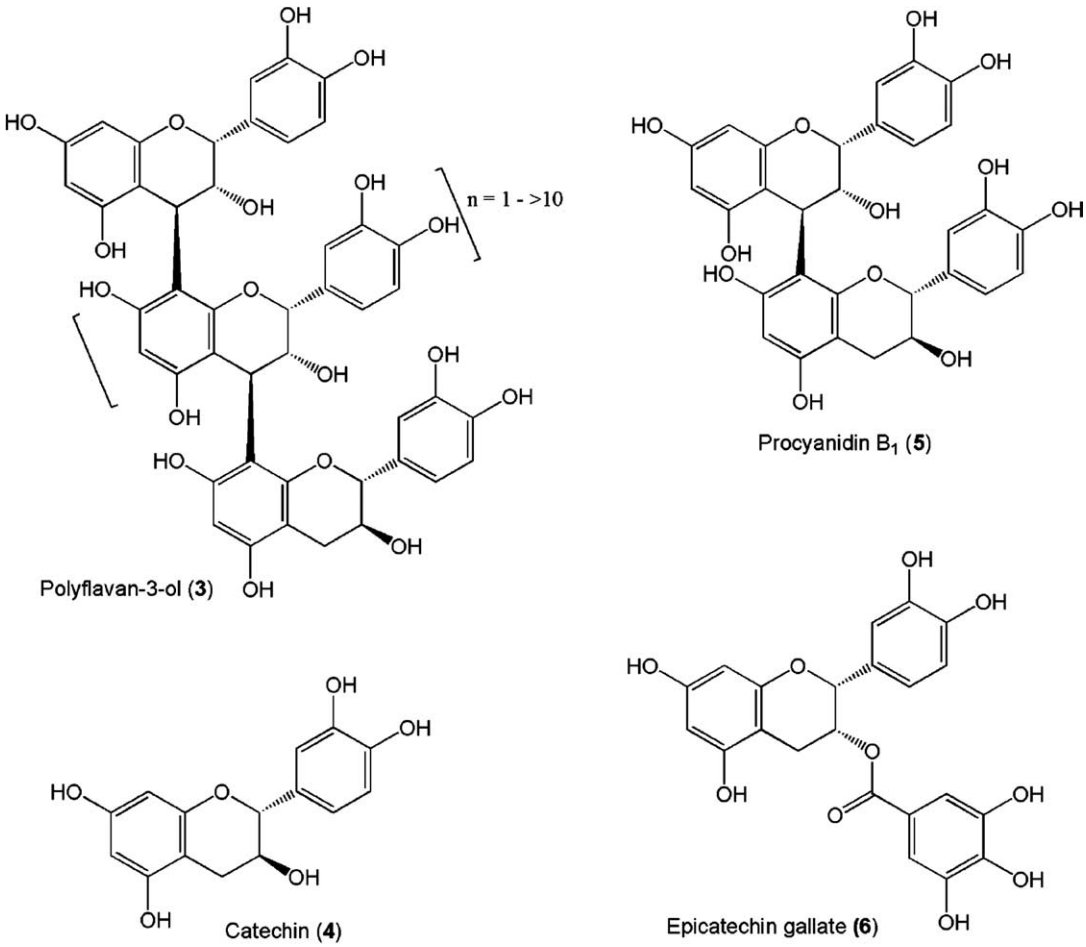


Fig. 3. The proanthocyanidins most commonly reported in sorghum: **3** and **4** (Gupta and Haslam, 1978; Gu et al., 2002); **5** (Gupta and Haslam, 1978); **6** (Awika, 2003).

3.2. Chemical composition and structure of tannins from sorghum

Tannins of sorghum are almost exclusively of the "condensed" type. They are mainly polymerized products of flavan-3-ols and/or flavan-3,4-diols (Fig. 3). Glycosylated and non-glycosylated polymers of flavan-4-ols with various substitution patterns have also been reported in sorghum (Fig. 4) (Gujer et al., 1986; Krueger et al., 2003). No tannic acid or hydrolysable tannins have been detected in sorghum. Gupta and Haslam (1978) identified proanthocyanidins with (−)-epicatechin chain extension units and (+)-catechin as chain termination units in sorghum (**3**). These results were largely confirmed by Gu et al. (2002) who identified (+)-catechin as the main chain-terminating unit (88%), with (−)-epicatechin accounting for the rest of the terminal units of sorghum procyanidins. Catechin (**4**) is the most commonly reported monomer, while procyanidin B1 (**5**) is the most common dimer present in sorghum (Gupta and Haslam, 1978; Awika, 2003). Chromatographic analysis also suggested presence of epicatechin gallate (**6**) as a significant component of the lower MW proanthocyanidins of tannin sorghums (Awika, 2003). However, a greater diversity of sorghum polymeric proanthocyanidins has been reported (Brandon et al., 1982; Gujer et al., 1986; Krueger et al., 2003). Brandon et al. (1982) reported the presence of prodelphinidins in sorghum. Gujer et al., 1986) also found dimers and trimers of hetero-polyflavans with glycosylated luteolinidin as a chain extender and eriodictoyl or its glucoside as chain terminators (**7** and **8**). Similar results were reported by Krueger et al. (2003) who observed the presence of DP2–DP7 mixtures of proluteolinidin (**7**) and proapigeninidin (**8**) polyflavans, with eriodictoyl or eriodictoyl-*O*-β-glucoside as the terminal units. Krueger et al. (2003) also observed great heterogeneity in the polyflavan-3-ol (procyanidin) polymers in terms of interflavan linkages (A or B type) and also the presence of gallocatechin/epigallocatechin hydroxylation patterns in sorghum tannin polymers (**9** and **10**). Some of the tannins found in sorghum have also been identified in other plant foods, e.g., cocoa, blueberries and cranberries (Hammerstone et al., 1999; Kennedy et al., 2001; Yang and Chien, 2000; Gu et al., 2002).

3.3. Levels of tannins in sorghum

The structural complexity of the tannins often creates a difficulty in isolating and characterizing them effectively. However, with the growing interest in natural antioxidants, an accurate characterization of the sorghum polyflavans (tannins) is essential to effectively predict their potential effects on health and nutrition. Knowledge of the polyflavan structures and chemical properties is also necessary for their accurate quantification.

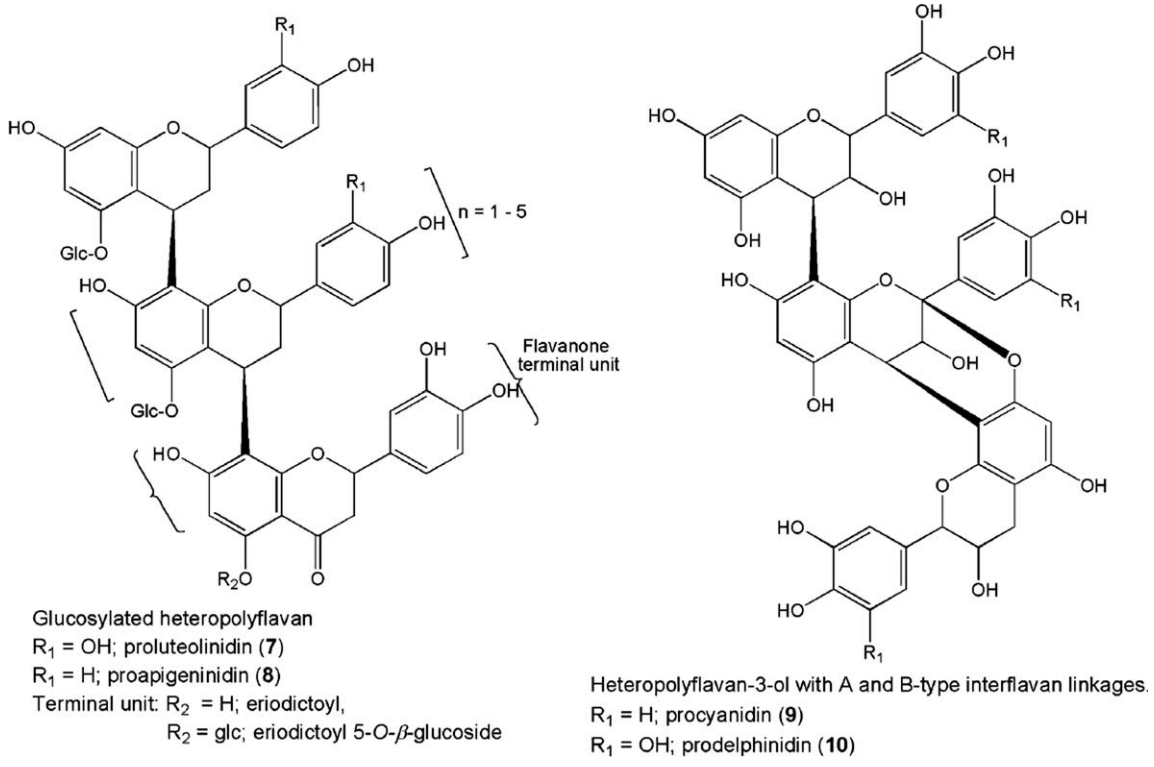


Fig. 4. Heterogenous polyflavans found in sorghum: **7** and **8** (Brandon et al., 1982; Krueger et al., 2003); **9** and **10** (Gujer et al., 1986; Krueger et al., 2003).

