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EVALUATION OF ASTRINGENCY OF PREPARATIONS WITH DIFFERENT DEGREE OF TANNIN POLYMERISATION

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Sensory and chemical methods were used for the evaluation of astringency of preparations differing in the degree of tannin polymerisation: (1) preparation of green tea (monomers), (2) preparation of grape seeds (oligomers), and (3) tannin polymers from black chokeberry fruits. The sensory and chemical astringency of the preparations was found to be diversified and occurred in the following order: tannin polymers > preparation of green tea > preparation of grape seeds. Of six modified starches under study, only two were able to negligibly neutralise the astringency sensations of tannins.

INTRODUCTION

Condensed tannins (*i.e.* proanthocyanidins) have been acknowledged as a health-promoting component of a diet. Tannin preparations obtained from grape seeds and green tea have been used as a dietary supplement of foodstuffs and as para-medicines. Tannins are composed of units of flavan-3-ol linked in the C_4-C_8 or C_4-C_6 position. Consequently, they have different degree of polymerisation, from simple molecules (monomers, dimers) to oligomers then polymers (Figure 1). The biological activity of tannins is determined by their structure, and mainly by the degree of their polymerisation. Catechins (monomers) present in green tea have been perceived as anticarcinogenic and antimutagenic compounds [Chung *et al.*, 1998; Katiyar & Mukhtar, 1996], whereas proanthocyanidins (oligomers) from grape seeds have been reported to exert a hypocholesterolemic effect in cholesterol-fed rats [Tebib *et al.*, 1994a, b, 1997].

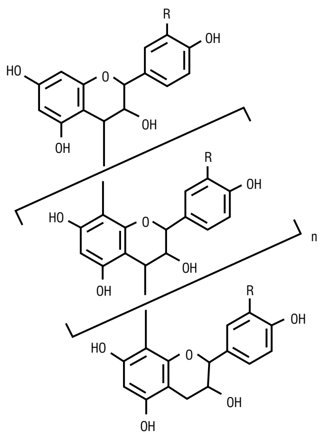

FIGURE 1. Chemical structure of condensed tanins.

Apart from valuable biological activity, tannins introduce into food some negative sensory attributes, namely bitterness and astringency. The sensory activity of tannins is determined also by the degree of their polymerisation. Researches have shown that larger molecules tend to be less bitter and more astringent [Lea & Arnold, 1978; Arnold *et al.*, 1980].

The objective of this study was to compare astringency of preparations differing in the degree of tannin polymerisation: (1) preparation of green tea (monomers), (2) preparation of grape seeds (oligomers), and (3) tannin polymers from black chokeberry fruit. Additional analyses were performed to determine the ability of soluble modified starches to neutralise astringency sensation.

MATERIALS AND METHODS

Material. Preparations: catechin - green tea leaves extract (*Cameiiia sinensis*) and proanthocyanidin - pure grape seed extract (*Vitis vinifera*) were purchased from General Nutrition Centre (GNC) in USA. Preparation of tannin polymers was obtained from black chokeberry fruit (*Aronia melanocarpa Elliot*). In brief, homogenised fruits were extracted with 70% aqueous ethanol for 30 min at the ratio of (1:7 w/v) in a shaking incubator. Supernatant containing low-molecular phenolic compounds was removed by centrifugation. Solid containing polymerised tannins was extracted twice more with 70% aqueous acetone. The resultant extract was evaporated under vacuum at 40°C and lyophilised.

Sensory panel and testing conditions. A total of 9 subjects, 6 female and 3 male students (from the University of Warmia and Mazury in Olsztyn), and staff from the Institute of Animal Reproduction and Food

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Research of the Polish Academy of Sciences, Olsztyn, participated in nine sessions of a 2-week training. One formal session was preceded by training sessions to familiarise the panelists with the concept of astringency and the computerised linear scaling system. For training sessions, reference standards for astringency were 0 and 1.5 g/L tannic acid. Both training and formal sessions were made in sensory laboratory room, fulfilling general requirements for sensory testing conditions [ISO 8589:1998].

Methods. The duo-trio test was applied for determination of the detection threshold of tannic acid. The number of correct judgements in the test was compared with the statistical values presented by Larmond [1977]. On the basis of these data, the detection threshold was determined as the concentration at which 75% of the judges distinguished the sample containing tannic acid.

