1. INTRODUCTION

1.1. Statement of the problem

Polyphenols are ubiquitous in plants and are an integral part of both human and animal diets (Bravo, 1998). Polyphenols protect crops from pathogens and predators by acting as phytoalexins and by increasing the astringency of food to make it unpalatable. To reduce bird damage, farmers grow condensed tannin-containing (tannin) sorghums, which are astringent during the immature stages when bird damage is highest (Bullard, Garrison, Kilburn and York, 1980). However, these agronomic advantages of condensed tannins to the farmer are accompanied by nutritional disadvantages (Butler, 1982; Chung, Wong, Wei, Huang and Lin, 1998). Tannins form complexes with proteins, starch and digestive enzymes causing a reduction in the nutritional value of food (Butler, 1982; Chung *et al.*, 1998). Nonetheless, the agronomic advantages of tannin sorghums outweigh such negatives as reduced nutrient availability or astringency (Awika and Rooney, 2004).

Interest in food phenolics has increased over recent years owing to their antioxidant properties (Bravo, 1998). High-tannin sorghums were found to have higher antioxidant capacity than is commonly found in fruits (Awika, Rooney, Wu, Prior and Cisneros-Zevallos, 2003b). Consumption of fruits, vegetables and cereals has been associated with lower risks of coronary heart disease and certain forms of cancer, due to the antioxidant properties of phenolic compounds, vitamins and dietary fibre in these foods (Steinmetz and Potter, 1996; Ness and Powles, 1997; Hollman and Katan, 1999; Ross and Kasum, 2002; Kamatha, Chandrashekarb and Rajinia, 2004). Thus, enhancing the content of phenolic compounds in plant foods through selective breeding and/or genetic improvement is viewed as a potent dietary option for disease prevention and control (Drewnowsky and Gomez-Carneros, 2000). However, phenolic compounds such as condensed tannins are well-known for eliciting negative consumer response (especially at high intensity) because of their dominant sensory properties, namely bitterness and astringency (Lesschaeve and Noble, 2005). Some of these bitter compounds include phenols found in tea, citrus fruits, wine and soy; triterpenes found in citrus fruits, and organo-sulphur compounds found in cruciferous vegetables like broccoli and cabbage (Reed, Tanaka and McDaniel, 2006). The objectionable sensory attributes of phenolic compounds may be the cause of the low

consumption of foods rich in these compounds (Drewnowski and Gomez-Carneros, 2000; Kamatha *et al.*, 2004).

Therefore, as research efforts focus on enhancing the content of phytochemicals like phenolic compounds in plant foodstuffs for health, it is necessary to determine how the sensory properties of these compounds affect consumer acceptance (Drewnowsky and Gomez-Carneros, 2000). Several studies have been carried out to identify and quantify phenolic compounds in sorghum (Kaluza, McGrath, Roberts and Schroder, 1980; Hahn, Faubion and Rooney, 1983; Awika, Dykes, Gu, Rooney and Prior, 2003a; Awika *et al.*, 2003b; Dykes, Rooney, Waniska and Rooney, 2005; Awika, McDonough and Rooney, 2005; Dlamini, Taylor and Rooney, 2007) as well as determining their antioxidant activity (Awika *et al.*, 2003a; Awika *et al.*, 2003b; Awika *et al.*, 2005; Dykes *et al.*, 2005; Dlamini *et al.*, 2007). However, quantitative assessment of the sensory attributes of phenolic compounds as well as their effect on the acceptability of sorghum foods is limited (Subramanian, Murty, Jambunathan and House, 1982; Yetneberk, de Kock, Rooney and Taylor, 2004; Yetneberk, Rooney and Taylor, 2005).

In eastern and southern Africa, traditional sorghum cultivars of moderate tannin content are widely grown and used as staple food and for alcoholic beverages (Awika and Rooney, 2004). According to these authors, some African cultures prefer tannin sorghums because the porridge from these sorghums 'remains in the stomach longer' and the farmer feels full for most of the day working in the field. These authors attributed this property to the slow digestibility and nutrient release from the tannin-complexed food matrix.

The question is, are there tannin sorghums that can address the needs of the sorghum producers, for whom condensed tannins have agronomic advantages, and simultaneously benefit the sorghum end users for whom condensed tannins are potentially potent antioxidants without compromising on their palatability?

1.2. Literature review

1.2.1. Sorghum (*Sorghum bicolor* [L] Moench)

Sorghum ranks fifth among the most important cereal crops in the world following wheat, rice, maize and barley (FAOSTAT, 2006). In the semi-arid tropics worldwide, sorghum is generally cultivated at a subsistence level and consumed as food by humans (Cothren, Matocha and Clark, 2000). Thus, it contributes significantly to the nutritional livelihood of impoverished populations of the world. Sorghum is eaten as porridge, fermented and unfermented breads, leavened and unleavened bread, snacks, non-alcoholic beverages and sorghum beer and malt (Murty and Kumar, 1995). In Japan, white tan-plant sorghums are processed into flour and other products such as snacks, cookies and ethnic foods (Awika and Rooney, 2004). In the USA, such sorghums are also gaining popularity as a substitute for wheat for people allergic to wheat gluten (Awika and Rooney, 2004).

1.2.1.1. Sorghum anatomical structure

The sorghum kernel is composed of three main parts: the outer covering (pericarp), the storage tissue (endosperm) and the embryo (germ) (Rooney and Miller, 1982) (Fig. 1.1; Taylor, 2003). The pericarp makes up 3-6%, the endosperm 84-90% and the embryo 5-10% of the grain depending on the kernel size. The sorghum kernel is called a caryopsis because the ovary wall dries and adheres strongly to the mature ovule. The pericarp originates from the ovary wall and is divided into the epicarp, the mesocarp, the cross cell layer and the tube cell layer. The epicarp is the outermost layer of the kernel and is divided into the epidermis containing pigments, and the hypodermis. The mesocarp is the middle part of the pericarp and may vary in thickness from thin (translucent) without starch granules to thick (chalky) with starch granules. The endocarp is the innermost part of the pericarp containing the cross and tube cells.

The endosperm consists of the aleurone layer, peripheral, horny (corneous) and floury portions (Fig. 1.1) (Rooney and Miller, 1982). The peripheral endosperm has starch granules embedded in a dense matrix of protein bodies and matrix proteins making the starch poorly available for hydrolysis. The corneous endosperm is located beneath the peripheral endosperm and is often called hard, vitreous or horny.

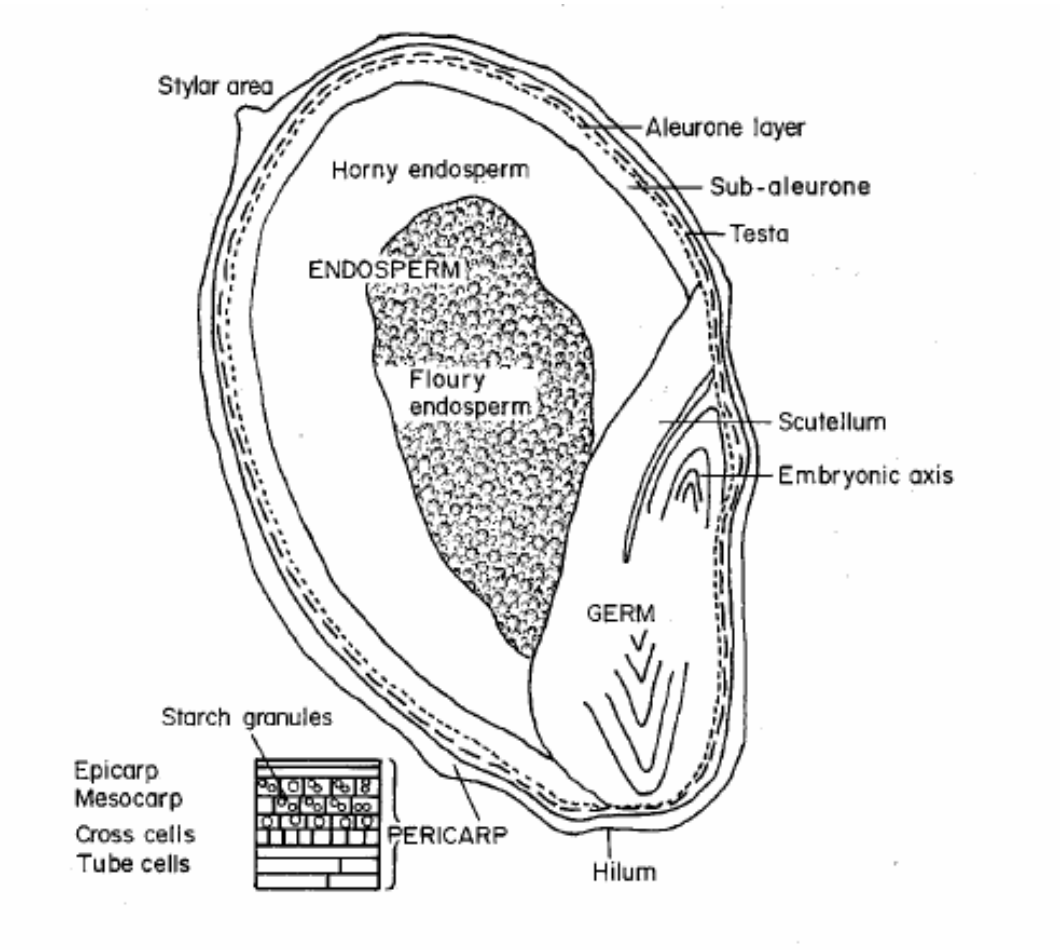


Figure 1.1. Cross-section of a sorghum kernel (Taylor, 2003).

The starch granules in this part of the endosperm are angular in shape with depressions where protein bodies were located. This part of the endosperm has strong starch-protein bonds and the starch granules often break easily rather than pull from the protein matrix. The floury endosperm is located in the inner most part of the kernel, and is composed mainly of starch with a smaller amount of protein bodies than found in the corneous endosperm. The relative proportion of corneous to floury endosperm within a sorghum kernel is often referred to as endosperm texture. The texture can be determined by visual examination of kernels cut longitudinally. A rating of 1 can be assigned to a kernel that contains very little floury endosperm and a rating of 5 can be assigned to a kernel which is predominantly floury (Rooney and Miller, 1982). Endosperm texture is important during processing. Sorghums with a corneous endosperm texture have higher milling yields

because the pericarp is more readily separated from the intact endosperm (Rooney and Miller, 1982). Sorghums with a corneous endosperm are also more resistant to insect attack during storage than those with a floury endosperm.

The embryo or germ is composed of two main parts, the embryonic axis and the scutellum (Fig. 1.1) (Rooney and Miller, 1982). The scutellum contains oil globules, protein bodies and a few starch granules. According to these authors, the embryo plays a major role in moisture uptake and mold susceptibility of the sorghum kernel.

Sorghum grain contains condensed tannins when there is the presence of a pigmented testa (Fig. 1.2) (Awika and Rooney, 2004). The pigmented testa is located just beneath the cross and tube cells (Rooney and Miller, 1982). The presence or absence of a pigmented testa is controlled by B_1 and B_2 genes and the testa is present when both B_1 and B_2 are dominant (B_1- B_2 -) (Rooney and Miller, 1982). These genes B_1- B_2 also affect pericarp colour when they are dominant in combination with the spreader gene (S-) and result with an intense pigment in the epicarp imparting a brown colour to the pericarp.