Quantification of tannins from any source is further complicated by lack of appropriate standards. Additionally, the choice of organic solvent and extraction procedure significantly affect results. Deshpande and Cheryan (1986) and Santos-Buelga and Scalbert (2000) provide good summaries of the several categories of methods used to quantify tannins and some specific considerations. The methods broadly fall under colorimetric (most common) and chromatography-based assays (more recent). Techniques based on ability of tannins to precipitate proteins have also been used (Hagerman and Butler, 1978, 1980; Makkar, 1989). The colorimetric and protein-based assays generally suffer from lack of specificity.

Among the colorimetric assays, the vanillin–HCl method originally reported by Burns (1971) and later modified (Maxson and Rooney, 1972; Price et al., 1978) is widely used for sorghums (Hahn and Rooney, 1986; Agullo and Rodriguez, 1995; Steadman et al., 2001). The 4-dimethylaminocinnamaldehyde (DMACA) method is also commonly reported (Treutter, 1989; Santos-Buelga and Scalbert, 2000). The DMACA and corrected vanillin–HCl methods are generally more appropriate for tannin estimation than the redox-based colorimetric assays like the Folin–Denis or Prussian blue methods which are less specific. With the vanillin–HCl method tannin levels of up to 68 mg/g have been reported for sorghum grains (Table 1). Additionally, factors like material particle size, type of solvent, extraction time and standards used, among others significantly influence the measured tannin content (Deshpande and Cheryan, 1986). For example Price et al. (1978) reported that extraction for 2–24 h with 1% HCl in methanol resulted in a 40–70% reduction in assayable tannins compared to a 20-min extraction time. We made similar observations in our laboratory. This phenomenon may be due to reaction of the tannins with other components in the extract to products that do not react positively in the vanillin–HCl test. Hence, it is always essential to specify extraction conditions, and caution is necessary when interpreting results. In general, better methods that are more stable to limited variations in sample composition, preparation and extraction procedures are necessary. Progresses in HPLC-based assays in the recent past are promising.

Table 1
Tannin contents[a] of sorghum grain and selected cereals and other food commodities

Commodity	mg/g (dry wt)	Reference
Tannin sorghum	10.0–68.0	b–e
Tannin-free sorghum	0.5–3.8	e
Finger millet	3.6–13.1	f
Buckwheat groats	1.7	g
Pinto beans	1.5–3.3	h
Faba beans	nd–0.7	i
Lentils	3.2–10.4	j
Cowpea	1.8–2.9	h, k

(b) Jambubathan and Mertz (1973); (c) Hahn and Rooney (1986); (d) Agullo and Rodriguez (1995); (e) Awika (2000); (f) Deshpande and Cheryan (1986); (g) Steadman et al. (2001); (h) Deshpande et al. (1985); (i) Helsper et al. (1993); (j) Vidal-Valverde et al. (1994); (k) Ummadi et al. (1995).

[a] Expressed as catechin equivalents, measured by the vanillin–HCl method. The vanillin–HCl method normally gives background noise (false positive), and in our experience values of up to 4.0 mg/g were obtained for sorghum samples that genetically have no tannins. Such samples do not show any peaks when analyzed by the more precise HPLC methods. Hence, the vanillin–HCl method should not be used for reporting presence or absence of tannins in a sample per se.

Quantitative assays of tannins based on HPLC were hampered in the past by difficulties in resolving the high molecular weight polyflavans, which are the most abundant forms of the tannins in sorghum (Hagerman and Butler, 1980; Rigaud et al., 1993; Hammerstone et al., 1999). Reversed phase HPLC techniques have been useful only for resolving the low MW procyanidins (up to trimers) (Lea et al., 1979; Putman and Butler, 1989). Rigaud et al. (1993) developed a normal phase HPLC method coupled with UV detection that separated procyanidins up to tetramers from grape seed and up to pentamers from cacao beans. Oligomers and polymers beyond pentamers tended to fuse into broad unresolved humps, making the method unsuitable for quantification. Improvements of the Rigaud et al. (1993) method that coupled the normal phase HPLC with fluorescence detection was used to successfully separate and quantify procyanidin oligomers up to decamers from cocoa beans and other food materials (Adamson et al., 1999; Hammerstone et al., 1999; Hammerstone et al., 2000). However, the method was not able to resolve polymers with DP >10, and hence underestimated total procyanidin content. Gu et al. (2002) later improved the method to effectively separate proanthocyanidin monomers to decamers, with the polymers beyond decamers resolving as a distinct quantifiable peak. With the improved method, these authors were able to quantify proanthocyanidin content of sorghum, cocoa, and some fruits. We recently successfully used the improved method of Gu et al. (2002) to characterize sorghum procyanthocyanidin polymers subjected to various food processing conditions (Awika et al., 2003a).

The advantage of the HPLC assay is that it allows for estimation of relative contributions of the various molecular weights to the tannin content of a sample (Table 2, Fig. 5). This is important since polymer chain length affects organoleptic, antioxidant and other properties of the tannins (Rigaud et al., 1993; Tebib et al., 1997; Lotito et al., 2000). Hence, information on relative proportions of the different proanthocyanidin oligomers and polymers may help predict their overall effectiveness as functional components of diet. For example, we recently demonstrated that extrusion of tannin sorghum (into direct expanded snacks) causes an overall reduc-

Table 2
Proanthocyanidin content[a] of brown sorghums compared to those of freeze-dried cocoa and blueberry[b]

DP[c]	CSC3*R28 grain	Sumac grain	Sumac bran	Cocoa[d]	Blueberry[d]
1	0.01 ± 0.00	0.18 ± 0.01	0.33 ± 0.07	14.24 ± 0.38	0.18 ± 0.01
2	0.09 ± 0.01	0.40 ± 0.01	1.33 ± 0.26	8.57 ± 0.51	0.46 ± 0.02
3	0.12 ± 0.01	0.51 ± 0.01	1.61 ± 0.33	8.10 ± 0.49	0.38 ± 0.02
4	0.21 ± 0.03	0.69 ± 0.01	2.32 ± 0.46	8.89 ± 0.54	0.50 ± 0.01
5	0.26 ± 0.04	0.74 ± 0.01	2.51 ± 0.51	8.86 ± 0.52	0.47 ± 0.01
6	0.49 ± 0.07	1.10 ± 0.02	3.61 ± 0.71	9.99 ± 0.61	0.69 ± 0.05
7	0.38 ± 0.06	0.79 ± 0.01	2.56 ± 0.50	6.38 ± 0.38	0.48 ± 0.02
8	0.38 ± 0.06	0.74 ± 0.01	2.29 ± 0.43	5.97 ± 0.31	0.61 ± 0.03
9	0.63 ± 0.10	1.17 ± 0.02	3.48 ± 0.62	7.36 ± 0.58	0.93 ± 0.02
10	0.31 ± 0.04	0.55 ± 0.01	1.52 ± 0.26	3.22 ± 0.23	nd[e]
P[f]	17.67 ± 3.92	15.09 ± 0.34	36.87 ± 6.12	16.17 ± 0.80	15.28 ± 0.51
Total	20.50 ± 4.35	21.97 ± 0.45	58.44 ± 10. 27	97.76 ± 5.32	19.99 ± 0.43
% oligo[g]	14.03	31.31	36.33	83.45	23.51

[a] mg/g, obtained by normal phase HPLC with fluorescence detection.
[b] Adapted from Awika et al. (2003a).
[c] Degree of polymerization.
[d] Gu et al. (2002).
[e] Not detected.
[f] Mixture of polymers with DP >10.
[g] Oligomers (DP <10) as a percent of total.

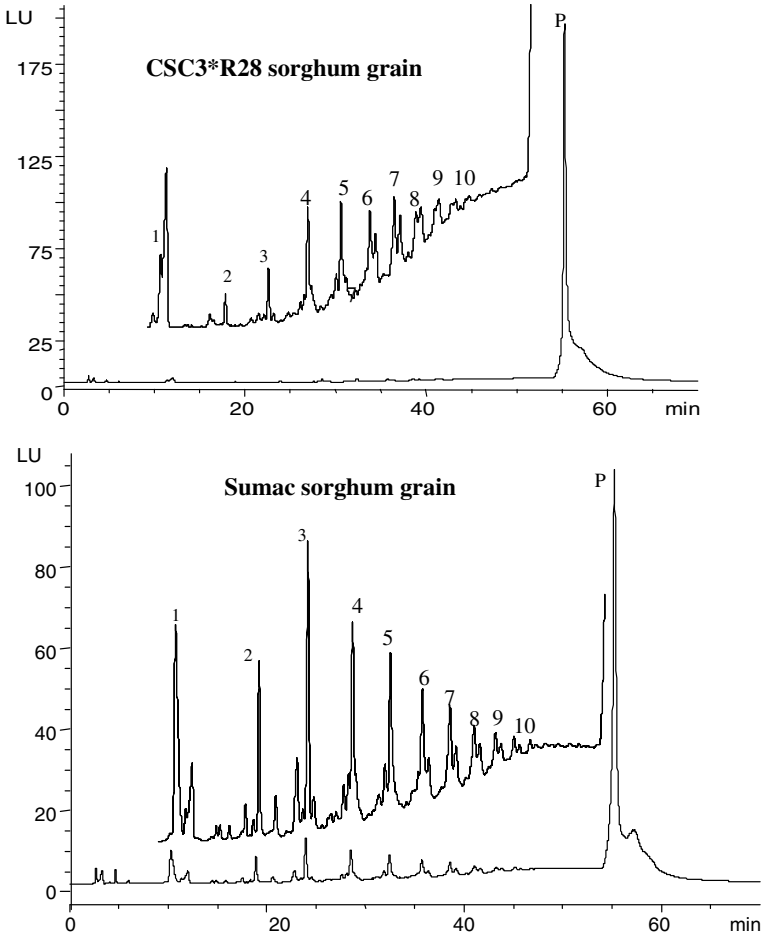
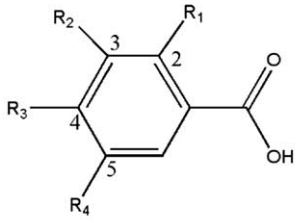


Fig. 5. Proanthocyanidin profiles of two brown sorghum grains, Sumac and CSC3*R28, separated by normal phase HPLC with fluorescence detection. Numbers on peaks denote degree of polymerization. 'P' is mixed polymers (DP >10). Figure insets are vertically magnified oligomer profiles (adapted from Awika et al., 2003a).

tion in extractable tannin content, but significantly improves (2–6-fold) the levels of the lower MW procyanidins (Awika et al., 2003a). The effect of processing on these tannins and their biological implications are currently under investigation using animal models.