Differential profile of the preparations was obtained with the method of multiple comparisons [Baryłko-Pikielna, 1975]. Concentration standard (tannic acid) accounted for 1.0 g/L, and that of each of the preparations - for 1.4 g/L of deionised water.

Sensation of Astringency Indices were calculated according to the method described by Matuszewska *et al.* [2000]. The intensity of astringency was measured on linear scale anchored from “none” to “very strong” [ISO 4121:1987]. The results were then converted into numerical values (10 units). On the basis of experimental curves, SAI were developed as a ration of half scale astringency (5 units) to evoking its preparations concentration.

“Chemical” astringency of the preparations measured by the amount of tannins complexed by bovine serum albumin (BSA) was assayed colorimetrically according to the method of Makkar *et al.* [1988]. Results were given in absorbance values (A_{510}) per gram of preparation.

Additional analyses were performed to determine the ability of soluble modified starches: tapioca, potato, wheat, and maize to neutralise astringency sensation. The panelists evoked the astringency sensation by taking into their mouth 15 mL of aqueous solution of polymer preparation (1.4 g/L). Then, they neutralized that sensation rinsing their mouth with 1% (v/w) solutions of modified starches. Distilled water was used as a reference solution.

RESULTS AND DISCUSSION

In a sensory analysis of phenolic compounds, tannic acid is most often used as a referential standard. The results of the duo-trio tests used in determination of the detection threshold values for that compound are presented in Table 1. The detection threshold value for tannic acid was low and reached 0.14×10^{-4} mol/L. It should be emphasised that some panelists reacted to lower concentrations, which indicates that their individual thresholds were below the accepted mean threshold. The results obtained proved that phenolic compounds present in food may limit its consumption to a considerable extent.

In order to compare the astringency of the preparations tested with the standard, differential profile of the samples was estimated. It was found out that the intensity of the astringency of the samples was very different. Monomers were shown to be characterised with astringency similar to that of the standard, while polymers demonstrated considerably higher intensity of that attribute (3.6 arbitrary units higher than the standard). Oligomers were characterised by the lowest astringency (4 arbitrary units lower than the standard) (Figure 2).

TABLE 1. Detection threshold of tannic acid.

No.	Concentration of tannic acid	
	% x 10^{-2}	mol/L x 10^{-4}
1	0.20	0.12
2	0.22	0.13
3	0.24*	0.14
4	0.26	0.15
5	0.28	0.16
6	0.30	0.17

* Threshold value; *Deionised water served as reference.

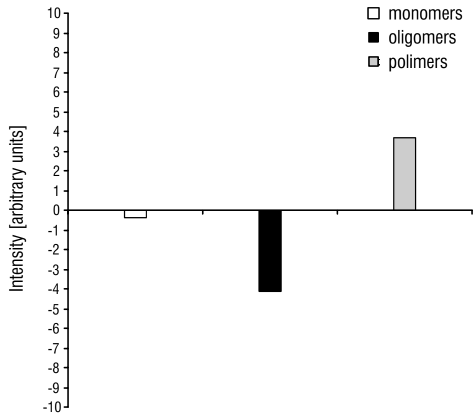

FIGURE 2. Differential profile of preparations with various degree of tannin polymerisation (standard – tannic acid 1.0 g/L).

In order to compare the strength of the astringency of the preparations, their Sensation of Astringency Indices (SAI) were calculated from concentration/intensity curves for each sample. It was found out that SAI reached 56.81, 10.00, and 65.78 for monomers, oligomers, and polymers, respectively (Table 2). The same order was reported for the absorbance values (A_{510}), characterising the “chemical” astringency of the preparations. The lowest values were reported for oligomers (5.24) and the highest ones for polymers (178.8) (Table 2). The results obtained suggest a correlation between two methods used in the study. According to literature data, catechins do not precipitate protein, therefore the “chemical” astringency of green tea preparation may be determined by phenolic compounds other than catechins [Hagerman *et al.,* 1998].

Sensory evaluation of the preparations faced a methodological problem of neutralising a long-standing astringency intensity in the mouth. Very little is known about the mechanism by which the drying sensation of

TABLE 2. Sensory (SAI) and chemical astringency (A_{510}/g) of preparation differing in the degree of tannin polymerisation.

Preparation	A_{510}/g	Sensation of Astringency Index (SAI)	Preparation concentration [%] by which the astringency intensity is equal to 5 units
Monomers	69.55 ± 0.56	56.81	0.088
Oligomers	5.24 ± 0.44	10.00	0.500
Polymers	178.8 ± 1.25	65.78	0.076

astringency is perceived. There is a view that astringency is caused by the precipitation of salivary proteins resulting in diminished oral lubrication [Hagerman & Butler, 1981]. According to that hypothesis, lubricating rinses (raised with hydrocolloids) should decrease the astringency of tannins.