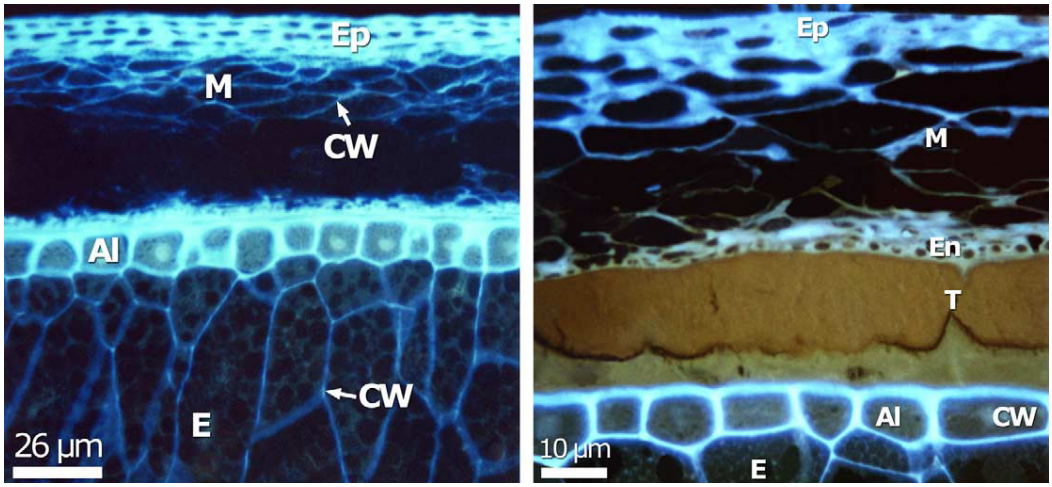


Figure 1.2. Fluorescence photomicrograph of sorghum bran cross-section, showing structural differences between a tannin-free sorghum (left) and a tannin sorghum with a pigmented testa (right). Al, aleurone layer; CW, cell wall; E, endosperm; En, endocarp; Ep, epicarp; M, mesocarp; T, pigmented testa (Awika and Rooney, 2004).

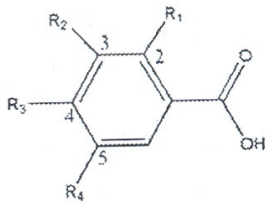
Sorghums have been classified into groups I, II and III based on the presence or absence of a pigmented testa (Rooney and Miller, 1982). Group I sorghums do not have a pigmented testa; group II have a testa (B_1- B_2- ss) and group III have a pigmented testa and a spreader gene (B_1- B_2- SS). According to Butler (1982) group I sorghums do not contain significant levels of tannins shown by low values of protein precipitation, Vanillin-HCl and anthocyanin production assays. Group II sorghum tannins are extractable in acidified methanol but not methanol alone. The typical high-tannin sorghums classified as group III sorghums, contain methanol-extractable tannins as well as group II type tannins extractable only in acidified methanol. Dicko, Hilhorst, Gruppen, Traore, Laane, Van Berkel and Voragen (2002) classified sorghums based on their whole grain tannin content as follows: low tannin sorghums $\leq$ 0.25%, medium tannin sorghums 0.26-0.5%, high tannin sorghums 0.51-0.75% and very high tannin sorghums $\geq$ 0.75% of tannin.

1.2.1.2. The chemistry of phenolic compounds of sorghum

Phenolic compounds are one of the most widely distributed groups of substances in the plant kingdom (Ross and Kasum, 2002). There are more than 8000 known phenolic compound structures, the common feature of which is an aromatic ring with at least one hydroxyl group (Ross and Kasum, 2002). There are more than 15 different classes of phenolic compounds in foods, ranging from simple phenolics with molecular weights of less than 500 to polymers of high (3000) molecular weight (Drewnowski and Gomez-Carneros, 2000).

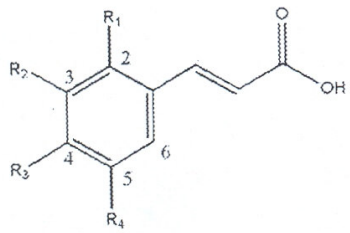
All sorghums contain phenolic compounds (Dykes *et al.*, 2005). Phenolic compounds are located mainly in the pericarp of the sorghum kernel (Awika and Rooney, 2004; Dykes and Rooney, 2006). Phenolic compounds identified in sorghum include phenolic acids, flavonoids and condensed tannins (Hahn *et al.*, 1983; Awika *et al.*, 2003a; Awika and Rooney, 2004). Phenolic acids in sorghum are mainly benzoic and cinnamic acid derivatives (Fig. 1.3) (Hahn *et al.*, 1983; Awika and Rooney, 2004). The benzoic derivatives have a C6-C1 structure and include gallic acid, *p*-hydroxybenzoic acid, vanillic, syringic and protocatechuic aids (Dykes and Rooney, 2006). Hydroxycinnamic acids have a C6-C3 structure and include coumaric, caffeic, ferulic and sinapic acids (Dykes and Rooney, 2006). Hahn *et al.* (1983) identified eight phenolic acids in sorghum namely: gallic, protocatechuic, p-hydrobenzoic, vanillic, caffeic, p-coumaric, ferulic and

cinnamic acids. According to these authors phenolic acids exist in sorghum as free forms mainly in the bran and bound forms esterified to cell wall polymers in the endosperm.



Benzoic acids

Gallic acid : $R_1 = H, R_2 = R_3 = R_4 = OH$
Gentisic acid : $R_1 = R_4 = OH, R_2 = R_3 = H$
Salicylic acid : $R_1 = OH, R_2 = R_3 = R_4 = H$
p-hydroxybenzoic acid : $R_1 = R_2 = R_4 = H, R_3 = OH$
Syringic : $R_1 = H, R_2 = R_4 = OCH_3, R_3 = OH$
Protocatechuic : $R_1 = R_4 = H, R_2 = R_3 = OH$



Cinnamic acids

Caffeic acid : $R_1 = R_4 = H, R_2 = R_3 = OH$
Ferulic acid : $R_1 = R_4 = H, R_2 = OCH_3, R_3 = OH$
o-coumaric acid : $R_1 = OH, R_2 = R_3 = R_4 = H$
p-coumaric acid : $R_1 = R_2 = R_4 = H, R_3 = OH$
Sinapic : $R_1 = H, R_2 = R_4 = OCH_3, R_3 = OH$

Figure 1.3. Basic structures of phenolic acids (benzoic and cinnamic acids) found in sorghum grain (Awika and Rooney, 2004).

Flavonoids are compounds with a C6-C3-C6 struture with two aromatic rings joined by a three carbon link (Fig. 1.4) (Dykes and Rooney, 2006). The basic structure of flavonoids allows a multitude of substitutions on the benzene rings A and B (Fig. 1.4.) (Hollman and Katan, 1999). There are two main subgroups of flavonoids namely, the 3-desoxyflavonoids (chalcones, flavanones, flavones) and the 3-hydroxyflavonoids (flavonols, anthocyanidins, leucoanthocyanidins and flavanols) (Brown, 1980). Flavanones, flavones, anthocyanins and flavanols have been identified in sorghum grain (Fig. 1.4) (Awika and Rooney, 2004). Flavanones identified in sorghum include naringenin and taxifolin and flavones include luteolin (Awika and Rooney, 2004).

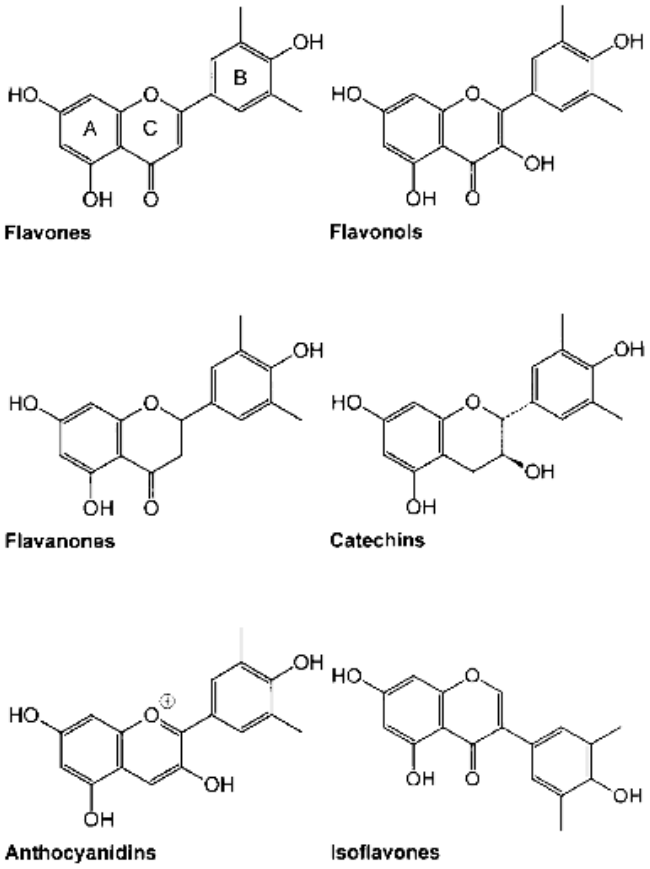


Figure 1.4. Structures of flavonoids (Hollman and Katan, 1999).

The most common anthocyanidins found in sorghum include 3-deoxyanthocyanidins: apigeninidin, luteolinidin and their derivatives (anthocyanins) (Fig. 1.5) (Awika and Rooney, 2004; Awika, Rooney and Waniska, 2004b).

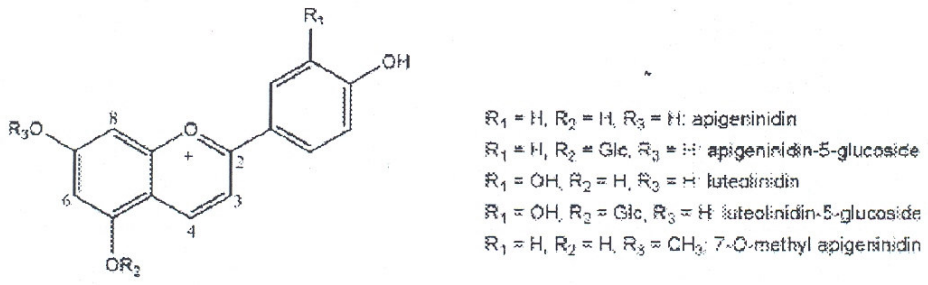


Figure 1.5. Basic structures of anthocyanidins (3-deoxycyanidins and their glucosides (anthocyanins)) found in sorghum grain (Awika and Rooney, 2004).

Monomeric flavan-4-ols such as luteoferol and apiforol may be present in either group I (tannin-free) or group III (tannin) sorghums (Fig. 1.6) (Butler, 1982). Luteoferol, with one OH-group, and apiferol, with two OH-groups, are flavan-4-ols of eriodictyl and naringenin respectively. A small amount of flavan-4-ols may be present in group II (tannin) sorghums. According to this author the presence of flavan-4-ols in sorghum grain is independent from that of tannins, but sorghums which contain flavan-4-ols but no tannin seem more abundant than those which contain tannin but not flavan-4-ols. Flavan-3-ol monomers identified in sorghum include catechin and epicatechin (Fig. 1.7) (Awika and Rooney, 2004).