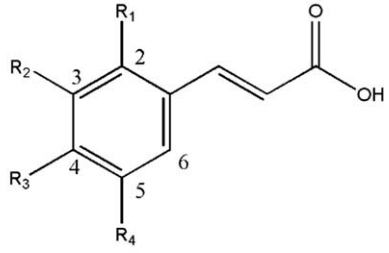
4. Phenolic acids of sorghum

The phenolic acids (PA) of sorghum largely exist as benzoic (**11**–**16**) or cinnamic (**17**–**21**) acid derivatives (Fig. 6). As in other cereals, the sorghum phenolic acids are mostly concentrated in the bran (outer covering of grain). The phenolic acids exist mostly in bound forms (esterified to cell wall polymers), with ferulic acid (**18**) being the most abundant bound PA in sorghum (Hahn et al., 1983) and other cereals (Nordkvist et al., 1984; Adom and Liu, 2002). Several other PA have been identified in sorghum including syringic (**15**), protocatechuic (**16**), caffeic (**17**), *p*-coumaric (**20**), and sinapic (**21**) as the more abundant (Hahn, 1984; Waniska et al., 1989). The PA-like other phenols are thought to help in plant defense against pests and pathogens. The PA show good antioxidant activity in vitro and thus may contribute significantly to the health benefits associated with whole grain consumption.



Benzoic acids (**11-16**)

Gallic acid (**11**): R_1 = H, R_2 = R_3 = R_4 = OH
Gentisic acid (**12**): R_1 = R_4 = OH, R_2 = R_3 = H
Salicylic acid (**13**): R_1 = OH, R_2 = R_3 = R_4 = H
p-hydroxybenzoic acid (**14**): R_1 = R_2 = R_4 = H, R_3 = OH
Syringic (**15**): R_1 = H, R_2 = R_4 = OCH_3, R_3 = OH
Protocatechuic(**16**): R_1 = R_4 = H, R_2 = R_3 = OH



Cinnamic acids (**17-21**)

Caffeic acid (**17**): R_1 = R_4 = H, R_2 = R_3 = OH
Ferulic acid (**18**): R_1 = R_4 = H, R_2 = OCH_3, R_3 = OH
o-coumaric acid (**19**): R_1 = OH, R_2 = R_3 = R_4 = H
p-coumaric acid (**20**): R_1 = R_2 = R_4 = H, R_3 = OH
Sinapic (**21**): R_1 = H, R_2 = R_4 = OCH_3, R_3 = OH

Fig. 6. Some phenolic acid monomers identified in sorghum.

Most quantitative studies specific to phenolic acids are performed by chromatographic analysis. In sorghums, the levels of PA do not correlate with the presence or levels of other phenols (anthocyanins or tannins). However, Waniska et al. (1989) observed increased levels of free PA in certain sorghums with pigmented testa (containing tannins) compared to ones without pigmented testa. In general sorghums have levels of PA comparable to those of other cereals (Table 3). Significant varietal differences are observed in phenolic acid composition and ratios of bound and free forms of these compounds in sorghum. In white sorghum varieties, and most other cereals which normally have very low levels of flavonoids, the bound PA are an important source of

Table 3
Contents of the major phenolic acids in cereals

Phenolic acid	Cereal	μg/g (dry wt)[a]	Reference
Grain			
Ferulic (**18**)	Sorghum	100–500	Hahn and Rooney (1986); Hahn (1984)
	Corn	1740	Adom and Liu (2002)
	Rye	900–1170	Adom and Liu (2002)
	Wheat	640	Adom and Liu (2002)
	Oats	360	Adom and Liu (2002)
	Rice	300	Andreasen et al. (2000)
	Barley	225[b]	Maillard and Berset (1995)
Sinapic (**21**)	Sorghum	50–140	Hahn et al. (1983)
	Rye	70–140	Andreasen et al. (2000)
p-Coumaric (**20**)	Sorghum	70–230	Hahn et al. (1983)
	Rye	40–70	Andreasen et al. (2000)
	Barley	80[b]	Maillard and Berset (1995)
Bran			
Ferulic (**18**)	Sorghum bran	1400–2170	Hahn (1984)
	Wheat bran	5410	Andreasen et al. (2001a)
	Rye bran	2780	Andreasen et al. (2001a)
Sinapic (**21**)	Sorghum bran	100–630	Hahn (1984)
	Wheat bran	75	Andreasen et al. (2001a)
	Rye bran	390	Andreasen et al. (2001a)
p-Coumaric (**20**)	Sorghum bran	0–970	Hahn (1984)
	Wheat bran	170	Andreasen et al. (2001a)
	Rye bran	190	Andreasen et al. (2001a)

[a] Total (free and bound) measured by HPLC.
[b] Bound only.

antioxidant activity. Adom and Liu (2002) found a strong correlation between antioxidant activity and levels of bound ferulic acid in wheat, corn, rice and oats.

5. Sorghum anthocyanins

Anthocyanins have been extensively studied in fruits and vegetables due to their antioxidant properties and potential as natural food colors. However, limited data exist on the types and levels of anthocyanins in cereals, probably because cereals have never been regarded as a commercially significant source. Nip and Burns (1969, 1971) identified apigeninidin (**22**), apigeninidin-5-glucoside (**23**), luteolinidin (**24**) and luteolinidin-5-glucoside (**25**) in red and white sorghum varieties (Fig. 7). Gous (1989) also reported **22** and **24** as the major anthocyanidins from a black sorghum variety. 7-*O*-methylapigeninidin (**26**) (Pale et al., 1997) and fisetinidin (**27**) (Blessin et al., 1963) were also reported in sorghum. Cyanidin (**28**) and pelargonidin (**29**) were also reported in corn (Francis, 1989), and sorghum (Yasumatsu et al., 1965). Cyanidin (**28**) and peonidin (**30**) glycosides were measured in wheat (Abdel-Aal and Hucl, 2003) and rice (Ryu et al., 1998). However, quantitative data on sorghum anthocyanins and their antioxidant properties have not been published.

The most common anthocyanins in sorghum are the 3-deoxyanthocyanidins (Sweeny and Iacobucci, 1981; Gous, 1989), which include **22** and **24**. These anthocyanins have a small distribution in nature (Clifford, 2000) and are distinct from the more widely distributed anthocyanidins in that they lack a hydroxyl group at the C-3 position (Fig. 7) and exist in nature substantially as aglycones (Stafford, 1965; Clifford, 2000). The 3-deoxyanthocyanidins were also reported to be more stable in acidic solutions relative to the anthocyanidins (**28**–**33**) commonly found in fruits, vegetables and other cereals (Timberlake and Bridle, 1980; Sweeny and Iacobucci, 1981). This suggests a potential advantage of sorghum as a viable commercial source of anthocyanins.

Like tannins, effective quantification of anthocyanins is hampered by lack of appropriate standards, efficient extracting solvents and separation techniques. For example, acidified methanol (Gous, 1989; Lu and Foo, 2001) and aqueous acetone (Kallithraka et al., 1995; Garcia-Viguera et al., 1998) are reported as optimum solvents for anthocyanin extraction. Our work (Awika, 2003) showed that acidified methanol extraction resulted in higher anthocyanin values than aqueous acetone extraction for sorghum anthocyanins. Lu and Foo (2001) demonstrated that acetone, when used as a solvent, reacts with anthocyanins in a time dependent manner to form pyranoanthocyanins (**34a** and **b**; Fig. 8), which are