TABLE 3. The usability of soluble modified starches to neutralise astringency sensation.

Modified starches	Usability for neutralisation
Hydrolysed starches from:	
tapioca	-
potato	-
wheat	-
maize	-
Pudding potato starch	+
Gelling potato starch	+

" - " lack of impact; " + " insignificant impact

Preliminary investigations of the ability of soluble modified starches to neutralise astringency sensation (Table 3) demonstrated that hydrolysed starches of tapioca, potato, wheat, and maize, did not reduce the long-standing sensation of astringency in the mouth of the panelists. Two potato starches only: pudding and gelling, reduced that sensation to a small extent. The usability of those starches for the sensory analysis of the preparations was troublesome due to their incomplete solubility in water at ambient temperature. In consequence, the evaluation panelists used mineral water and non-salted crackers to neutralise the astringency sensation.

CONCLUSIONS

In conclusion, we found out that both sensory and chemical astringency of the preparations, differing in the degree of tannin polymerisation, were ordered as follows: tannin polymers > preparation of green tea > preparation of grape seeds. More intense astringency of preparation from green tea than that of preparation from grape seeds suggests that not only tannins but also other phenolic compounds or/and non-phenolic compounds may be responsible for that sensory attribute. These investigations point to the need of continuing studies into the neutralisation of astringency sensation as it is an important methodological issue in the analysis of phenolic compounds.

REFERENCES

1. Arnold R.A., Noble A.C., Singleton V.L., Bitterness and astringency of phenolic fractions in wine. J. Agric. Food Chem., 1980, 28, 675–678.
2. Baryłko-Pikielna N., 1975, Zarys Sensorycznej Analizy Żywności. WNT, Warszawa.
3. Chung K.T., Wong T.Y., Wei C.I., Huang Y.W., Lin Y., Tannins and human health: a review. Crit. Rev. Food Sci. Nutr., 1998, 38, 421–64.
4. Hagerman A.E., Rice M.E., Ritchard N.T., Mechanisms of protein precipitation for two tannins, pentagalloyl glucose and epicatechin$_{16}$ (4–8) catechin (procyanidin). J. Agric. Food Chem., 1998, 46, 2590–2595.
5. Hagerman A.E., Butler L.G., The specificity of proanthocyanidin-protein interactions. J. Biol. Chem., 1981, 256, 4494–4497.
6. ISO 8589:1998, Sensory analysis – General guidance for the design of test rooms.
7. ISO 4121:1987, Sensory analysis – Methodology – Evaluation of food products by methods using scales.
8. Katiyar S.K., Mukhtar H., Tea in chemoprevention of cancer: epidemiologic and experimental studies. Int. J. Oncol., 1996, 8, 221–38.
9. Larmond E., Laboratory methods for sensory evaluation of food. Canadian Goverment Publishing Centre, 1977.
10. Lea A.G.H., Arnold G.M., The phenolics of ciders: bitterness and astringency. J. Sci. Food Agric., 1978, 29, 478–483.
11. Makkar H.P.S., Dawra R.K., Singh B., Determination of both tannin and protein in a tannin-protein complex. J. Agric. Food Chem., 1988, 36, 523.
12. Matuszewska I., Baryłko-Pikielna N., Szczecińska A., Skąpska S., Sensory characteristics of industrially manufactured apple aroma condensates, 2000, *In*: Proceedings of the 6th Wartburg Aroma Symposium, Eisenach, 10–13 April 2000, pp. 380–381.
13. Tebib K., Bitri L., Besancon P., Rouanet J.M., Polymeric grape seed tannins prevent plasma cholesterol changes in high cholesterol-fed rats. Food Chem., 1994a, 49, 403–406.
14. Tebib K., Besancon P., Rouanet J.M., Dietary grape seed tannins affect lipoproteins, lipoprotein lipases and tissue lipids in rats fed hypercholesterolemic diets. J. Nutr., 1994b, 124, 2451–2457.
15. Tebib K., Rouanet J.M., Besancon P., Antioxidant effects of dietary polymeric grape seed tannins in tissue of rats fed a high cholesterol-vitamin E-deficient diet. Food Chem., 1997, 59, 135–141.