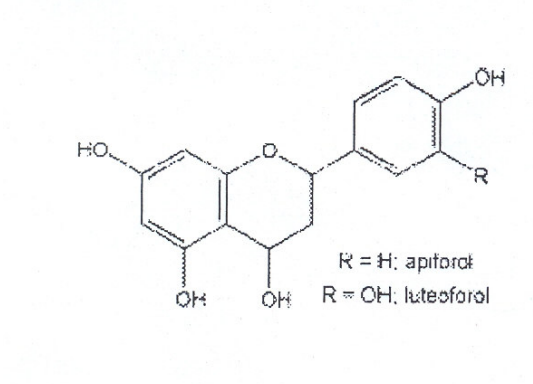


Figure 1.6. Basic structure of flavan-4-ols (apiforol and luteoforol) identified in sorghum (Awika and Rooney, 2004).

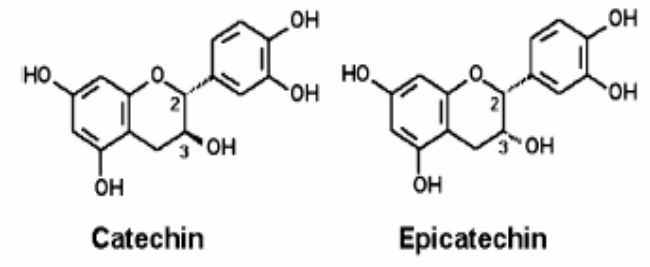


Figure 1.7. Basic structure of flavan-3-ols, (+)-catechin and (-)-epicatechin (Dixon, Xie and Sharma, 2005)

Plant tannins are defined as water-soluble phenolic compounds with molecular weights ranging from >500 to 3000 with the ability to precipitate gelatine and other proteins

(Swaim and Bate-Smith, 1962; Strumeyer and Malin, 1975). Structures of tannins vary in the nature of constitutive sub-units, degree of polymerization or chain length and linkage position (Vidal, Francis, Noble, Kwiatkowski, Cheynier and Waters, 2004). For instance major constituents of proanthocyanidins (condensed tannins) from grape seeds and skins include (+)-catechin, (-)-epicatechin and (-)-epicatechin-3-*O*-gallate and (-)-epigallocatechin (Souquet, Cheynier, Brossaud and Moutounet, 1996). Proanthocyanidins in sorghum are mainly composed of a series of condensed flavan-3-ols (Fig. 1.8) and flavan-3,4-diols molecules (Bullard *et al.*, 1980). According to Butler (1982) the condensed tannins in sorghum often exist as oligomers of five to seven flavan-3-ols which depolymerize into monomeric anthocyanidin pigments and thus are designated as proanthocyanidins.

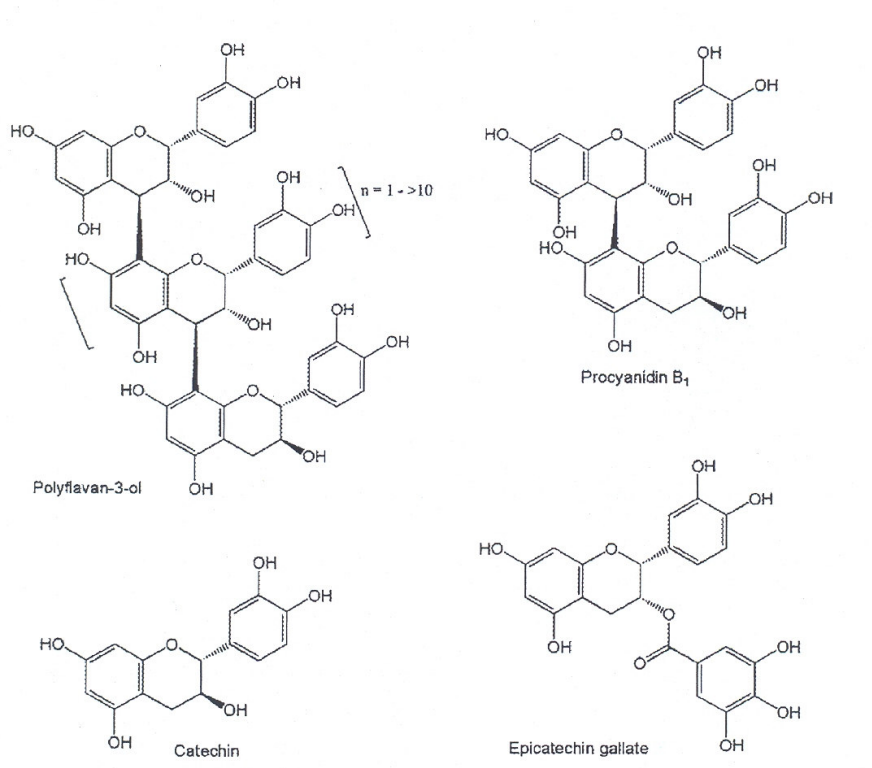


Figure 1.8. Structures of proanthocyanidins commonly found in sorghum grain (Awika and Rooney, 2004).

Tannins are a major phenolic component of sorghums with a pigmented testa (Awika *et al.*, 2003a). Some proanthocyanidins have been identified in sorghum with (-)-epicatechin chain extension units and a (+)-catechin chain termination units (Fig. 1.8) (Awika and

Rooney, 2004). Procyanidin B1 (epi (C4-C8) cat) is the most common dimer present in sorghum (Fig. 1.8) (Awika and Rooney, 2004).

1.2.1.3. Content of phenolic compounds in sorghum

The amount of phenolic compounds present in any particular sorghum cultivar is influenced by its genotype and the environment in which it is grown (Dykes *et al.*, 2005). These authors determined total phenol, condensed tannins, flavan-4-ols, anthocyanins and antioxidant activity of sorghum grain of clearly identified genotypes. The sorghum grain varied in pericarp colour, mesocarp thickness and the presence and intensity of the pigmented testa layer. Sorghum grains grown from purple/red coloured plants had higher total phenol content than tan plant types. Sorghums with a thick pericarp had higher total phenol content than sorghums with a thin pericarp. Sorghums with a pigmented testa gene B_1- B_2- and the spreader gene S had increased total phenol content, with B_1- B_2- S genes having the highest total phenol contents. Sorghums with a red pericarp contained flavan-4-ols such as luteoforol and apiforol, produced by flavanones, naringenin and eriodictyol. Consequently tan plant sorghums had the lowest content of flavan-4-ols, followed by purple/red plant sorghums with a thin pericarp. Purple/red plant sorghums with a thick pericarp had the highest content of flavan-4-ols. In tannin-free sorghums with a red pericarp, the total phenols were contributed mostly by the flavan-4-ols. Anthocyanin content followed the same trend as flavan-4-ols. Sorghums with a black pericarp contained the highest levels of anthocyanins. According to these authors, sorghums with a black pericarp are genetically red but turn black during maturation in the presence of sunlight.

Generally, cereals that contain condensed tannins (pigmented) like sorghum have higher levels of phenols than non-pigmented tannin-free sorghums and cereals like wheat and barley (Table 1.1 and Table 1.2) (Dykes and Rooney, 2007). Condensed tannin content levels reported in scientific literature using the Vanillin-HCl method range between 0.0-4.7 mg CE/g in tannin-free sorghums and 10-73 mg CE/g in tannin sorghums (Table 1.1). Phenol content levels reported in sorghum range from 0.8-5.6 mg gallic acid equivalents (GAE)/g in whole grain tannin-free sorghums and 11.7-22.5 mg GAE/g in whole grain tannin sorghums using the Folin-Ciocalteu method (Table 1.2). Phenolic content in the other cereals without a pigmented testa was comparable to that of the tannin-free sorghums (Table 1.2). Phenol content levels in bran are about four times the amount found in the

whole grain, supporting the view that phenolics are concentrated in the pericarp of sorghum (Table 1.2; Dykes and Rooney, 2007).

Table 1.1. Tannin content in different sorghum grains

Sorghum (whole grain)	Tannin content (mg CE/g dry wt) [1, 2]	Reference
Tannin-free sorghums	0.5-3.8	Awika and Rooney (2004)
Tannin-free sorghum (IS 2284)	4.7	Yetneberk *et al.* (2005)
Tannin sorghum (Sumac)	50.1	Awika *et al.* (2005)
Tannin sorghums	10.0-68.0	Awika and Rooney (2004)
Tannin sorghum (SC 103)	28.2	Awika *et al.* (2005)
Tannin sorghum (Seredo)	73	Yetneberk *et al.* (2005)
Tannin sorghum (Red Swazi)	33.6	Dlamini *et al.* (2007)
Tannin sorghum (NS 5511)	49.1	Dlamini *et al.* (2007)

[1] mg catechin equivalents (CE)/g (dry wt)

[2]Vanillin-HCl method

Processing procedures, such as decortication (Chibber, Mertz and Axtell, 1978; Beta, Rooney and Taylor, 2000; Awika *et al.*, 2005), fermentation (Towo, Mutuschek and Svanberg, 2006; Dlamini *et al.*, 2007), chemical treatment (Beta, Rooney, Marovatsanga and Taylor, 1999; Beta *et al.*, 2000), cooking (Butler, 1982; Towo *et al.*, 2006; Dlamini *et al.*, 2007) and extrusion cooking (Awika *et al.*, 2003a; Dlamini *et al.*, 2007) lower the total phenol and condensed tannin content of sorghum. On the other hand, germination (malting) increases the phenolic content of the sorghum (Butler 1982; Beta *et al.*, 1999). The apparent increase in phenolic content as germination proceeded is attributed to the production of non-tannin phenolic compounds by the developing roots and shoots (Beta *et al.*, 1999).

Table 1.2. Total phenolic content in the grain and bran of different sorghums and selected cereal grains

Type of Cereal	Total phenol content (mg GAE/g [1,2])		Reference
	Grain	Bran	
White sorghum (tannin-free)	0.8	4.8	Awika *et al.* (2003b)
White (Macia) (tannin-free)	2.7	N/D	Dlamini *et al.* (2007)
Red (Tx2911) (tannin-free)	4.8	19.5	Awika *et al.* (2004b)
Black (Tx430) (tannin-free)	5.6	26.1	Awika *et al.* (2004b)
Sorghum(tannin-free)	4.0	N/D	Ragaee, Abdel-Aal & Noaman (2006)
Tannin sorghum (NS 5511)	22.4	N/D	Dlamini *et al.* (2007)
Tannin sorghum (Red Swazi)	19.7	N/D	Dlamini *et al.* (2007)
Tannin sorghum (SC103)	11.7	48.7	Awika *et al.* (2004b)
Tannin sorghum (CSC3*R28)	12.9	56.6	Awika *et al.* (2004b)
Tannin sorghum (Sumac)	22.5	88.5	Awika *et al.* (2004b)
Pearl millet (standard)	3.4	N/D	El Hag, Tinay & Yosif (2002)
Pearl millet (Ugandi)	4.4	N/D	El Hag *et al.* (2002)
Rye	1.0	N/D	Ragaee *et al.* (2006)
Barley	0.9	N/D	Ragaee *et al.* (2006)
Hard wheat	0.6	N/D	Ragaee *et al.* (2006)

[1] mg Gallic Acid Equivalent (GAE)/g (Folin-Ciocalteu method)

[2] N/D – not determined

1.2.2. Harmful and beneficial effects of phenolic compounds

Phenolic compounds are thought to be both harmful and beneficial to the consumer (Chung *et al.*, 1998). Tannins appear to decrease the nutritive value of diets when added or when found naturally in high levels in certain foodstuffs (Strumeyer and Malin, 1975). This is because tannins are known to bind macromolecules such as proteins, starch and digestive enzymes (Butler, 1982; Haslam and Lilley, 1988). This tannin binding action causes a reduction in the nutritional and functional value of the bound constituents (Beta, 2003). According to Awika and Rooney (2004) the tannins in sorghum bind to and reduce digestibility of various food/feed nutrients, thus negatively affecting productivity of

livestock. According to Butler (1982), the tannin in high-tannin sorghums (2-3% dry weight) is sufficient under optimum conditions to bind considerably more protein than is present in the grain. Thus, it is likely that dietary tannins may be available to bind proteins of the digestive tract and thus interfere with digestion and absorption. This was demonstrated by Mamary, Habori, Aghbari and Obeidi (2001) who studied the extent of the *in vivo* inhibitory effects of two levels (1.4% and 3.5% catechin equivalent [CE]) of dietary sorghum tannins on rabbit digestive enzymes as well as mineral absorption. Addition of sorghum grain with 1.4% CE tannin content to the diet of rabbits did not significantly change the growth rate, food consumption or the feed conversion ratio. However, addition of sorghum grains with 3.5% CE tannin content significantly reduced the animals' body weight gain, feed conversion ratio, and slightly increased food consumption with respect to the control. Mamary *et al.* (2001) proposed that the lack of impact in the growth rates of animals fed the low-tannin (1.4% CE) sorghum grains may suggest the existence of a threshold-limit.