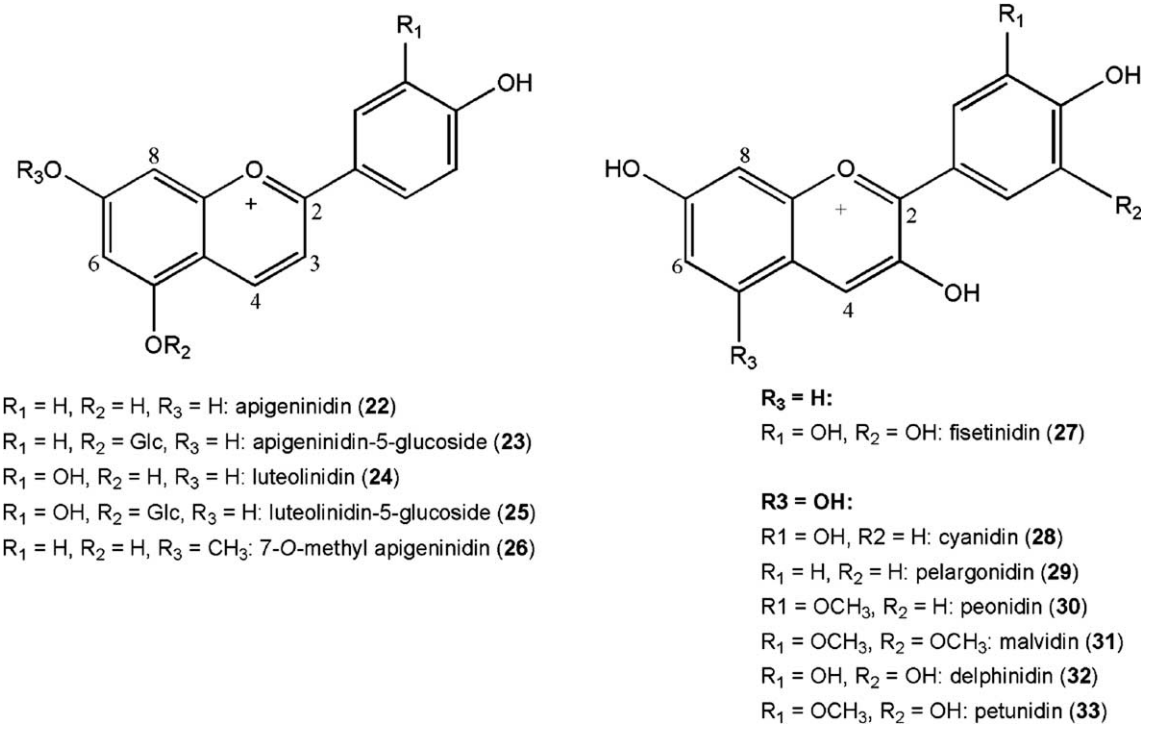


Fig. 7. The 3-deoxyanthocyanidins and their glucosides identified in sorghum compared to the anthocyanidins found in fruits, vegetables and other cereals (see Table 4).

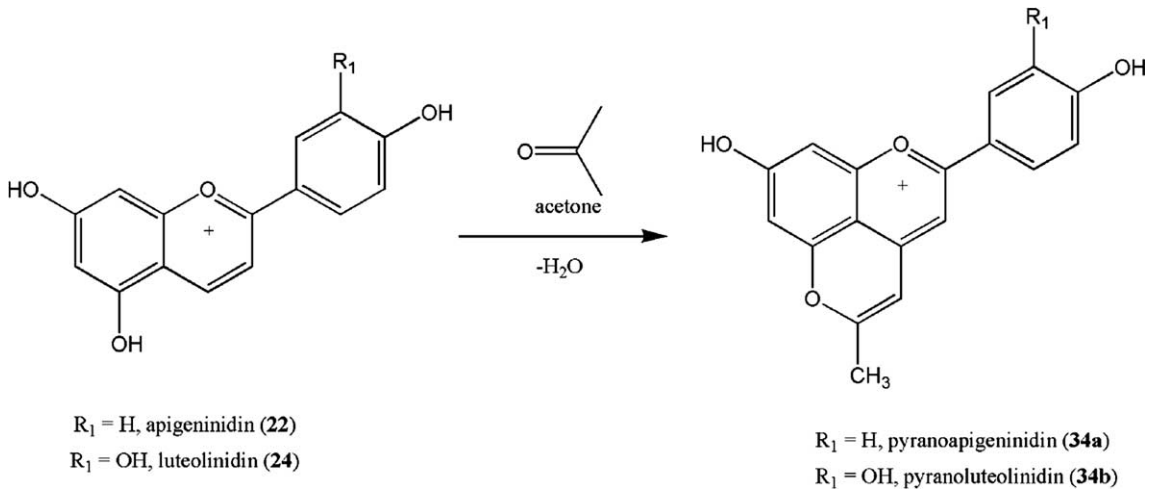


Fig. 8. Acetone interaction with sorghum anthocyanidins during extraction to form pyranoanthocyanidins as reported by Awika (2003). The reaction mechanism was proposed by Lu and Foo (2001).

Table 4
Anthocyanin content of sorghum brans relative to other cereals and common fruits and vegetables

Commodity	Content[a]	Major anthocyanidins	Source
Black sorghum bran	4.0–9.8	**22** and **24**	b
Brown sorghum bran	1.6–3.9	**22** and **24**	b
Red sorghum bran	3.3	**22**	b
Blue wheat bran	0.5	**28**	c
Purple wheat bran	0.2	**28**	c
Black rice grain	0–4.9	**28** and **30**	d
Purple corn	1.6	**28**	e
Blueberry	0.2–5.0	**31** and **32**	f, g
Red grapes	0.3–7.5	**30**, **31** and **33**	h
Black raspberry	1.7–4.0	**28**	f, i
Red raspberry	0.1–0.6	**28**	f, i
Strawberry	0.2–0.9	**29**	g, j
Elderberry	2.0–10.0	**28**	k
Red cabbage	0.3–0.9	**28**	f, h

(b) Awika (2003); (c) Abdel-Aal and Hucl (2003); (d) Ryu et al. (1998); (e) Cevallos-Casals and Cisneros-Zevallos (2003); (f) Wang et al. (1997); (g) Clifford (2000); (h) Bridle and Timberlake (1997); (i) Torre and Barrit (1997); (j) Garcia-Viguera et al. (1998); (k) Bronnun-Hansen et al. (1985).

[a] mg/g, fresh weight.

spectrally different from anthocyanins. We made similar observations in our laboratory (Awika, 2003). Additionally, anthocyanins are sensitive to pH and also polymerize with other phenols to form structures that are hard to effectively isolate or characterize (Remy et al., 2000). It is not certain whether the anthocyanins normally isolated are natural forms or are mostly solvent-modified derivatives.

The pH differential method of Fuleki and Francis (1968) is still the most common spectroscopy method for crude anthocyanin estimation. The method is rapid and does not involve expensive equipment, and estimates both monomeric as well as polymerized anthocyanins. However, the values by this method are greatly affected by the standard used. The HPLC procedure is also used for anthocyanin analysis in cereals (Ryu et al., 1998; Abdel-Aal and Hucl, 2003). The anthocyanin levels in sorghums compare well to those of fruits and vegetables commonly used as commercial anthocyanin sources (Table 4). Black sorghums generally have high levels of anthocyanins and may be a useful source of these compounds.

6. Other phenolic compounds from sorghum

Several other phenolic compounds have been isolated from sorghum (Table 5, Fig. 9). Naringenin (**35**) (a flavanone) was quantified in our laboratory as a major phenolic component of a bright red (TX 2911) sorghum variety (0.95 mg/g of bran) (Awika, 2003). Naringenin (**35**) and its glucoside were previously reported in sorghum by Gujer et al. (1986). Monomeric forms of proapigeninidin, apiforol (**36**) (Watterson and Butler, 1983), and proluteolinidin, luteoforol (**37**) (Bate-Smith, 1969) were also identified in sorghum leaves and grain. The presence of these units in sorghum grain polyflavans (tannins) was later confirmed by Gujer et al. (1986) and Krueger et al. (2003). Gujer et al. (1986) also reported taxifolin (**38**) and eriodictoyl (**40**) and their glucosides (**39** and **41**) in sorghum grain. The flavones, luteolin (**42**) and 7-*O*-methyl luteolin (**43**) were also reported in sorghum (Stafford, 1965; Misra and Seshadri, 1967). We did not find any information on presence of lignans in sorghum, but given these important phenolic compounds have been reported in many other cereals, there is a strong possibility that they exist in sorghum. Due to the biological benefits attributed to these compounds, there is a need to determine their presence and levels in sorghum.

Table 5
Major flavonoids isolated from sorghum grain

Compound	Reference
Anthocyanins	
23 and **25**	Nip and Burns (1969, 1971)
Anthocyanidins	
22 and **24**	Nip and Burns (1969, 1971); Gous (1989)
26	Pale et al. (1997)
27	Blessin et al. (1963)
Flavan-4-ols	
36	Watterson and Butler (1983)
37	Bate-Smith (1969)
Flavones	
42	Stafford (1965)
43	Misra and Seshadri (1967)
Flavanones	
35	Gujer et al. (1986); Awika (2003)
40	Kambal and Bate-Smith (1976)
41	Gujer et al. (1986)
Dihydroflavonol	
38	Gujer et al. (1986)
39	Gujer et al. (1986)
Proanthocyanidins (polyflavans)	
3	Gujer et al. (1986); Gu et al. (2002)
4	Gupta and Haslam (1978)
5	Gupta and Haslam (1978)
7 and **8**	Gujer et al. (1986); Krueger et al. (2003)
10	Brandon et al. (1982); Krueger et al. (2003)

7. Antioxidant properties of sorghum phenols and their bioavailability

Currently antioxidant activity is the most common in vitro parameter used to assess or predict potential benefits of plant phytochemical compounds. However, correlations between in vitro antioxidant activity and actual health benefits are unknown. Such in vitro antioxidant data ignore other potentially beneficial or harmful effects of phytochemicals like modification of enzyme activity and/or cell signaling pathways. For example, vitamin C and E, and the carotenoids, which were previously recognized only for their antioxidant characteristics, were shown to induce other biological responses (Astley, 2003). Antioxidant activity data are also hard to compare since there are no standardized methods; the methods currently used do not always agree in terms of ranking samples for antioxidant efficacy. Additionally, measured antioxidant activity in vitro tells us nothing about release and uptake of the compounds, as well as their distribution and metabolism in the body. However, antioxidant activity data still provide useful information for screening plant materials and products with desirable compounds and properties that can be used for further biological testing. In sorghum, phenol content correlates most strongly with antioxidant activity measured by various methods (Table 6, Fig. 10) indicating the phenols are largely responsible for the activity (Awika, 2003).

