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SHORT COMMUNICATION

POLYPHENOLS OF IMMATURE SAPOTA FRUIT

A. G. MATHEW and S. LAKSHMINARAYANA

Central Food Technological Research Institute, Mysore-2, India

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Abstract—The polyphenols of sapota fruit at a tender stage have been separated by paper chromatography. The simple phenols identified are *cis*- and *trans*- forms of chlorogenic acid, gallic acid, catechins and leucocyanidin. The polymeric proanthocyanidins contain leucodelphinidin, leucocyanidin and leucopelargonidin and traces of catechin.

INTRODUCTION

THE CHEMISTRY of sapota *Achras sapota* L. (Sapotaceae) (Sapodilla), which is an important crop in India, has not been studied in detail until recently.[1] Because of the high polyphenol content, the fruit is astringent, especially when immature. The astringency gradually decreases as the fruit attains maturity and the fruit becomes sweet when fully ripe. Although the anthocyanidins that are formed on treatment of the total polyphenols with hot acid have been identified as mainly delphinidin and cyanidin and to a lesser extent pelargonidin,[2] a better knowledge of the exact nature of the individual polyphenols has been found necessary to understand the astringency in this fruit. In view of the high degree of astringency and larger concentration of polyphenols noted at the immature stage, fruit at this low maturity was chosen for detailed study.

RESULTS

The chromatographic and chemical characteristics of the sapota polyphenols, after two-dimensional separation, are presented in Table 1. The spot designated 16 (Table 1) was present in traces and could only be detected in a concentrated ether extract. Chromatograms obtained from spotting the pressed juice also gave essentially the same pattern, suggesting that the extraction was efficient and that little or no change occurred during the short-term processing.

EXPERIMENTAL

Fruit Sample

Fruits of 2 months' maturity from fruit-set were collected from the Institute orchard. At the 2-month stage, the weight of the fruit was 0·28 g with a moisture content of 66·4 per cent. Estimation by Folin Denis method of freeze-dried sapota fruit extract using areca mixed polyphenols as standard[3] showed 19·63 per cent of polyphenols on dry basis. A hot-water extract of the immature fruits was freeze-dried and used for different studies as described in an earlier communication.[2]

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[2] S. LAKSHMINARAYANA and A. G. MATHEW, *J. Food Sci.* **32**, 451 (1967).
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TABLE 1. R_f VALUES, PROPERTIES AND IDENTITY* OF SAPOTA POLYPHENOLS

Spot No.	Intensity in $K_3Fe(CN)_6$ + $FeCl_3$ spraying	R_f in BAW	R_f in 2% acetic acid	Bis-diazotized benzidine test	Vanillin-HCl test	Leucoanthocyanidin (*n*-butanol–HCl) test	Probable identity	% of total (Folin-Denis)
1	2	3	4	5	6	7	8	9
1	Very intense	0·68	0·74	Yellow–brown	−ve	−ve	*trans*-Chlorogenic acid	6·2
2	Very intense	0·66	0·58	Yellow–brown	−ve	−ve	*cis*-Chlorogenic acid	5·1
3	Intense	0·63	0·40	Yellow–brown	Very faint	Very faint	Gallic acid and (+) catechin	2·3
4	Faint	0·60	0·10	Faint brown	−ve	−ve	Flavanol	1·6
5	Very faint	0·52	0·38	Faint brown	+ve	−ve	(−)-Epicatechin	1·2
6	Faint (elongated)	0·44	0·42 to· 0·52	Faint brown	+ve	+ve	Leucocyanidin	3·5
7	Very faint	0·42	0·76	Faint brown	−ve	−ve	Non-flavan-phenol (unidentified)	1·1
8	Fairly intense	0·28	0·79	Brown	+ve	−ve	Flavan (unidentified)	1·5
9	Fairly intense	0·32	0·72	Brown	+ve	−ve	Flavan (unidentified)	1·3
10	Intense	0·22	0·72	Brown	+ve	Very faint	Flavan (unidentified)	1·9
11	Intense	0·22	0·63	Brown	+ve	Very faint	Flavan (unidentified)	2·0
12	Intense	0·11	0·65	Brown	+ve	Faint	Flavan (probably a proanthocyanidin)	3·0
13	Faint	0·1 to 0·4	0·0 to 0·55	Brown	+ve	+ve	Proanthocyanidin complex†	16·2
14	Very intense	0·0	0·0 to 0·6	Brown	+ve	+ve	Proanthocyanidin complex†	26·3
15	Very intense	0·0	0·0	Brown	+ve	+ve	Proanthocyanidin complex†	27·2
16	Very faint	0·5	0·0	Brown	−ve	−ve	Flavanol	Negligible

* Each of these values given in the Table represents an average of three values.

† Compound 13 is composed mainly of leucocyanidin with traces of leucopelargonidin, compound 14 of leucodelphinidin as well and compound 15 of mainly leucodelphinidin with lesser amounts of leucopelargonidin.

Chromatographic Procedure

Two-dimensional chromatograms on Whatman No. 3 paper were developed with *n*-butanol–acetic acid–water (BAW) 4:1:2·2 v/v in the first direction and 2% aqueous acetic acid in the second direction. They were sprayed with specific spray reagents.[4] The compounds were identified (Table 1) by standard procedures. Quantitative separation and estimation was carried out by using four identical papers. Corresponding spots were marked from one of the papers already sprayed with $K_3Fe(CN)_6 + FeCl_3$ and then cut into small pieces for estimation in triplicate.[5] The leucoanthocyanidin test was done by the method described by Swain and Hillis.[6]

Spots 1 and 2 in alcoholic solution showed the u.v. spectral characteristics of chlorogenic acid. Chlorogenic acids, gallic acid and catechins[3] were confirmed by co-chromatography with authentic samples.

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