Since studies on the negative effects of phenolic compounds on sorghum foods have mostly focused on protein in general, Emmambux and Taylor (2003) investigated the interaction of sorghum-kafirin with phenolic compounds because like proline-rich saliva proteins, sorghum-kafirin is also rich in proline (Taylor, Von Benecke and Carlsson, 1989). Phenolic acids and flavonoid-type phenolics did not complex kafirin to form haze, whilst tannic acid and sorghum condensed tannins did. It was concluded that since condensed tannins in sorghum complex kafirin, this complexation might be involved in decreasing the protein digestibility of high-tannin sorghums. In contrast, the endogenous phenolic compounds found in tannin-free sorghums, such as flavonoids and phenolic acids, may not play a significant role in decreasing protein digestibility when such sorghums are wet cooked.

On the other hand, the phenolic compounds play an important agronomic role by reducing grain damage (pre-harvest and post-harvest losses) and bird predation (Strumeyer and Malin, 1975; Hahn *et al.*, 1983). The agronomic advantages such as resistance to bird predation are associated with high-tannin sorghums, which have low nutritional value for non-ruminants (Butler, Riedl, Lebryk and Blytt, 1984). Generally, higher concentrations of phenolic compounds are found in sprouts and seedlings than in the mature plant (Bravo, 1998; Chung *et al.*, 1998; Goldman, Kadar and Heintz, 1999). Mature grain of tannin

sorghums contains 2% condensed tannins or more while the immature grain has even higher tannin levels (Butler *et al.*, 1984).

Phenolic compounds are also thought to be harmful in that incidences of certain cancers, such as oesophageal cancer, have been associated with consumption of tannin-rich foods such as betel nuts and herbal tea (Chung *et al.*, 1998). Polyphenols in foodstuffs have been implicated in carcinogenesis (Lule and Xia, 2005). In the presence of oxygen, transition metal ions such as Cu and Fe catalyze the redox cycling of phenolics, leading to the formation of reactive oxygen species (ROS) and phenoxyl radicals that can damage DNA, lipids, and other biological molecules (Li and Trush, 1994; Lule and Xia, 2005). However, the dosage of tannins required to induce cancers probably far exceeds the level encountered during normal food intake; as such, tannins are not believed to be potent carcinogens (Chung *et al.*, 1998).

However, the role of phenolic compounds as antioxidants has been linked to low incidences of certain forms of cancer (Block, Patterson and Subar, 1992) and coronary heart diseases (Ness and Powles, 1997; Hollman and Katan, 1999). The cardio-protective effects of phenolic compounds stem from their ability to inhibit lipid peroxidation, chelation of redox-active metals and attenuation of other processes involving reactive oxygen species (Heim, Tagliferro and Babilya, 2002). According to Krishnaswamy and Polasa (2001) it has been established through epidemiological studies that vitamins A, C and E, β-carotene, selenium and calcium are micronutrients with cancer chemopreventive properties, while flavonoids, plant sterols, saponins, phytic acid, glucosinolates and terpenoids are non-nutritive cancer chemopreventers. The non-nutrient inhibitors of carcinogenesis have different modes of action (Krishnaswamy and Polasa, 2001). Some, like ferulic acid and ellagic acid, act as blocking agents, by inhibiting the activity of enzymes which convert pro-carcinogens to carcinogens. Others, like isoflavones and epigallocatechin gallate, act as suppressing agents, by restraining different steps in the metabolic pathways required in tumour development. Others like ellagic acid are trapping agents that physically react with carcinogens and detoxify them. Sources of these non-nutritive chemopreventers include cereal grains, vegetables, fruits and spices like turmeric, cloves, ginger, thyme, mustard, cinnamon and anise (Krishnaswamy and Polasa, 2001). This protective effect has been attributed to the antioxidant property of these compounds (Krishnaswamy and Polasa, 2001).

1.2.3. Sensory properties of phenolic compounds

Sensory attributes associated with smaller phenolic compounds like phenolic acids include sweet, sour, bitter and astringency (Peleg and Noble, 1995). Peleg and Noble (1995) investigated the sensory properties of phenolic acids (benzoic acid derivatives) commonly found in fruits, vegetables, grains and spices. These included salicylic acid (2-hydroxy benzoic acid), *m*-hydroxyl benzoic acid (3-hydroxy benzoic acid), gentisic acid (2,5-hydroxyl benzoic acid) protocatechuic acid (3,4-hydroxy benzoic acid) and gallic acid (3,4,5-trihydroxy benzoic acid) in water. Each of these compounds elicited multiple sensations including sweetness, sourness, astringency, bitterness and prickling. Although the compounds were structurally similar their sensory properties differed qualitatively and quantitatively. Gentisic acid was most sour, benzoic acid was highest in prickling sensation, salicylic acid was most astringent, *m*-hydroxyl benzoic acid was the sweetest and gentisic, benzoic and protocatechuic acids were most bitter.

Polyphenols of high molecular weight such as condensed tannins are predominantly bitter and astringent (Lesschaeve and Noble, 2005). Bitterness and astringency in some fruits and beverages, such as tea, cider and red wine, are elicited primarily by polyphenols (Lesschaeve and Noble, 2005). Flavan-3-ols, such as catechin, epicatechin and their oligomers and polymers (proanthocyanidins or condensed tannins) are abundant in tea and wine. Bitterness and astringency are sensory attributes mostly cited as the cause of condensed tannins in sorghum being objectionable (Bullard *et al.*, 1980; Hahn *et al.*, 1983; Asante, 1995; Mugula and Lyimo, 2000; Awika and Rooney, 2004; Yetneberk *et al.*, 2005). A bitter taste and after-taste has been reported in *injera* produced from tannin sorghum (Yetneberk *et al.*, 2004; Yetneberk *et al.*, 2005). As more of the pericarp was removed, the bitterness of the *injera* decreased and the overall rating improved.

Variation in phenol composition such as molecular size or chain length (monomer, dimer, trimer), extent of galloylation, small differences in configurations such as stereochemistry of the sub-units (catechin or epicatechin) and site of linkage between the sub-units (C4→C6 or C4→C8) produce significant differences in the intensity and duration of the bitterness and astringency of phenolic compounds (Arnold, Noble and Singleton, 1980; Peleg, Gacon, Schilch and Noble, 1999; Vidal, Francis, Guyot, Marnet, Kwiatkowski,

Gawel, Cheynier and Waters, 2003). Arnold *et al.* (1980) determined the bitterness and astringency of four grape seed phenolic fractions in wine: (I) catechin, (II) dimeric anthocyanogens, (III) trimeric and tetrameric anthocyanogens and (IV) condensed tannins in model wine. All the fractions were found to be bitter and astringent. All the fractions, including the most astringent fraction (IV), were more bitter than astringent. Astringency increased with increasing molecular weight from fraction I to IV ($p < 0.001$). The condensed tannin fraction (IV) was the most intensely bitter and astringent fraction. Peleg *et al.* (1999) also examined seven flavonoids (+)-catechin, (-)-epicatechin, dimer B3 (catechin (4→8) catechin), dimer B6 (catechin (4→6) catechin), dimer B4 (catechin (4→8) epicatechin), trimer C2 (cat (4→8) cat (4→8) cat) and trimer C (cat (4→8) cat (4→8) epi) in water (Fig. 1.9). (-)-Epicatechin was significantly more bitter and more astringent and had a longer duration than its chiral isomer (+)-catechin. Difference in molecular size was the major factor influencing the sensory properties of bitterness and astringency in the phenolic compounds investigated. As the degree of polymerization increased, maximum bitterness intensity and duration decreased, whereas astringency increased. The monomers were more bitter than they were astringent while the trimers were more astringent than they were bitter. The bond linking the monomeric units also influenced both bitterness and astringency. The catechin-catechin dimer linked by a 4→6 bond (B6) was more bitter than both catechin-catechin (4→8) (B3) and catechin-epicatechin (4→8) (B4) dimers. Catechin-catechin (4→8) (B3) was less astringent than both catechin-catechin (4→6) (B6) and catechin-epicatechin (4→8) (B4). In agreement with the findings of Arnold *et al.* (1980) relative astringency increased with increasing molecular weight. Contrary to the findings on Arnold *et al.* (1980), Peleg *et al.* (1999) reported that procyanidin fractions with higher degree of polymerization (DP) were less bitter than fractions with lower DP. Vidal *et al.* (2003) carried out a descriptive sensory analysis on a range of purified apple, grape seed and grape skin tannin fractions differing in chain length and degree of galloylation in a model wine. The degree of polymerization appeared to be the most discriminatory variable among the fractions. Overall astringency increased with increasing chain length. Increased degree of gallolylation of the fractions increased a rough sensation associated with coarseness, drying and chalkiness. Like Arnold *et al.* (1980), Vidal *et al.* (2003) also reported that chain length did not affect bitterness perception. The bitterness scores were very low for all the samples.

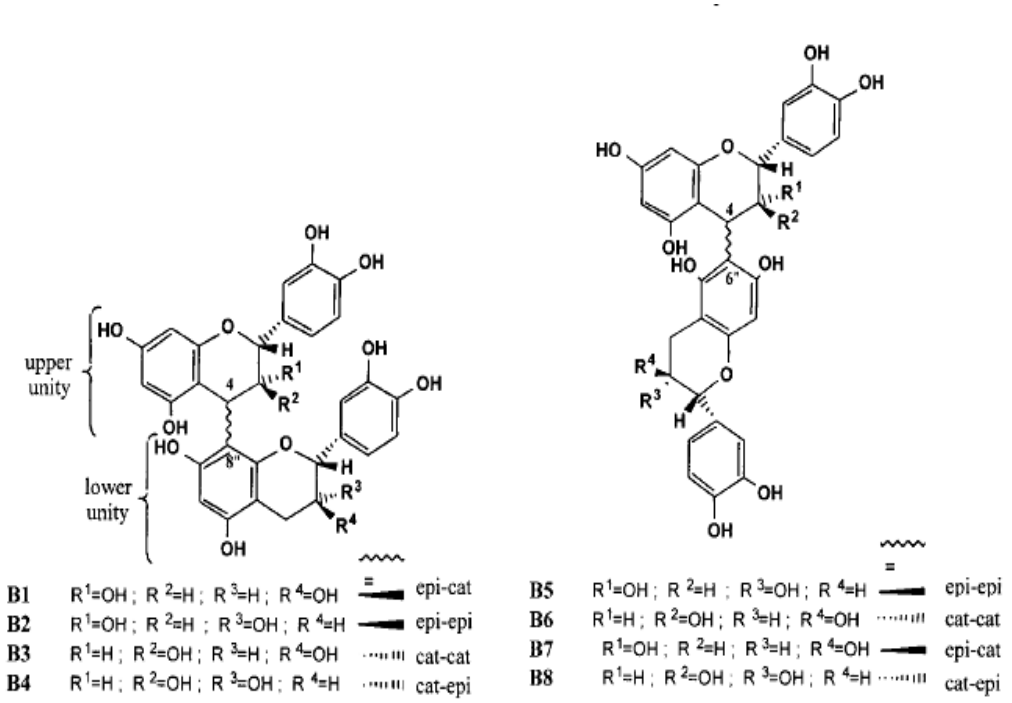


Figure 1.9. Molecular structures of procyanidin dimers with a C4-C8 linkage (B1-B4) and dimers with a C4-C6 linkage (B5-B6) (De Freitas and Mateus, 2001).