7.1. Condensed tannins

Tannins from sorghum show powerful antioxidant activity in vitro (Hagerman et al., 1998; Riedl and Hagerman, 2001). We also found that tannin (brown) sorghums had antioxidant activities higher than most non-tannin sorghums (Table 6). High MW tannins have the greatest antioxidant activity in vitro (on a molar basis) among natural antioxidants (Hagerman et al., 1998; Bors et al., 2000). This was attributed to the proximity of many aromatic rings and hydroxyl groups and the fact that tannins were not able to act as prooxidants (Hagerman et al., 1998). Procyanidin *o*-quinone is capable of producing oligomeric compounds through various coupling reactions that retain the number of hydroxyl groups, unlike the simple flavanoid *o*-quinones that can act as prooxidants by forming reactive oxygen species through futile redox cycling (Bors et al., 2000). A major concern about tannins though is that they may not be biologically effective antioxidants due to their large molecular size and their tendency to bind food molecules into insoluble complexes. However, Riedl and Hagerman (2001) demonstrated that even when complexed with proteins, sorghum tannins retained at least 50% of their antioxidant activity. Such protein-complexed tannins may serve as free radical sinks in the digestive system thus sparing other antioxidants.

Recent data show that tannins are more bioavailable than previously thought (Ross and Kasum, 2002). Deprez et al. (2001) reported that procyanidins, up to trimers, could be absorbed by the intestinal cell monolayer. Additionally, Spencer et al. (2000) reported that the interflavan bond in the procyanidins was unstable in a simulated gastric juice (pH 2) environment, which degraded the higher MW procyanidins to monomers and dimers. This potentially improves bioavailability of the procyanidins. However, Rios et al. (2002) reported that procyanidins are not degraded (are stable) during gastric transit in humans, hence only the monomers and dimers may be directly bioavailable. Still, part of the non-absorbed tannins is degraded by colon microflora into phenolic acids that are absorbed and may provide additional benefits (Pietta et al., 1997; Deprez et al., 2000; Tapiero et al., 2002).

7.2. Phenolic acids

Phenolic acids are more readily absorbed than other phenols from food due to their small molecular sizes (Scalbert et al., 2002). These compounds are readily extractable from fruits and vegetables, where they

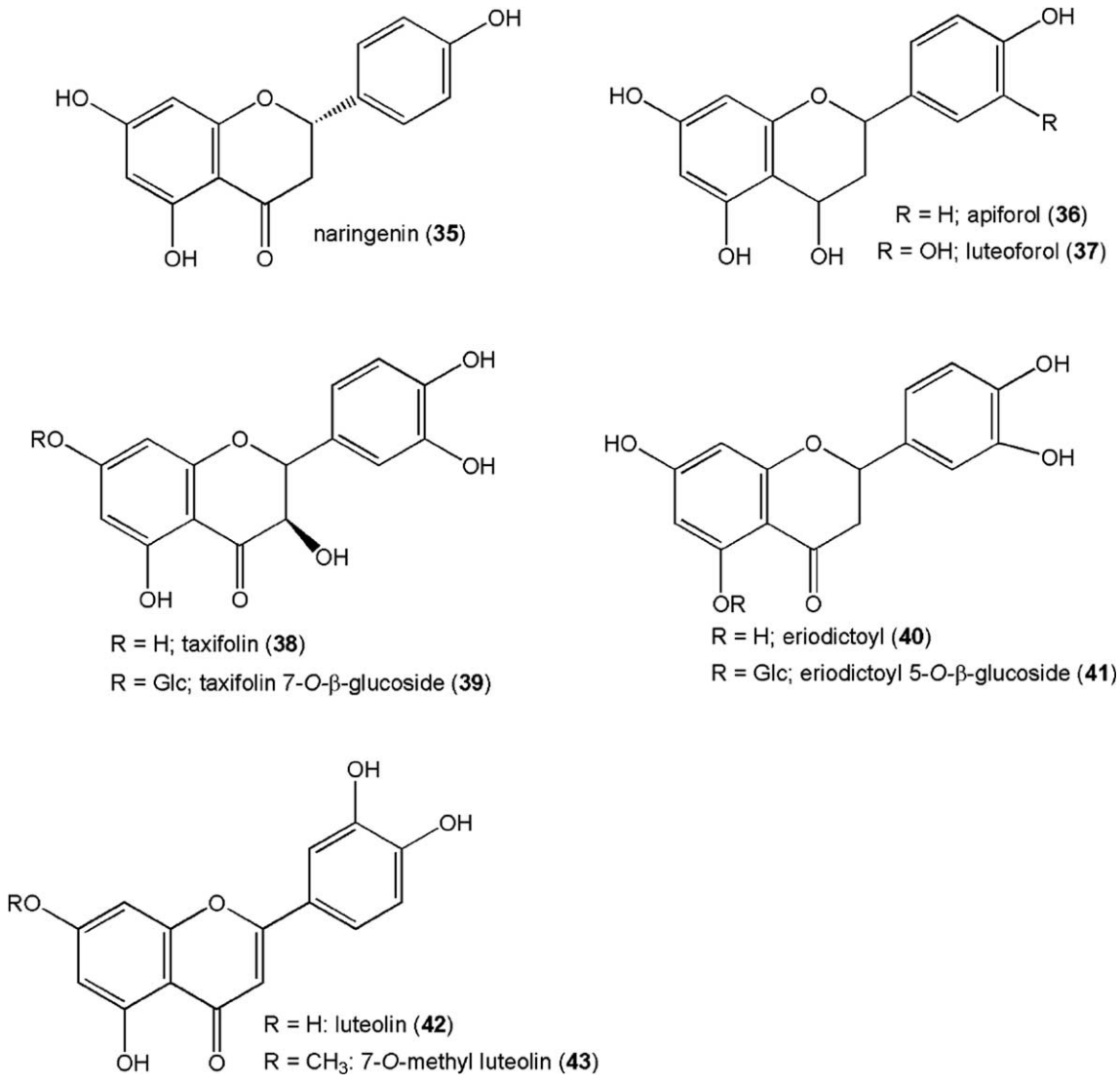


Fig. 9. Additional flavonoid monomers identified in sorghum.

Table 6
Phenol contents and antioxidant activities (μmol TE/g sample, dry wt) of sorghum and sorghum products measured by different methods

Sorghum	ORAC[a]	ABTS[b]	DPPH[c]	Phenols[d]
White grain	22	6	6	0.8
White bran	64	28	21	4.8
Red grain	140	53	28	6.6
Red bran	710	230	71	19.9
Black grain	220	57	41	6.4
Black bran	1000	250	184	26.0
CSC3*R28 grain	450	108	118	12.3
CSC3*R28 bran	2400	512	495	54.9
Sumac grain (brown)	870	226	202	19.8
Sumac bran (brown)	3100	768	716	66.3
CV%	6.8	3.5	5.3	5.97

[a] Oxygen radical absorbance capacity, fluorescein used as a probe.
[b] 2,2′-Azinobis (3-ethyl-benzothiaziline-6-sulfonic acid); activity was measured after 30 min reaction in pH 7.4 phosphate buffer saline.
[c] 2,2-Diphenyl-1-picrylhydrazyl; activity was measured after 8 h reaction in methanol.
[d] mg GAE/g (Folin–Ciocalteu method). Samples were extracted in 70% aqueous acetone. Adapted from Awika et al. (2003b).

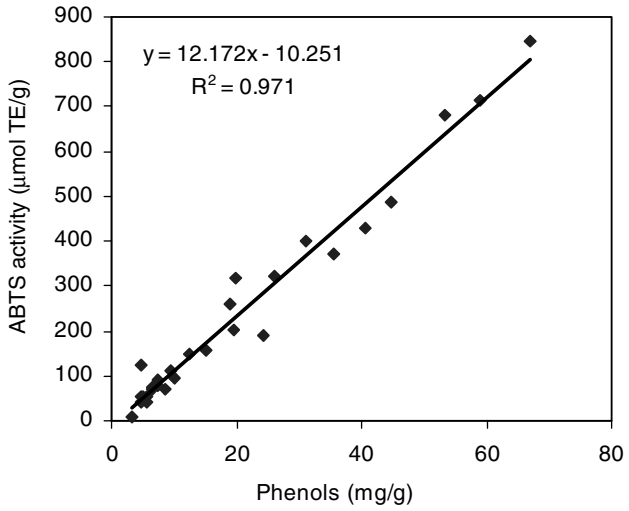


Fig. 10. Correlation between antioxidant activity (ABTS) and level of phenols in sorghum and sorghum brans. Samples were extracted in acidified methanol (1% HCl) (Awika et al., 2003b).

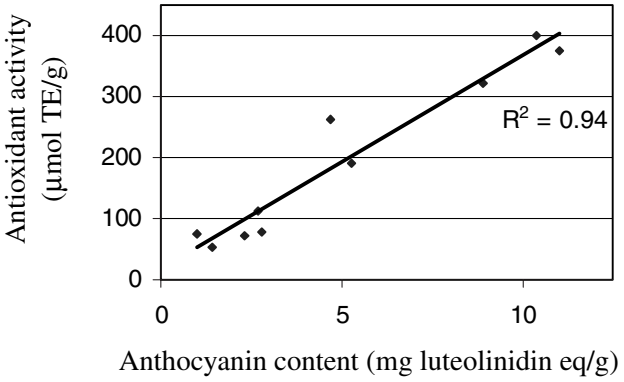


Fig. 11. Correlation between anthocyanin content and antioxidant activity (ABTS) of black sorghums and their brans. Anthocyanins were determined by the pH differential method. Acidified methanol (1% HCl) was the extracting solvent.

significantly exist in free forms. However, in sorghum and other cereals, most of the PA are esterified to cell wall components and can only be extracted in meaningful quantities by alkaline hydrolysis. Such bound forms of phenolic acids were, until recently, considered unavailable for absorption. However, Kroon et al. (1997) and Andreasen et al. (2001a,b) demonstrated that human colonic esterases (mostly of microbial origin) are capable of releasing esterified diferulates and other hydroxycinnamic acids from cereal brans. This implies that the bound PA are potentially bioavailable and the actual contribution of PA to health benefits associated with consumption of whole grains may be greater than previously assumed. In sorghum, bound PA generally account for over 85% of total PA (Hahn, 1984; Waniska et al., 1989). Adom and Liu (2002) also reported that more than 90% of ferulic acid (the most abundant PA in cereals) in corn, wheat, rice and oats exists in bound form.

7.3. Anthocyanins

Anthocyanins were reported to have low absorption compared to other flavanoids (Wu et al., 2002). However significant absorption of these compounds was demonstrated (Lapidot et al., 1999; Matsumoto et al., 2001; Milbury et al., 2002). Most of these data were obtained for fruit anthocyanins which are thought to contribute significantly to the health benefits of fruit consumption. We have not found any work reporting bioavailability of the 3-deoxyanthocyanidins commonly found in sorghum. This is probably because these relatively rare anthocyanins were not considered to be of economic interest.

The high antioxidant capacity of black sorghums and their brans were correlated with their anthocyanin contents (Fig. 11). Hence, anthocyanins may contribute significantly to any potential health benefits of these sorghums. Boveris et al. (2001) demonstrated that a 3-deoxyanthocyanidin (apigeninidin) isolated from soybean had strong dose-dependent quenching ability against ascorbyl and lipid radicals. Anthocyanins from fruits have been shown to possess several therapeutic benefits, including vasoprotective and anti-inflammatory properties (Lietti et al., 1976), anti-cancer and chemoprotective properties (Karaivanova et al., 1990), as well as anti-neoplastic properties (Kamei et al., 1995). The sorghum anthocyanins should be investigated for any unique health properties.

7.4. Comparing sorghum with fruits

Blueberries are considered an excellent source of antioxidants (Heinonen et al., 1998; Prior et al., 1998) and are commonly used as ingredients in various baked foods (e.g., muffins). Antioxidant activities of blueberries and other common fruits are compared to those of sorghum brans in Table 7. The sorghum brans show significantly higher values than the fruits. The high

Table 7
Antioxidant (ORAC) activity in sorghum brans relative to common fruits

Sample	ORAC[a] (dry wt)	Reference
Black sorghum bran	1010	Awika (2003)
Brown sorghum bran	2400–3100	Awika (2003)
Blueberries	87–870	Moyer et al. (2002)
Strawberries	356–400	Wu et al. (2002)[b]
Plums	452–600	Wu et al. (2002)
Grapes	100	Wu et al. (2002)
Watermelon	15	Wu et al. (2002)
Orange	80–150	Wu et al. (2002)

[a] µmol TE/g, using fluorescein as a probe.
[b] Unpublished data, courtesy of Wu, X., Arkansas Children's Nutrition Center, USDA-ARS, AR.

ORAC (oxygen radical absorbance capacity) levels in sorghum brans demonstrate a high potential of the sorghum brans compared to fruits as a source of natural antioxidants. The sorghum fractions can provide high antioxidant properties when used as ingredients in cereal-based foods. However, comparable antioxidant properties do not necessarily translate into comparable health benefits.

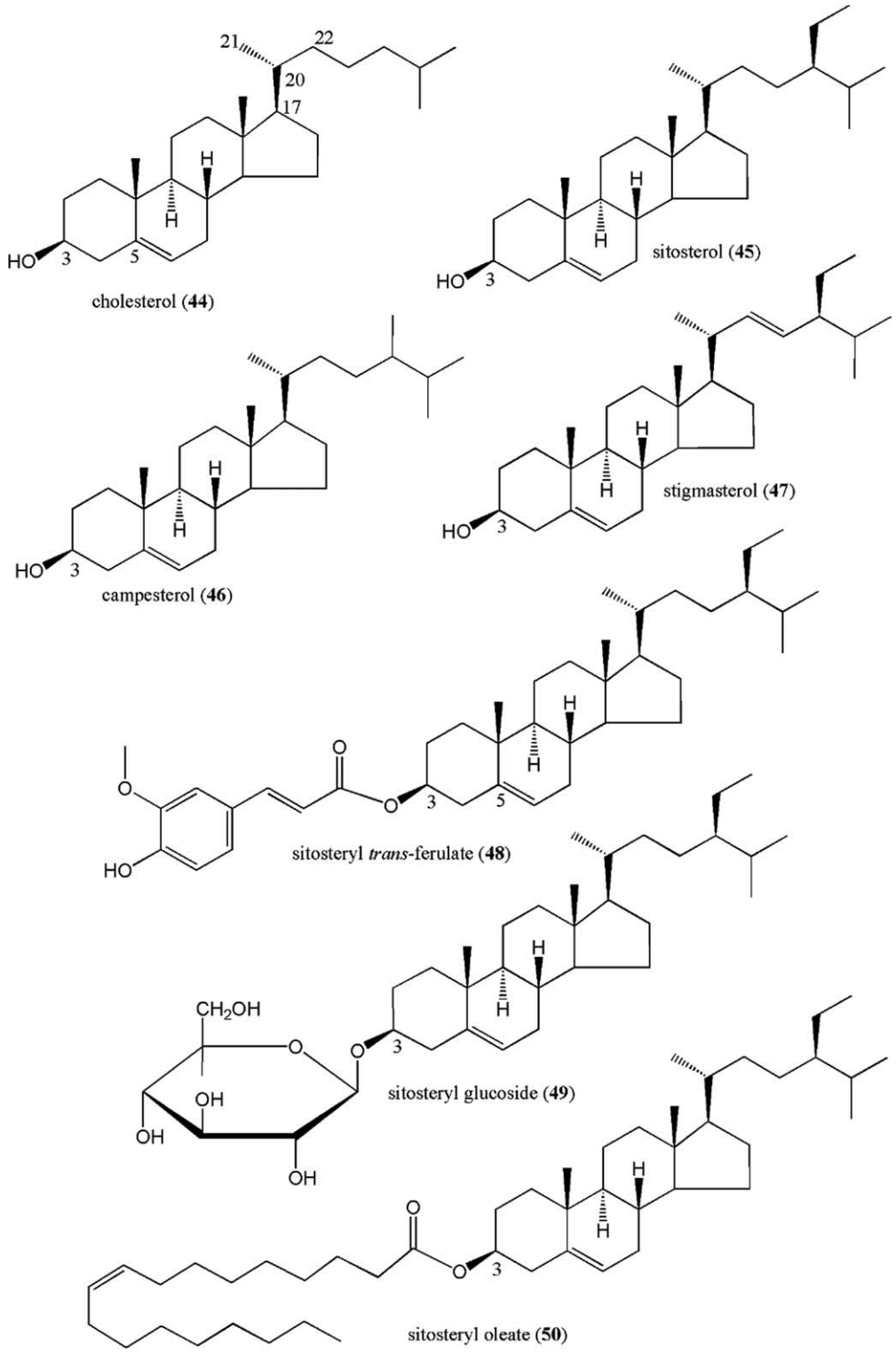


Fig. 12. Structure of cholesterol and common phytosterols, and their esters are found in sorghum and other cereals. *Notes*. Only sitosterol esters are illustrated, but campesterol and stigmasterol esters are also found in sorghum. Stanols are saturated forms of the sterols (single bond at position 5).

8. Phytosterols

Phytosterols are cholesterol (**44**)-like compounds that are structural components of plant cell membranes (Fig. 12). In cereals grains they are mostly found in bran and are extractable as part of bran oil waxes. There is a considerable interest in these compounds due to their promotion of cardiovascular health, especially through their cholesterol-lowering properties. Cereal brans reported to have high levels of these compounds include rice (Rogers et al., 1993; Dunford and King, 2000; Fang et al., 2003) and corn (Moreau et al., 1996; Singh et al., 2003). The phytosterols in cereals exist in free forms, as esters of fatty acids or hydroxycinnamic acids (usually ferulate), or conjugated with sugars (mostly glucose). In sorghum the free phytosterols identified include sitosterol (**45**), campesterol (**46**) and stigmasterol (**47**) (Avato et al., 1990). Esterified forms (**48**–**50**), with fatty acid chains of C14–C24 (Avato et al., 1990) and ferulates (Singh et al., 2003) were also identified in sorghum. Similar compounds are found in rice and corn brans (Rogers et al., 1993; Moreau et al., 1996). Stanol forms of the phytosterols (without a double bond at position 5) have also been reported in cereals (Ostlund, 2002). These compounds are not as commonly studied as the sterols, but are reported to offer similar health benefits. Ostlund (2002) reported that the stanols comprise about 10% of phytosterols in diet.