Since the astringency sensation is important in many beverages, Valentová, Skrovánková, Panovská and Pokorný (2002) compared the time dependence of astringency sensations in model aqueous solutions (tannic acid and (+)-catechin solutions) and different beverages (orange drink, model vermouths, red wines and Ceylon black tea), and investigated the interactions of astringency with other basic tastes and ethanol. Astringency was detected without difficulty in the presence of other tastes. The time dependence of the astringency in black tea was similar to that of (+)-catechin in aqueous solutions. The effect of astringent substances in wine was much more difficult to ascertain than in model solutions. Wine, particularly red wine, contains many phenolic substances that may taste either astringent or bitter or both (Lea and Arnold, 1978). Certain relations exist between the astringency and bitterness as most phenolic substances may taste both astringent and bitter (Lea and Arnold, 1978). Valentová *et al.* (2002) found that the time dependence of astringency in beverages was similar to that of bitterness. Differences among assessors were similar for the two sensations (bitterness and astringency) and dependent on saliva flow. Experienced assessors could distinguish both sensations (bitterness and astringency)

in the presence of each other. The development of astringency and its fading after swallowing followed an exponential course, but it was different for different beverages.

Bitterness and astringency contribute to the good taste of ciders and wines (Lule and Xia, 2005). Lea and Timberlake (1974) reported that as the bitterness and astringency in ciders increased, the concentrations of oligomeric and polymeric flavan-3-ols also increased. They concluded that the highly polymerized material was primarily responsible for both astringency and bitterness, while the isolated monomers, dimers and trimers contributed only slightly to these sensations. Lea and Timberlake (1974) and Lea and Arnold (1978) also reported that astringency and bitterness of cider procyanidins increased with increasing molecular weight. No specific polyphenol fraction was found to be exclusively responsible for bitterness and astringency (Lea and Arnold, 1978; Lea and Arnold, 1983). Kallithraka, Bakker and Clifford (1997c) reported (+)-catechin and (-)-epicatechin as bitter and astringent in a model wine and real wine. (-)-Epicatechin was found to be more astringent than (+)-catechin on an equal weight basis. According to Noble (1995) young wines with a high amount of smaller oligomers (dimers and trimers) are described as 'hard' (bitter and astringent), whilst older wines with more polymerized phenols (8-10 units) are described as 'soft' (less bitter and mainly astringent). Yamanishi (1990) reported that (+)-catechin and (-)-epicatechin in tea were exclusively bitter, while (-)-epicatechin gallate had an astringent threshold of 50 mg/l.

Delcour, Vandenberge, Corten and Dondeyne (1984) determined the taste detection thresholds of polyphenolics in deionised water. Phenolics evaluated were (+)-catechin (flavanol), procyanidin B3 (catechin-catechin (4→8) dimer), quercetin dehydrate (flavonol), tannic acid (hydrolysable tannin) and a mixture of trimeric and tetrameric procyanidins. The detection threshold depended on their degree of polymerization. The higher the molecular weight of these substances the lower their detection threshold values. For instance, the detection threshold levels were: 46.1 mg/l for (+)-catechin, 17.3 mg/l for procyanidin B3, 8.9 mg/l for quercetin dehydrate, 14.1 mg/l for tannic acid and 4.1 mg/l for a mixture of trimeric and tetrameric procyanidins.

1.2.4. Bitterness

There are five basic tastes: sweet, sour, salty, bitter and umami (Kim, Breslin, Reed and Drayna, 2004). According to these authors these tastes are mediated by taste receptor proteins residing on the surfaces of taste receptor cells (TRCs) within the taste buds of the tongue. At a molecular level, taste is a gustatory stimulus that stimulates a taste receptor cell (TRC), which in turn conveys the message to a sensory neuron (McLaughlin and Margolskee, 1994). A nerve impulse then relays the message to the gustatory centres of the brain where it registers as a taste. Stimuli for a single taste may come from several different types of chemicals; in the case of bitterness for example caffeine, a purine; morphine, an alkaloid; and potassium chloride, a simple salt are all bitter. The first step in taste recognition takes place in the taste pore, where molecules that are perceived to have taste (tastants) enter the taste pore and interact with receptor molecules and channels within the microvillar membrane of the TRCs (McLaughlin and Margolskee, 1994). Besides detecting taste stimuli, TRCs also convey taste information to the brain through a neuron (McLaughlin and Margolskee, 1994). The neurons make contact with taste cells at the synapse, a specialized region between the receiving end of the neuron, and the sending end of the taste cell. Information is then passed from the TRC to the neuron via chemical transmitters called neuro-transmitters secreted by the taste cell into the synapse. When the neurons detect these transmitters they react to them with a nerve impulse that is transmitted to the brain (McLaughlin and Margolskee, 1994). This process of receiving sensory information that is translated into a useful signal to the nervous system is called sensory transduction (McLaughlin and Margolskee, 1994).

1.2.4.1. Bitter taste transduction and other basic tastes

According to Kinnamon (1996), research data suggest that different mechanisms are utilized for the transduction of different taste stimuli. Ionic taste stimuli such as salts (Na^+), acids and some bitter compounds interact directly with apically located ion channels to depolarize taste cells. Amino acids, sweet stimuli and other bitter compounds bind to specific membrane receptors usually coupled to G-proteins and secondary messenger systems. According to Herness and Gilertson (1999) sour and salty tastes depolarize TRCs by directly interacting with ion channels. In contrast amino acids, sugars and other compounds perceived as sweet and most bitter compounds activate G-protein-coupled receptors (GPCRs) (Kim *et al.*, 2004). According to Kinnamon (1996) both H^+ and Na^+ use the same channel. However since salt (Na^+) can be distinguished from the taste of acids, other mechanisms must exist for acid transduction. Other mechanisms of acid

transduction include a proton-gated Ca^{++} channel and a proton transporter. Some bitter compounds are transducted by the same apical K^+ conductance involved in acid transduction (Kinnamon, 1996). Quinine and divalent salts like $CaCl_2$ directly block the K^+ conductance while K^+ salts permeate the conductance to depolarize taste cells. Specific membrane receptors appear to be required for the transductance of sugars, synthetic sweeteners, amino acids and most bitter compounds. Most of these receptors are coupled to G- proteins and second messengers. Thus, bitter stimuli interact with both apical ion channels and specific membrane receptors for transduction (Kinnamon, 1996). It is not clear whether the bitter taste of procyanidins (flavan-3-ols) is a result of receptor or surface membrane interaction (Peleg *et al.*, 1999). Regardless of whether bitterness of procyanidins is elicited by interaction with a specific bitter membrane-bound receptor or through surface membrane interactions, increasing the size of procyanidins decreased their bitterness. The monomers were perceived as more bitter than the dimers which were in turn more bitter than the trimers. These authors suggested that this could be a result of increased steric interference reducing the strength of interactions between the flavonoid and the receptor or the receptor membrane thus causing the trimers to be perceived as least bitter.

The bitter taste appears to be the most complex taste quality in humans, given the variety of chemically diverse structures that elicit bitterness on an apparently large number of gene encoding receptors (McLaughlin and Margolskee, 1994; Kim *et al.*, 2004). Bitter taste can be detected at very low concentrations (Glendinning, 1994). It is believed to have evolved to enable organisms to detect and avoid environmental toxins (Glendinning, 1994; Kim *et al.*, 2004). There are many compounds identified as tasting bitter including inorganic salts (KCl), amines (denatonium), amino acids (tryptophan), peptides, alkaloids (quinine and morphine), acetylated sugars (sucrose octa-acetate), flavanols/phenols (epicatechin), carbamates/thioureas (6-n-propylthiouracil [PROP] and phenylthiocarbamide [PTC]), to name a few (Keast and Breslin, 2002). In order to be able to taste such divergent structures, mammals have evolved multiple mechanisms which have an affinity for the divergent chemical structures. According to McLaughlin and Margolskee (1994), some bitter compounds have an affinity for fatty acid molecules in the cell membrane and are termed lipophilic, whilst other molecules are hydrophilic. Thus, it is assumed that bitter compounds share taste receptor sites and transduction mechanisms,

since it seems improbable that each of the thousands of bitter compounds would have its own unique transduction sequence (Keast and Breslin, 2002).

There are many substances for which different individuals show great differences in their taste thresholds. Yokomukai, Cowart and Beauchamp (1993) investigated individual differences among humans in their perception of different bitter tasting compounds. They found that sensitivity to quinine sulphate (QSO_4) and sensitivity to urea were unrelated. Subjects who were highly sensitive to one bitter compound could be insensitive to another. Out of the 52 subjects tested, 18 found these compounds to be equally bitter, 17 found QSO_4 to be more bitter than urea, and 17 found urea to be more bitter than QSO_4. QSO_4-sensitive subjects found the bitterness of caffeine and sucrose octa-acetate (SOA) to be more than that of magnesium sulphate; whereas the reverse was true for the urea-sensitive subjects. Thus, it was concluded that the results support the existence of multiple bitter transduction sequences, in that individual differences in response to various bitter compounds may reflect differences in the relative availability of specific transduction sequences. Delwich, Buletic and Breslin (2001) investigated whether classes of bitter transduction processes in the general population might be revealed by examining and correlating individual differences in sensitivities to bitter compounds namely: quinine HCl, caffeine, (-)-epicatechin, L-phenylalanine, L-tryptophan, tetralone, magnesium sulphate, urea, SOA, denatonium benzoate and PROP. It was assumed that bitter tasting compounds that cluster together as a function of the subject's perceptual sensitivities share some common physiological mechanism. The subjects rated the bitterness intensities of different compounds followed by ranking the intensity of bitterness of the compounds from the weakest to the strongest. By examining the subject's (n=26) individual differences in ratings and rankings of the bitter compounds, four clusters emerged. The first group included urea, phenylalanine, tryptophan and epicatechin, the second group included quinine, caffeine, SOA, denatonium benzoate and tetralone, the third group included magnesium sulphate and the fourth included PROP. From these results, it was concluded that bitterness appears to be transduced in humans via several different transduction mechanisms. A separation was found between those that contain at least one primary amine (group 1) and those that contain at least one methyl group (group 2). Magnesium sulphate does not contain methyl groups or amines and was thus seen as an isolated compound. The panellists differed by their sensitivity to PROP and also differed in their bitterness ratings; however they did not differ in rankings. Therefore it was

concluded that there are subjects who possess diminished absolute sensitivity to bitter stimuli but do not differ from other subjects in the relative sensitivity to these compounds.