Quantitative data on sorghum phytosterols are limited. Singh et al. (2003) reported total phytosterol levels of 0.5 mg/g for sorghum grain, compared to 0.9 mg/g for corn (Table 8). The fiber fraction isolated from sorghum by a corn wet-milling procedure had 0.7–0.8 mg/g phytosterols. Values of 1.7–5.6 mg/g were reported for rice bran under optimized extraction conditions (Xu and Godgers, 2000). However, Singh et al. (2003) reported that the wet milling procedure they used resulted in a loss of 53–73% of the phytosterols in the isolated sorghum fractions relative to whole grain. Additionally the values they reported were only for the hexane-extractable (non-bound) forms of the compounds. Actual phytosterol values for sorghum may be higher with hydrolytic, or other optimized extraction procedures commonly used for cereals (Weihrauch and Gardner, 1978; Jonker et al., 1985; Piironen et al., 2002). Particle size of material (Moreau et al., 1996) as well as type of solvent (Xu and Godgers, 2000) are also known to significantly affect amount of extractable phytosterols. However, indications are that sorghums are a viable source of phytosterols (Table 8), and procedures to extract these compounds especially from sorghum bran should be explored.

Table 8
Phytosterol contents of cereals

Grain	Phytosterols (mg/100 g)	Reference
Sorghum	46–51[a]	b
Corn	70–88	c–e
Barley	55–76	e, f
Wheat	40–74	e–g
Oats	35–60	g
Rye	96	f
Brown rice	72	e
Buckwheat	96	e

(b) Singh et al. (2003); (c) Ostlund (2002); (d) Moreau et al. (1996); (e) Piironen et al. (2002); (f) Jonker et al. (1985); (g) Weihrauch and Gardner (1978).

[a] Non-bound phytosterols only (from hexane extracts).

9. Policosanols

Policosanols are a mixture of high molecular weight aliphatic alcohols (also called fatty alcohols) that are part of the wax components of plants. The compounds are currently commercially obtained from sugarcane wax by hydrolytic cleavage and further purification (Gouni-Berthold and Berthold, 2002). In sorghum, wax comprises about 0.2% of the grain, generally higher than in other cereals. The policosanols represent 19–46% of the sorghum wax, with octacosanol (C28) (**1**) and triacontanol (C30) (**2**) as the most abundant (Bunger and Kummerow, 1951; Dalton and Mitchell, 1959; Seitz, 1977; Avato et al., 1990). This translates to approximately 38–92 mg of policosanols in every 100 g sorghum grain. Weller and Hwang (2003) reported that these compounds may be the most commercially valuable component of sorghum grain based on their current market value. The policosanols have not been extensively studied in cereals, and most of the available literature is based on sugarcane wax policosanols. However, the compounds are gaining rapid popularity due to their cholesterol lowering ability and other sources, including sorghum, will definitely be of considerable interest.

The policosanols have cholesterol-lowering potency comparable to that of statins (currently popular but expensive and potentially harmful drugs) (McCarthy, 2002). Castano et al. (2002) reported that 10 mg/day of policosanol was more effective than 20 mg/day of lovastatin in reducing LDL cholesterol and raising HDL cholesterol levels. Other studies have shown similar benefits (reviewed by Gouni-Berthold and Berthold, 2002; Pepping, 2003). These authors also report that the policosanols present no toxic effects even at high doses. Other positive benefits provided by policosanols include effects on lipid peroxidation, platelet aggregation and smooth muscle cell proliferation (Fraga et al., 1997; Castano et al., 2002; Gouni-Berthold and Berthold, 2002). The policosanols are destined to gain importance as natural, safe and effective dietary alternatives to statin medication. Efficacy and economic potential of the sorghum policosanols should be investigated.

10. Sorghum phytochemicals and human health

10.1. Sorghum and cardiovascular disease

Cardiovascular disease (CVD) is the number one killer in the USA (Sistino, 2003). Various epidemiological data indicate that whole grain consumption significantly lowers mortality from CVD (Kushi et al., 1999; Slavin et al., 2000; Anderson, 2003). The phytosterols in the cereal brans are believed to contribute to beneficial effects. Other components of the whole grains, including polyphenols and fiber, also play a role in CVD prevention. For example, a cholesterol-lowering effect of tea and grape catechins and tannins is widely reported (Lin et al., 1986; Tebib et al., 1997; Santos-Buelga and Scalbert, 2000). We did not find any study that directly assesses effects of sorghum polyphenols on cholesterol.

In vivo studies on effects of sorghum on CVD are scarce. Klopfenstein et al. (1981) reported a cholesterol-lowering effect of low-tannin sorghum grain when fed to guinea pigs at 58% of diet. This effect was greater than that produced by wheat, rolled oats or pearl millet. More recently, Cho et al. (2000) found that sorghum and proso millet hexane extracts inhibit rat liver microsomal 3-hydroxy-3-methylglutaryl CoA (HMG-CoA) reductase in a dose-dependent manner. They also observed that fecal bile acid excretion, as well as levels of HDL cholesterol were increased, without a change in total cholesterol, when whole sorghum, proso millet or buckwheat were fed to rats at 30% of diet. Rooney et al. (1992) on the other hand observed that when sorghum bran was added to rat diet to achieve a 6% actual sorghum fiber, both high-tannin and non-tannin sorghums as well as wheat bran increased blood serum total cholesterol in rats. However, they did not measure changes in HDL and LDL cholesterol components. It is apparent that more data is necessary on how the diverse sorghum types affect cholesterol metabolism in controlled animal studies. Available data showing sorghum are a significant source of phytosterols and policosanols (Dalton and Mitchell, 1959; Seitz, 1977; Avato et al., 1990; Singh et al., 2003) will hopefully stimulate additional work in this field.

Benefits of sorghum to cardiovascular health may not be limited to positive effects on cholesterol. Lee and Pan (2003) demonstrated that dietary tannin-sorghum distillery residues inhibited 63–97% of hemoglobin-catalyzed oxidation of linoleic acid in cultured mullet fish compared to soybean (13%) and rice bran (78%). The authors also found that the sorghum residues significantly improved blood-thinning and erythrocyte membrane integrity of the fish blood cells during winter, thus maintaining normal blood fluidity and preventing RBC hemolysis induced by H_2O_2. They attributed the prevention of RBC hemolysis to the antioxidant activity of the tannins and other polyphenols present in the sorghum residue. Such protective effect on RBC was previously demonstrated for tea (Grinberg et al., 1997; Yang and Koo, 2000) and red wine (Pace-Asciak et al., 1996; Tedesco et al., 2000) polyphenols. The blood-thinning effect is also in agreement with epidemiological data that suggest that polyphenols from tea may reduce risk of stroke (Sato et al., 1989; Keli et al., 1996; Higdon and Frei, 2003).

In general several epidemiological studies show that red wine and tea consumption correlate with reduced risk of CVD (StLeger et al., 1979; Renaud and De Lorgeril, 1992; Higdon and Frei, 2003). The effects are largely attributed to the tannins and other polyphenols in these food products. There is no epidemiological data on tannin sorghum consumption and CVH in humans. This may partly be due to the fact that in the regions with high human sorghum consumption, CVD is not regarded as a major problem since there are other diseases that are of bigger concern. Such a study is important to help validate some of the evidence reported for in vitro and controlled animal studies, and determine the role sorghum may play in fighting CVD. The presence of unique polyphenols in sorghum (discussed earlier) is likely to produce positive benefits.

10.2. Sorghum and obesity

Obesity is an ever-increasing problem in the western world and is related to several disease conditions including CVD and diabetes. As a result a plethora of weight loss regimens, diets, pills, etc., has emerged. However, such regimens do not seem to produce the desired effects and obesity cases are on the rise (Hill et al., 2003; Wyatt, 2003). For example, in 1994 approximately 56% of US population was overweight and by 2000 the figure was 65% (Hill et al., 2003). Food (high calorie) consumption and lifestyle (lack of physical activity) account for the bulk of obesity cases. However, a radical change in either of these factors, as demanded by many weight loss strategies, is not easy to sustain for long periods of time, hence the failure of the strategies to produce an impact on obesity in the population.

Numerous reports on reduced weight gain of animals (rats, pigs, rabbits, poultry) fed high tannin sorghum are available (Jambubathan and Mertz, 1973; Featherson and Rogler, 1975; Cousins et al., 1981; Lizardo et al., 1995; Al-Mamary et al., 2001; Muriu et al., 2002). The mechanisms by which tannin sorghums reduce nutritive value include binding of food proteins (Haslam, 1974; Hagerman and Butler, 1981) and carbohydrates (Naczk and Shahidi, 1997) into insoluble complexes that cannot be broken down by digestive enzymes. Another mechanism involves the direct binding of digestive enzymes including sucrase, amylases, trypsin, chymotrypsin and lipases (Lizardo et al., 1995; Carmona et al., 1996; Nguz et al., 1998; Al-Mamary et al., 2001), thus inhibiting

their activity. Inhibition of intestinal brush-border bound amino acid transporters by sorghum tannins was also reported (King et al., 2000). There is evidence that the higher DP tannins are more involved in these interactions that the low DP ones (Bacon and Rhodes, 1998; Sarni-Manchado et al., 1999).

Effects of the sorghum tannins on animal weight gain depend on levels fed as well as animal species. Al-Mamary et al. (2001) found addition of 1.4% catechin equivalents (CE) sorghum to rabbit diet had no effect on growth rate and weight gain, whereas at a CE of 3.5%, there was a marked decrease in live weight gain and feed conversion ratio. Cousins et al. (1981) reported a 10% reduction in feed efficiency (relative to corn) when high tannin (3.1–3.4% CE) sorghums were fed to pigs. A low tannin sorghum (0.8% CE) had similar feed efficiency as corn. Lizardo et al. (1995) also observed a 10% reduction in weight gain when pigs were fed tannin-sorghum diet compared to corn. Broiler chicks fed low tannin sorghums (0.6–0.9% CE) had similar weight gain and feed efficiency compared to corn (Ambula et al., 2001). However, these authors observed an average of 50% reduction in weight gain when high tannin sorghums (2.7–3.5% CE) were fed to the broiler chicks. Such data suggest that there is a threshold for the inhibitory effects of sorghum tannins, and that animal species are affected differently.

Despite the evidence on animal studies, there is no reported work on how these attributes of tannin sorghums can be used to help lower calorie intake in overweight humans. Epidemiological data on humans in this area is also lacking, probably because in the places where tannin sorghums are consumed, obesity is often not a problem due to different lifestyles, daily food (calorie) intake and other factors. In certain cultures in Africa tannin sorghum is preferred to other cereals since it has a longer "staying power" in the stomach, i.e., offers durable satiety value. This property may be related to the slow digestibility and nutrient release from the tannin-complexed food matrix and should be investigated.

It is essential to assess the possibility of using this information to fight obesity in humans. Grain-based foods (breakfast cereals, bread, cookies, extruded snacks, etc.) are common and widely consumed in many parts of the world. High tannin sorghum grains and their milled fractions (bran) can find use as parts of ingredients in such foods. Brans from high tannin sorghums were incorporated in bread and cookies at up to 15% and 30%, respectively, without significant differences in texture or flavor profiles compared to whole-wheat products (Gordon, 2001; Mitre-Dieste et al., 2000). Cereal-based foods are a viable means of fighting obesity since they are consumed more consistently than most other foods and contribute a significant portion of the daily calorie intake. However, to use tannin sorghums in human diet to help fight obesity, the following need to be determined:

1. Whether the effects observed in animals are reproducible in humans.
2. The levels of tannins in sorghum necessary to produce desired effects.
3. Potential side effects of using tannin sorghums and their fractions at such levels, e.g., effects on availability of other essential micronutrients, especially divalent minerals like iron (which is chelated by tannins), and how the effects can be overcome.
4. How various food processing conditions affect potential activity of the tannins.

10.3. Sorghum and cancer

Positive effects of sorghum and/or millet consumption on cancer have been documented. Van Rensburg (1981) reported that sorghum consumption consistently correlated with low incidences of esophageal cancer in various parts of the world (including several parts of Africa, Russia, India, China, Iran, etc.) whereas wheat and corn consumption correlated with elevated incidences. Such regions also had deficiencies of certain minerals and vitamins in their diets. In attempting to explain this phenomenon, the author proposed (with considerable evidence) that the nutrient deficiencies were responsible for the high esophageal cancer incidences, and that sorghum and millet consumption promoted resistance to esophageal cancer risk. Chen et al. (1993) reported similar results from epidemiological data from Sachxi Province, China. These authors studied 21 communities within the province over a period of 6 years and found that regions that consumed highest amounts of sorghum, and to a lesser extent millet, had 1.4–3.2 times lower mortality from esophageal cancer than areas that primarily consumed wheat flour or corn. Consumption of other foods like alcohol, tea, meats and vegetables did not contribute significantly to esophageal cancer mortality. These evidences suggest presence of anti-carcinogenic compounds in sorghum that are either lacking or are not present in significant quantities in wheat or corn.

On the other hand, consumption of plants containing tannins (tea, sorghum, betel nuts, etc.) was previously implicated in incidences of cancer in the upper digestive tract (Morton, 1970, 1972). Morton (1970) implicated high incidences of esophageal cancer in certain parts of South Africa, China and Russia on consumption of high tannin sorghums. Oterdoom (1985) theorized that the carcinogenic effect of the high tannin sorghums reported by Morton (1970) was due to the fact that "tannins destroyed proteins both in the mucosa and in enzymes". However, the carcinogenic evidence suggested by Morton (1970) was criticized by Yu and Swaminathan (1987), and we concur, for lack of acceptable experimental design or control for confounding variables. Actually, Morton (1970) did not present any data to back her claim, but merely theorized that the common denominator in

regions with elevated risk of esophageal cancer was probably the consumption of plants containing tannins.

In vitro studies have also revealed anti-carcinogenic properties of sorghum. Grimmer et al. (1992) demonstrated anti-mutagenicity of sorghum polyphenol extracts. They found the high MW procyanidins (tannins) had the highest anti-mutagenic activity compared to lower MW tannins. Gomez-Cordovez et al. (2001) showed that sorghum tannins had anti-carcinogenic activity against human melanoma cells, as well as positive melanogenic activity (melanogenesis is believed to help protect human skin against UV irradiation damage (Eller et al., 1996)). The authors did not observe such melanogenic activity in red wine extracts. On the other hand, Parbhoo et al. (1995) reported that sorghum procyanidin extracts may induce cytochrome P-450, a protein that is capable of converting certain promutagens to mutagenic derivatives, in rat liver.

There is plenty of literature available (and often conflicting) on properties of various polyphenol-rich foods, especially tea and red wine/grapes in relation to various types of cancer in humans (reviewed by Chung et al., 1998; Santos-Buelga and Scalbert, 2000; Higdon and Frei, 2003). The overall trend suggests positive (anti-carcinogenic) effects of the polyphenol-rich products in diet rather than negative (carcinogenic) ones, but a consensus is far from deducible. For example, even though tea is reported as anti-carcinogenic in many in vitro and epidemiological studies, Higdon and Frei (2003) in an excellent review concluded that when all is considered, available data do not support the notion that tea is protective against cancer. Yang et al. (2001) also provided useful insight on the limitations of various cancer-related studies.

For sorghum, available data on cancer are too limited to draw reasonable conclusions. However, the corroborative epidemiological evidences reported by Van Rensburg (1981) and Chen et al. (1993) against esophageal cancer warrant some follow up. Additional in vitro data as well as controlled animal studies are necessary to understand how the levels and composition of polyphenols in sorghum affect cancer, and which specific components are responsible. Whole grain consumption has long been correlated with reduced risks to other forms of digestive tract cancer, especially colon cancer. How much of these effects are contributed by the dietary fiber or the phytochemicals concentrated in the brans of the grains is still unknown.

11. Perspective

Sorghum has a diversity of phytochemicals with a potential to significantly impact human health. The sorghum phytochemicals show high antioxidant activity against different free radicals in vitro relative to fruits and vegetables, and may offer similar benefits attributed to fruits and vegetables. Information on how sorghum phytochemicals affect human health is scarce. However, overall epidemiological evidence suggests sorghum has anti-carcinogenic properties when consumed regularly in diet. Animal studies also indicate that sorghum consumption promotes cardiovascular health better than other cereals. It is absolutely essential to determine if the positive effects observed in animals can be reproduced in humans, given cardiovascular disease is a leading killer in the west.

The most negatively reported effect of sorghum tannins in animals is reduced weight gain. However, with obesity a major and an ever-increasing problem in the developed world (more than 60% of Americans are reported to be overweight), this attribute of sorghum tannins has the potential of helping alleviate the problem. But we first need to determine if and what levels of tannin sorghum fractions in food can produce a desired weight reduction in humans and address any potential side effects that such levels may create.

Overall, the most effective (cost or otherwise) solutions to many major human health conditions lie in the natural components of foods we eat, rather than expensive medical intervention. The challenge is to find and incorporate a balance of the functional ingredients in everyday foods at adequate levels. Sorghum, which is currently underutilized, is definitely worth attention as a source of health-promoting phytochemicals for such foods. Some potential applications of sorghum include:

1. *Direct food use*. Whole sorghum grain can be used in baked, extruded and other cereal-based products (bread, cookies, expanded snacks, pasta, breakfast cereals, etc.) as partial or complete substitutes for other cereals. The sorghum brans can be used to fortify bread, cookies and other snacks, to improve the phytonutrient content, as well as dietary fiber and sensory properties. We previously demonstrated that pigmented sorghum brans in combination with other ingredients produce acceptable quality, dark-colored baked goods that appeal to consumers due to their natural 'healthy' appearance (no need for artificial darkening, e.g., as caramel is used to darken rye bread).
2. *Extraction of active components for commercial use*. Most phytochemicals in sorghum are concentrated in bran fractions. These fractions are easily separated from sorghum by decortication and can then be used to extract the various phytochemicals for dietary supplementation, food quality improvement or therapeutic applications. For example, sorghum anthocyanins, which were reported as more stable than the fruit anthocyanins, can be used as natural food colors with functional properties. The phytosterols and policosanons are found in sorghum bran and spent

distiller's grain in relatively large quantities; these sorghum fractions may provide a low cost source of these valuable compounds. Sorghum tannins can be extracted for use as antioxidant supplements as well as anti-caloric agents for obese individuals. Other phenolic antioxidants from sorghum can also be used as natural food preservatives, antioxidant supplements and therapeutic agents, among other uses.

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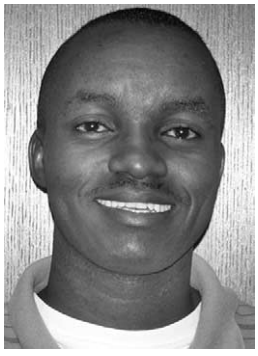

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