1.2.4.2. Genetic variation

Genetic variation in taste perceptions has been investigated by different researchers since Fox (1931) accidentally discovered that his colleague could taste the bitterness of a chemical compound he was working with, phenylthiocarbamide (PTC), whilst he found it tasteless. Blakeslee and Fox (1932) solicited volunteers to taste PTC by posing the question: "What taste world do you live in?" Twenty eight percent of the people found it to be tasteless whilst the remainder described it as bitter to varying degrees. The responses essentially showed a bimodal distribution that distinguishing two phenotypes: tasters and non tasters. In subsequent studies, chemical compounds sharing the H-N-C=S chemical moiety like PROP also showed the same bimodal threshold distribution, leading to the designation of 'tasters' for the more sensitive and 'non tasters' for the less sensitive individuals (Hall, Bartoshuk, Cain and Stevens, 1975). The incidences of PROP taste 'blindness' varies around the world. In western Africa about 3% of the population are non tasters, > 40% in India, and about 30% of the adult Caucasian population in North America are non tasters (Tepper, 1998). However, it should be noted that so-called "non tasters" can taste PROP at high concentrations (Tepper, 1998).

Kalmus (1958) reported that sensitivity to the bitter taste of PTC is genetically linked to the dominant allele - 'T.' Non tasters of PTC being genotype – 'tt', and tasters being genotypes – 'Tt' and 'TT'. Some offspring of non taster parents have been found to be tasters (Olson, Boehnke, Neiswanger, Roche, and Siervogel, 1989). Sex and age have also been found to influence PTC sensitivity thresholds (Kalmus, 1958). Females were found to be more sensitive to PROP than males. Using threshold and supra-threshold tests, Bartoshuk, Fast, Karrer, Marino, Price and Reed (1992) demonstrated that there are three phenotypical groups and not two. The threshold is defined as the lowest concentration of a test solution that can be distinguished from plain water (Tepper, 1998). Tasters have very low thresholds ($< 1.0 \times 10^{-4}$ mol/l; high sensitivity to PROP at very low concentrations), whereas non tasters have higher thresholds ($> 2.0 \times 10^{-4}$ mol/l; poor sensitivity at low concentrations). Roughly one third of the taster population is homozygous (TT) tasters and is classified as super tasters and two thirds are heterozygous (Tt) tasters and classified as medium tasters. Bartoshuk *et al.* (1992) identified the three phenotypic groups using

supra-threshold taste scaling for NaCl (1.0×10^{-2} – 1.0 mol/l) and PROP (3.2×10^{-5} – 3.2×10^{-3} mol/l). The supra-threshold is a range above the threshold. Non tasters rated the intensity of PROP considerably lower than NaCl, super tasters rated the intensity of PROP considerably higher than NaCl, and the medium tasters' ratings for PROP and NaCl overlapped.

Sensitivity to PROP has been reported to be correlated with the density of both fungiform taste papillae and taste pores or buds (Miller and Reedy, 1990a; Miller and Reedy, 1990b; Bartoshuk, Duffy and Miller 1994; Duffy, Miller and Bartoshuk, 1994). In these studies, super tasters were found to have the highest densities followed by medium tasters followed by non tasters. This might explain the greater sensitivity of PROP tasters to basic tastes like bitterness and sweetness (Tepper, 1998).

1.2.4.3. Sensitivity to PROP and bitterness of other compounds

In addition to differing in the ability to perceive the bitterness of thioureas (PTC and PROP), tasters and non tasters have been reported to differ in their perception of the bitterness of other compounds (Mela, 1989). According to Drewnowski and Rock (1995) not all studies have found associations between the taste of PTC/PROP and other bitter compounds. Discrepancies have been found in studies investigating the relationship between PROP, urea, caffeine and quinine. Gent and Bartoshuk (1983) and Leach and Noble (1986) reported a significant positive relationship between PROP/PTC and quinine, whereas Kalmus (1958) and Mela (1989) did not find such a relationship. Gent and Bartoshuk (1983) reported that tasters found quinine hydrochloric acid (QHCl) significantly ($p < 0.02$) more bitter than non tasters. Leach and Noble (1986) compared the bitterness of PROP to quinine and caffeine using the time intensity (TI) sensory method. The tasters (n=8) rated the maximum intensity of quinine significantly ($p < 0.001$) higher than the non tasters (n=6). However, there was no significant difference between the two groups for their ratings of maximum intensity of caffeine. When comparing the intensity ratings for PTC and quinine by tasters and non tasters, Kalmus (1958) reported lower ratings (2.5) for intensity of PTC by non tasters than tasters (10.6), whereas the intensity ratings for quinine by the two groups (non tasters and tasters) were essentially similar (10.0 and 10.6, respectively). Mela (1989) assessed the perceived intensity of NaCl and five bitter compounds: caffeine, denatonium benzoate, QHCl, SOA and urea by tasters and non tasters. Mela (1989) reported that non tasters did not differ

significantly from the tasters in their bitterness ratings of both quinine and caffeine. However, significant group x concentration interactions were noted for urea, denatonium benzoate and SOA. Hall *et al.* (1975) studied thresholds of non tasters (n=10) and tasters (n=10) for PTC, caffeine, urea, QHCl and NaCl. A bimodal distribution was reported for caffeine at lower concentrations (albeit to a lesser degree) as noted for PTC. The caffeine thresholds were correlated with the PTC thresholds (Spearman rank correlation coefficient = 0.83, $p< 0.001$). Urea, like caffeine, was also perceived as slightly less bitter by non tasters than tasters at low concentrations; suggesting that although urea and caffeine do not possess a HNCS group they may stimulate the same receptor sites as PTC. Although QHCl also tastes bitter, it did not follow the same trend, and is seemingly coded by a different receptor site. In agreement with the findings of Kalmus (1958), Hall *et al.* (1975) reported that QHCl was equally bitter to tasters and non tasters. Contrary to the findings of Hall *et al.* (1975), but in agreement with the findings of Leach and Noble (1986), Mela (1989) reported that PROP tasters and non tasters did not differ in their ratings of the intensity of caffeine. While Hall *et al.* (1975) reported that urea was slightly less bitter to non tasters than tasters Mela (1989) did not find a relationship between PROP and urea. Delwich *et al.* (2001) reported significant differences between the ratings of the non tasters (n=4) and tasters (4 super tasters and 18 medium tasters) for the bitterness of QHCl, caffeine, (-) epicatechin, tetralone, L-phenylalanine, L-tryptophan, magnesium sulphate, urea, SOA and denatonium benzoate. However, such differences were not observed for the bitterness rankings of these compounds as a function of PROP taster status. It was concluded that a lack of significant difference in the compound rankings is an indication that the subjects in each group differ only quantitatively, not qualitatively. In other words, non tasters have lower system gains for bitterness due to their fewer taste buds and/or fewer taste pores compared to the medium and super tasters.

Frank and Korchmar (1985) studied the taster group's reaction time (RT) as well as the intensity ratings for different taste stimuli (sucrose, NaCl, QSO_4, HCl, PTC and water). The existence of two non taster sub-groups was reported. One group was insensitive to thiourea compounds only, whilst the other group was insensitive to thiourea and a number of the other compounds (sucrose, NaCl, QSO_4 and HCl). The second sub-group appeared to have a specific PTC sensitivity deficit that did not influence their processing of other taste stimuli. This finding is consistent with the view that insensitivity to PTC is a result of a lack of a PTC taste receptor (Frank and Korchmar, 1985). Delwich *et al.* (2001)

surmised that some of the discrepancies noted in the different studies (Kalmus, 1958; Hall *et al.*, 1975; Gent and Bartoshuk, 1983; Leach and Noble, 1986; Mela, 1989) on the bitterness of PROP, urea, quinine, caffeine and other bitter compounds may be due to the inclusion of differing proportions of the non taster sub-groups reported by Frank and Korchmar (1985). According to Delwich *et al.* (2001) studies with a high percentage of non tasters insensitive to PROP and other bitter compounds would be more likely to find a significant relationship between PROP and other compounds than study groups with a lower percentage this non tasters sub-group.

1.2.4.4. Sensitivity to PROP and phenolic compounds

Thorngate and Noble (1995) studied the time-course of bitterness and astringency of monomeric flavan-3-ols (-)-epicatechin and (+)-catechin in water, using the time intensity sensory method. Epicatechin was significantly more bitter and more astringent and had longer total duration than catechin. According to these authors, PROP status had no significant effect on any of the parameters: time to max (T_{max}), intensity at max (I_{max}) and total duration for both bitterness and astringency. Ishikawa and Noble (1995) investigated the interaction between astringency and sweetness of red wine using the time intensity methodology. As the level of tannic acid in the experimental wine was increased, all the astringency parameters increased. Maximum intensity and total duration of astringency were significantly reduced as the sucrose concentration increased. No differences were noted in the perception of astringency and sweetness between the PROP tasters (n=14) and non tasters (n=10). However, there was a significant difference in the intensity and persistence of astringency as a function of salivary flow rate. Low flow subjects rated the astringency higher and longer than high flow subjects. Smith, June and Noble (1996) examined the effects of viscosity and sweetness on astringency of aqueous solutions of grape seed tannin thickened with carboxymethyl cellulose or sweetened with aspartame. Maximum intensity and total duration of astringency decreased significantly as viscosity increased. Maximum intensity and total duration of bitterness were not significantly affected by increasing viscosity. Increasing sweetness had no effect on astringency parameters, but maximum intensity of bitterness was significantly decreased. PROP status and salivary flow rate had no effect on the perception of bitterness or astringency of the grape seed tannin aqueous solutions.

1.2.4.5. PROP sensitivity on acceptability of bitter foods

Greater sensitivity to the bitterness of PROP has been linked to reduced acceptability of bitter foods and beverages such as dry milk products and cheese (Marino, Bartoshuk, Monaco, Anliker, Reed and Desnoyers, 1991), brussels sprouts (Van Doorn, Van der Kruk, Van Holst, Raaijmakers-Ruijs, Postma, Groenweg and Jongen, 1998), broccoli and cheese (Tepper, 1999; Keller, Steinmann, Nurse and Tepper, 2002), broccoli, spinach, brussels sprouts, black coffee, soy milk and soybean tofu (Kaminski, Henderson and Drewnowski, 2000), grapefruit juice (Drewnowski, Henderson and Shore, 1997) and red wine (Pickering, Simunkova and DiBattista, 2003). Marino *et al.* (1991) investigated how tasters and non tasters would rate the sensory attributes (bitterness, sweetness, saltiness, sourness and creaminess) of a variety of cheeses. Cheddar and Swiss cheese were reported to taste more bitter to tasters than non tasters; and American and cottage cheeses were saltier to tasters than non tasters. On the other hand, sweetness, sourness and creaminess showed no taster/non taster association. Dry milk powders were also perceived to be more bitter to some tasters than non tasters. Casein was found to be more bitter to some adult tasters than non tasters. Since protein molecules are too large to stimulate taste, the bitter taste was attributed to fragments of proteins (amino acids) resulting from processing (Marino *et al.*, 1991). According to Tepper (1998), PROP and PTC are chemically related to the isothiocyanates and goitrin, which are bitter compounds found in cruciferous vegetables such as cabbage, broccoli, brussels sprouts, turnips and kale. Kaminski *et al.* (2000) studied food preferences of young women for brussels sprouts, broccoli, spinach, black coffee, soy milk and soybean tofu. PROP super tasters rated brussels sprouts as significantly more bitter than non tasters. The subjects who rated the foods as more bitter also rated them as less pleasant and less palatable. Bitterness was most frequently responsible for decreased food preference. Thus, food preferences were linked to taste preferences. Tepper (1999) investigated the influence of PROP taster status on the acceptance of broccoli, cheese and whole milk. PROP taster children gave significantly lower hedonic ratings for raw broccoli, cheese and whole milk than non taster children. Keller *et al.* (2002) determined the acceptance of bitter and fatty foods by taster and non taster children. Taster children showed a significantly lower acceptance of raw broccoli and American cheese; and taster girls showed a significantly lower acceptance of full-fat milk than non taster girls. According to Drewnowski *et al.* (1997) increased taste acuity for both PROP and naringin was associated with greater dislike for each bitter compound. Naringin is the primary bitter compound in grapefruit juice. PROP super tasters disliked bitter naringin solutions significantly more than non tasters. PROP sensitivity was also

associated with reduced acceptability of grapefruit juice. Drewnowski *et al.* (1997) reported that increased taste acuity for naringin, the primary bitter compound in grapefruit juice, and PROP were associated with greater dislike for each of the compounds. PROP super tasters disliked bitter naringin solutions significantly more than non tasters. PROP sensitivity was also associated with reduced acceptability of grapefruit juice.

1.2.5. Astringency

In addition to the taste and smell systems, there is a chemical and tactile responsiveness mediated by trigeminal nerves (Lawless and Heymann, 1998). According to these authors a variety of everyday flavour experiences arising from trigeminal stimulation including the fizzy tingle of carbonated drinks, burn of capsaicin in hot peppers, and the pungency of spices such as ginger and cumin. The trigeminal nerves also signal tactile, thermal, and pain sensations. Unlike bitterness which is mediated through taste receptors, astringency is an oral sensation signalled by trigeminal nerves (Vidal *et al.,* 2003). Astringency is an important sensory attribute of foods and beverages that contain tannins such as coffee, tea, beer, wine, apples, ciders and many berry crops and nuts (Lee and Lawless, 1991). The word astringent is derived from the Latin word *ad* (to) and *stringere* (bind). Thus, astringency is defined as a binding reaction relating to the ability of astringent materials to bind and precipitate proteins (Haslam and Lilley, 1988; Lee and Lawless, 1991). Saliva contains a considerable quantity of proteins (proline-rich proteins and possibly mucins) that lubricate the mouth (Siebert and Chassy, 2003). When these salivary proteins (especially those rich in proline) bind preferentially with polyphenols in foods, they form insoluble complexes (Gawel, Iland and Francis, 2001; Siebert and Chassy, 2003). Like sorghum kafirin (Taylor *et al.*, 1989), saliva contains proline-rich proteins (PRP) that interact strongly with tannins (Muenzer, Bildstein, Gleason, and Carlson, 1979; Hagerman and Butler, 1981). This results in a decrease in salivary lubrication properties and thus elicits the astringency sensation (Gawel *et al.*, 2001; Siebert and Chassy, 2003). According to Mehansho, Hagerman, Clements, Butler, Rogler and Carlson (1983) rats fed with a diet containing 2% CE tannin had an increased secretion of proline-rich salivary proteins. The increase in the proline-rich protein fraction was attributed to the tannins in the diet. This is seen as a protective mechanism for other dietary and digestive proteins not to interact with tannin (Hagerman and Butler, 1981). According to Mehansho, Clements, Sheares, Smith and Carlson (1985) proline-rich proteins are very efficient in

selectively binding to tannins and removing them from ingested food, hence reducing their detrimental effects. Also unlike bitterness, astringency is a tactile sensation because it has to do with feeling and not taste (Breslin, Gilmore, Beauchamp and Green, 1993). The tactile sensations caused by increased friction (decrease in salivary lubrication) between oral membranes are the primary basis of astringent sensations (Breslin *et al.*, 1993). Astringency belongs to mouth-feel sensations, particularly important in beverages such as fruit juices, tea and wine (Valentová *et al.*, 2002).

1.2.5.1. Compounds that cause astringent sensations

There are four main groups of compounds that cause astringency: plant polyphenols, salts of multivalent metallic cations (Al, Cr, Zn, Pb, Ca) particularly aluminium salts such as alum, mineral and organic acids, and dehydrating agents such as alcohol (ethanol) (Haslam and Lilley, 1988; Siebert and Chassy, 2003). Tannins make the mouth feel rough and dry because they cause a drawing, puckering or tightening sensation in the cheeks and muscles of the face as a result of coagulating saliva and mouth proteins (Haslam and Lilley, 1988; Lawless and Hyman, 1998). Polyphenols such as tannins form complexes with mucoproteins of the saliva and by either precipitating them or causing sufficient conformational changes so that they lose their lubricating power (Bate-Smith, 1973). According to Bate-Smith (1973), a threshold exists for the subjective experience of astringency to be sensed in the mouth. At low concentrations, not eliciting the 'puckery' sensation, the sensation is described as 'body' or 'substance' in wine or fruit. According to Asano, Shinagawa and Hashimoto (1982), the proline-rich haze forming proteins in beer have unfolded molecular structures that facilitate the entry of polyphenols into them. Peptides that contain proline were found to combine with polyphenols to form complexes that scatter light (indicating the presence of colloidal or larger size particles) in proportion to their proline content (Asano *et al.*, 1982; Siebert, 1999).

Organic and inorganic acids have also been reported to be astringent even though they do not resemble plant tannins (Rubico and McDaniel, 1992; Corrigan Thomas and Lawless, 1995). Organic acids (malic, citric and quinic acid) have been found to be both astringent and sour in model solutions (Rubico and McDaniel, 1992). Corrigan Thomas and Lawless (1995) compared astringency, astringent sub-qualities (drying, roughing and puckering), and sourness of organic and inorganic acids. The astringency profile of the organic acids (lactic, citric, acetic, fumaric and malic) was similar but slightly different from the

inorganic acids (HCl and phosphoric). HCl and phosphoric acids were more astringent and less sour while the organic acids were more sour than astringent. The accepted astringency mechanism of salivary proteins binding with tannins, involves hydroxyl groups on the tannin molecule binding to an electronegative site like the keto-imide linkage on the protein forming a complementary hydrogen bond pair (McManus, Davis, Lilley and Haslam, 1981). This mechanism might explain why some acids like tartaric acid, which has adjacent hydroxyl (–OH) groups, are potent astringents (Corrigan Thomas and Lawless, 1995). For the inorganic acids used here, a mechanism such as denaturation of salivary proteins may be responsible for the astringent sensation (Corrigan Thomas and Lawless, 1995). Salts like potassium aluminium sulphate (alum) and alcohols (ethanol) have dehydrating properties, and the resulting removal of water is presumably the cause of their astringency (Haslam and Lilly, 1988; Siebert and Chassy, 2003). Small multiple charged cations such as aluminium bind water very tightly (Siebert and Chassy, 2003). Peleg, Bodine and Noble (1998) proposed that alum interacts with salivary proteins or epithelial proteins to elicit the astringency sensation.

1.2.5.2. Sensory perception of astringency

Astringency, unlike true tastes, is a complex and persistent sensation that does not demonstrate adaptation (Lyman and Green 1990; Ishikawa and Noble, 1995). The intensity and duration of the astringency sensation has been found to increase with repeated ingestion (Guinard, Pangborn and Lewis, 1986a; Guinard *et al.*, 1986b; Lyman and Green 1990; Lee and Lawless 1991). Lyman and Green (1990) found that the astringent sensation can be altered by the presence of other compounds. Sweeteners in particular, reduced the astringency sensation. Sucrose was found to suppress the astringency of vermouths. The authors presumed it was probably due to an increase in the salivary flow rate. The salivary flow rate was measured as a function of pre-exposure to water, tannic acid, a mixture of tannic acid and sucrose (tannic acid+sucrose), and sucrose. Sucrose and a mixture of tannic acid+sucrose increased the salivary flow rate significantly more than water and tannic acid. However, although salivary flow rate was highest when subjects sipped sucrose alone, there was no significant difference between the salivary flow rates of sucrose and tannic acid+sucrose. Different theories were proposed as to why sucrose decreased the astringency sensation. It was postulated that by sucrose stimulating more saliva production, additional proteins in the saliva reversed the phenol/protein complexes causing the astringent sensation in the mouth as proposed by Haslam and Lilley

(1988). Alternatively, it was proposed that the increased salivation simply helped clear out the phenols from the mouth, as has been proposed by Lagerlof and Dawes (1985), and/or provided new proteins to replace the precipitated proteins as has been proposed by Joslyn and Goldstein (1964). Another theory was that the viscosity of the sucrose solution provided lubrication that helped mask the astringency sensation. Peleg and Noble (1999) reported that increasing the viscosity of cranberry juice using carboxymethylcellulose lowered its perceived astringency. Smith *et al.* (1996) also reported that increased viscosity caused by carboxymethylcellulose lowered the perceived astringency of aqueous solutions of grape seed tannin.

Astringency of phenolic compounds has been reported to increase in the presence of added acid (to lower pH) (Fischer, Boulton and Noble, 1994; Peleg *et al.*, 1998). Astringency of aqueous solutions of phenolic compounds (grape seed tannin, tannic acid, catechin and gallic acid) increased upon addition of citric acid, whereas the astringency of alum was reduced (Peleg *et al.*, 1998). The difference noted between the phenolic compounds and alum was attributed to the chemical modifications affecting the binding capacity of the different astringents to salivary proteins. Chelation of the aluminium ion in alum by acids reduced its availability to bind the salivary proteins. On the other hand, the increased astringency noted for the phenolic compounds upon acidification was speculated to result from the pH driven increase in affinity of the phenols for binding with proteins. Kallithraka, Bakker and Clifford (1997a) assessed how addition of malic acid and lactic acid affected the bitterness, astringency and sourness of red wines and model solutions. The intensity and duration of astringency and sourness increased with decreasing pH in both the model solutions and red wine. Bitterness was not affected by the addition of either acid. Peleg and Noble (1999) also reported that the astringency of cranberry juice could be modified by altering the pH.

Kielhorn and Thorngate (1999) used a multidimensional scaling (MDS) study of ten diverse compounds: (+)-catechin, (-)-epicatechin, caffeine (bitter), citric acid (acid), alum, tannic acid, grape seed tannin, gallic acid, ethanol and capsaicin. Three recognizable groupings emerged: a bitter neighbourhood comprising of (+)-catechin, (-)-epicatechin, caffeine and ethanol; an acid neighbourhood comprised of citric acid and gallic acid; and an astringent neighbourhood comprised of tannic acid and grape seed tannin. Thus, although (+)-catechin and (-)-epicatechin are described as astringent, they were more

closely associated with caffeine and ethanol than the traditional astringents (tannic acid and grape seed tannin) in the MDS plot. Dimension 2 was defined by capsaicin, indicating that it was unique to the perceptual space. According to Kielhorn and Thorngate (1999) as aspartame and sucrose share 'sweetness' even though the quality of the sweetness is different, so may the monomeric flavan-3-ols and their polymeric counterparts share 'astringency' although the true quality of the sensation is different. Although small chemically, benzoic acid derivatives: salicylic acid (2-hydroxy benzoic acid), *m*-hydroxyl benzoic acid (3-hydroxy benzoic acid), gentisic acid (2,5-hydroxyl benzoic acid) protocatechuic acid (3,4-hydroxy benzoic acid) and gallic acid (3,4,5-trihydroxy benzoic acid) in water, are also reported to elicit astringency (Peleg and Noble, 1995). McManus *et al.* (1981) determined the association of small phenols: resorcinol (1,3-dihydroxybenzene), catechol, and pyrogallol with bovine serum albumin (BSA) (Fig. 1.10). The astringency of these small phenols was attributed to precipitation of or strong binding with proteins due to the presence of 1,2-dihydroxy or 1,2,3-trihydroxy groups. The affinity of resorcinol for BSA was weaker than that of catechol and pyrogallol, which have two and three *ortho*-disposed phenolic groups respectively, to more strongly bind the protein.

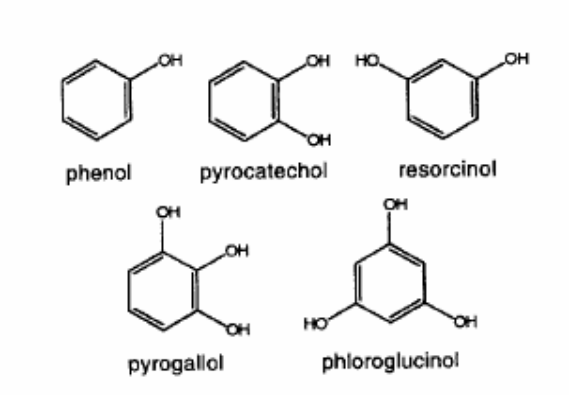


Figure 1.10. Basic structures of small phenolic compounds (Kennedy, Saucier and Glories, 2006).

One difficulty in studying astringency is that many untrained observers confuse astringency with bitterness (Lee and Lawless, 1991). Lea and Arnold (1978) classified bitterness and mouth drying as 'twin sensations' because nearly all astringents are also bitter, and untrained panellists sometimes confuse the two qualities. In addition to being

bitter, many astringent materials (particularly organic acids) also have a sour side-taste. Lee and Lawless (1991) examined quantitative and qualitative perceptual reactions to astringent materials for three diverse chemical substances (alum, tannic acid and tartaric acid) at several concentrations producing moderate to strong levels of perceived sensation. A trained sensory panel developed six descriptors for the sensory sensations elicited by the astringent substances as follows: astringency, mouth-drying, puckery feeling, mouth-roughing, bitterness and sourness. The time intensity ratings for each attribute were found to depend on both the particular astringent substance and concentration tested. These authors recommended the use of alum as a standard in future structure activity studies using time intensity procedures because it was relatively low in perceived bitterness and sourness, but produced pronounced drying, roughing, puckering/drawing sensations.

1.2.5.3. Acceptability of astringency in food

Several predominantly astringent and bitter beverages such as tea, wine, beer and coffee are widely consumed (Guinard *et al.*, 1986a; Mattes, 1994; Drewnowsky and Gomez-Carneros, 2000; François, Guyot-Declerck, Hug, Callemien, Govaerts and Collin, 2006). Astringency is an essential characteristic of wine caused by procyanidins, affecting perceived 'mouth-feel' that is informally described as 'soft', 'hard' or 'rough' especially when referring to red wine (Guinard *et al.*, 1986a; Ishikawa and Noble, 1995). Pickering *et al.* (2003) examined the relationship between taste and astringency perception elicited by red wines and sensitivity to PROP. Bitterness, astringency and acidity intensities were all correlated with PROP taster status. PROP non tasters gave significantly lower intensity ratings for astringency, bitterness and acidity of the red wines than did PROP tasters. However, Ishikawa and Noble (1995) investigating the astringency and sweetness of Carnelian red wine using the time intensity methodology and found no relationship between PROP taster status and the astringency perception of the wine. According to these authors the magnitude of astringency and sweetness of wine did not differ between PROP tasters (n=14) and non tasters (n=10).

1.2.6. Time intensity sensory evaluation procedure

The time intensity sensory evaluation method is used to continuously capture, in great detail, nuances of flavour growth, decay and disappearance (Lawless and Heymann, 1998). Over the years, the time intensity evaluation method has been used in tracking the changes

in the sensory properties (flavour and texture) of different foods and beverages. Foods possess a composite of many taste attributes, but in most cases the evaluation of these attributes is typically made for each individual attribute (Duizer, Bloom and Findlay, 1997). Furthermore, many sensory evaluation methods, such as quantitative descriptive analysis and difference tests, measure the perception of food flavour and texture as static events even though the intensity perception does not occur at a single point in time (Bloom, Duizer and Findlay, 1994; Dijksterhuis and Piggott, 2001). Processes involved in eating, such as mastication and salivation are dynamic (Dijksterhuis and Piggott, 2001); both flavour and texture intensities change as the food moves through the mouth and is prepared for swallowing (Bloom *et al.*, 1994). Therefore methods taking these dynamic processes into consideration are likely to produce more valid results than static methods (Dijksterhuis and Piggott, 2001). This is why the time intensity method is gaining wide application because it measures changes in the perception of product attributes over time (Bloom *et al.*, 1994).

1.2.6.1. Single attribute time intensity (SATI) and dual attribute time intensity (DATI) sensory methods

According to Leach (1984), bitterness and astringency are characterized by a persistent after-taste and thus cannot be estimated solely by scalar intensity procedures. Scalar or point estimates of intensity are inadequate when the sensory properties of samples vary differentially over time (Noble, 1995). For instance, wines that may be equally bitter when first sipped, may vary in the persistence of bitterness after the wine is swallowed. Thus, to fully characterize the differences in their sensory properties requires analysis of the time-course of perceived intensity. Time intensity sensory evaluation has mostly been used to measure single attributes.

The time intensity method has been used in the study of the bitterness and/or astringency of wine (Boulton and Noble, 1994; Valentová *et al.*, 1997; Kallithraka, Clifford and Bakker, 1997b), beer (François *et al.*, 2006; King and Duineveld, 1999) and soymilk (Courregelongue, Schlich and Noble, 1999). Zimoch and Findlay (1998) used the time intensity sensory method to study the juiciness and toughness of beef samples. McGowan and Lee (2006) also used it in a study of artificial sweeteners in chewing gums. Most of these studies used the single attribute time intensity (SATI) method. However, in recent years time intensity is gaining popularity measuring dual attributes simultaneously.

Duizer *et al.* (1997) compared the SATI method to the dual attribute time intensity (DATI) method in investigating dual attributes of sweetness and peppermint flavour of four samples of chewing gum with varying rates of sweetness and peppermint flavour release. They observed that in general, the DATI method was as sensitive as the SATI method in distinguishing sweetness and peppermint flavour of the chewing gum.

According to Duizer *et al.* (1997), advantages inherent with using the DATI method include collection of sensory data that more accurately reflects what is taking place in the mouth during consumption of a food; using the DATI method means that only half the time was required to collect the same information by the SATI method since both attributes were measured simultaneously; the DATI method can also possibly provide solutions to two known methodological problems: dumping and inter-sample variability. Dumping is a problem that occurs in single attribute measurements when a single attribute is rated more intense when evaluated alone than when evaluated with other attributes (Duizer *et al.*, 1997; Zimoch and Findlay, 1998).

1.2.6.2.‘Panellist’s signature’

According to Boulton and Noble (1994), the human judge is a multi-purpose instrument who can be trained to measure many attributes. However, despite extensive training of judges to calibrate their use of descriptive terms and rating scales, individual physiological and psychological differences affect perception of sensory properties. Time intensity studies are subject to different biases, one of which is panellist variation (Valentová *et al.*, 2002). Valentová *et al.* (2002) paid particular attention to the variability of different judge’s responses. They reported that there were slow, medium and rapidly reacting subjects. Differences in the salivary flow rates of judges have been attributed to noted differences among judges (Boulton and Noble, 1994; Fischer *et al.,* 1994; Ishikawa and Noble, 1995; Kallithraka, Bakker, Clifford and Vallis, 2001). Consequently, individual curve shapes showed a high variance among judges (Pangborn, Lewis and Yamashita, 1983).

Fischer *et al.* (1994) studied the physiological factors contributing to the variability of sensory assessments, i.e. the relationship between salivary flow rate and temporal perception of gustatory stimuli using wines varying in ethanol, pH and phenolic composition. They reported that the perceived intensity and duration of bitterness and

astringency were affected by salivary flow rate, possibly due to salivary volume, salivary pH and protein composition. Subjects with low saliva flow rates took longer to reach maximum intensity (T_{max}) and had a longer duration (D_{tot}; persistence) of bitterness and astringency than subjects with high flow rates. Low flow subjects also perceived the intensity (I_{max}) of bitterness and astringency higher than subjects with high flow rates. Ishikawa and Noble (1995) studied the temporal perception of astringency and sweetness in red wine using the time intensity methodology. They found a significant difference for both intensity (I_{max}) and duration (D_{tot}) of astringency of red wine between the low and high saliva flow subjects. The low-flow subjects rated astringency higher and longer than the high-flow subjects.

1.3. Conclusions

A lot of research has been done to determine the sensory properties of phenolic compounds in fruits, tea, wine, beer and other foods but information on the sensory properties of phenolic compounds in sorghum is limited. Condensed tannins are potentially important antioxidants, but consumption of tannin-containing (tannin) sorghums is hampered by the general belief that tannins confer objectionable sensory attributes to this food. Therefore it is necessary to determine the sensory attributes of sorghums containing varying amounts of phenolic compounds, especially condensed tannins and to determine their acceptability to consumers. Bitter taste perception has been genetically linked to sensitivity to PROP, in that some people can taste its bitterness (tasters) whilst others cannot (non tasters). Since condensed tannins are bitter and astringent, preference ratings of sorghum sensory attributes may be influenced by genetic sensitivity to PROP.

1.4. Hypotheses

Tannin sorghums will be more bitter and more astringent than tannin-free sorghums because the total phenol content (including tannins) of these sorghums exceeds that of the tannin-free sorghums. It has been found that phenolic compounds in sorghum contribute significantly to the perceived bitterness and astringency of sorghum products (Asante, 1995; Yetneberk *et al.*, 2004).

High molecular weight phenolic compounds are known to be predominantly astringent; while the low molecular weight compounds are known to be predominantly bitter (Leach, 1984; Peleg *et al.*, 1999). Therefore, astringency will predominate in tannin sorghums, while bitterness will predominate over astringency in the tannin-free sorghums.

Condensed tannins in foods are well-known for eliciting negative consumer response at high intensity because of their dominant sensory attributes: bitterness and astringency (Cheynier, 2005; Lesschaeve and Noble, 2005). Therefore the bitterness and astringency of tannin sorghums will be more intense than tannin-free sorghums and as a result these sorghums and will be less acceptable to consumers.

Bitter taste perception has been genetically linked to sensitivity to 6-*n*-propylthiouracil (PROP), in that some people can taste its bitterness (tasters) whilst others cannot (non tasters; Bartoshuk, 1993). Since condensed tannins are bitter and astringent, acceptance ratings of these sorghums will be influenced by genetic sensitivity to PROP. Therefore, the bitterness of the tannin sorghums will have a negative influence on the acceptability of these sorghums to PROP tasters, while non tasters will find these sorghums equally acceptable.

1.5. Objectives

The objectives of the study were:

1. To determine the bitterness, astringency and other sensory attributes of bran infusions and sorghum rice of sorghums containing different levels of phenolic compounds using a trained sensory panel.

2. To determine the intensity and time course of bitterness and astringency of bran infusions of sorghums varying in condensed tannin content using the time intensity sensory method for dual attributes.

3. To determine which sensory attribute (bitterness or astringency) predominates in the bran infusions from tannin and tannin-free sorghums.

4. To determine consumer acceptability of the rice of sorghums containing different levels of condensed tannins.

5. To determine whether PROP taster status influences the acceptability of the rice of sorghums containing different levels of condensed tannins.