Chemistry of Natural Compounds, Vol. 34, No. 2, 1998

CHEMICAL CHARACTERISTICS OF THE SEPTA OF THE FRUIT OF *Juglans regia*

M. G. Bezhuashvili and N. Z. Kurashvili

The qualitative composition of the lipids, the triacylglyceride content, and the fatty acid composition of the septa of the fruit of Persian walnut Juglans regia *have been determined. The phenol fraction is represented in the form of phenolic carboxylic acids, phenolic aldehydes, catechins, proanthocyanidins, and oligomeric and polymeric phenolic compounds, including lignin. Polysaccharides are represented mainly in the form of xylan and glucan.*

The species Persian walnut *Juglans regia* is widely distributed in Georgia. The septa of its fruit form a renewable raw material. A study of the chemical composition of this raw material is necessary for the rational use of the products obtained from it. With this aim, we have investigated the chemical composition of the septa of *Juglans regia* fruit.

The results of an experiment (Table 1) show that the fraction soluble in diethyl ether was represented mainly by lipid compounds, together with low-molecular-mass phenolic substances. The lipids of the material under investigation included triacylglycerides, sterol esters, fatty acid methyl esters, and hydrocarbons. Among these components the triacylglycerides predominated. The following fatty acids participated in their formation: palmitic, stearic, oleic, linoleic, and linolenic.

The phenolic fraction of the septa of Persian walnuts was represented in the form of low-molecular-mass phenolic aldehydes and phenolic carboxylic acids, catechins, proanthocyanidins, and oligomeric and polymeric phenolic compounds, including lignin. The raw material investigated was rich in low-molecular-mass aromatic substances including, in particular, gallic, protocatechuic, *p*-coumaric, vanillic, syringic, 4-hydroxybenzoic, and ferulic acids; they also contained protocatechualdehyde, vanillyl alcohol, syringyl alcohol, and 4-hydroxybenzaldehyde. Catechins were represented in the form of (+)-catechin and (−)-epicatechin.

Transformations of the Persian walnut septa in an acid medium formed petunidin, which showed the presence of the corresponding proanthocyanidin in the raw material. The high content of polyphenols must be mentioned, although their accurate quantitative determination could not be achieved by titration with $KMnO_4$ solution. We assume that this was due to the low content of free hydroxy groups, causing their partial oxidation or, perhaps, to their participation in polymerization. To some degree, the polyphenols were dissolved by hot extraction of the raw material with ethyl alcohol and water. After hot-water extraction with the production of a dark brown extract (yield 14%), the raw material gave a faintly colored alcoholic extract with a low content of extractive substances.

The unextracted fraction of the Persian walnut septa consisted mainly of a complex of lignin with polysaccharides. It showed a high content of lignin — 39% a.d.m. — which is greater than in woody plants. This contradiction is explained by the inhomogeneity of the lignin. During the performance of the experiment, part of the polyphenols condensed with the lignin, increasing the yield of true lignin. When the raw material was first treated with a 0.125 N solution of NaOH, only the lightly polymerized phenolic compounds dissolved, and the others formed a lignin complex as "kino." On ethanolysis, this inhomogeneous lignin of the raw material investigated underwent cleavage to monomers of the protocatechuic, guaiacyl, and 4-hydroxyphenyl types.

The septa of the fruit of *Juglans regia* contain a small amount of polysaccharides. They are present in readily hydrolyzable and difficultly hydrolyzable forms. On acid hydrolysis, the readily hydrolyzable polysaccharides gave xylose and

Scientific Research Institute of Horticulture, Viticulture, and Wine-Making of the Georgian Academy of Agricultural Sciences, 6 Gelovani pr., Tbilisi 380015. Translated from Khimiya Prirodnykh Soedinenii, No. 2, pp. 161-164, March-April, 1998. Original article submitted February 10, 1997.

 0009-3130/98/3402-0128$20.00

TABLE 1. Chemical Composition of the Septa of the Fruit of *Juglans regia*

Fractions and compounds	Mass	Fractions and compounds	Mass
Fractions soluble in diethyl ether, % a.d.m.	8.1	protocatechuicaldehyde	13.2
Triglycerides, % in the lipid fraction	35.8	vanillic acid	18.7
Fatty acids in the triglycerides, % of total:		vanillyl alcohol	11.0
palmitic, $C_{16:0}$	8.4	syringic acid	25.0
stearic, $C_{18:0}$	3.0	syringyl alcohol	14.7
oleic, $C_{18:1}$	19.3	*p*-coumaric acid	4.2
linoleic, $C_{18:2}$	56.8	4-hydroxybenzaldehyde	6.4
linolenic, $C_{18:3}$	12.5		
Fraction soluble in 96% ethanol (after extraction of the raw material with diethyl ether), % a.d.m.	10.5	4-hydroxybenzoic acid	24.8
Catechins, % a.d.m.	1.2	ferulic acid	3.8
Proanthocyanidins, % a.d.m.	2.1	Fraction of polyphenols soluble in 0.125 N NaOH solution, % a.d.m.	14.5
Low-molecular-mass aromatic compounds, ml/liter (in the alcoholic extract): gallic acid	17.8	Lignin, % a.d.m.	39.0
protocatechuic acid	22.7	Readily hydrolyzable polysaccharides, including, %: xylan	9.2
			7.5
		Difficulty hydrolyzable polysaccharides, including, %: glucan	1.2
			1.0

mixtures of galactose and arabinose. Glucose predominated in a hydrolsate of the difficultly hydrolyzable polysaccharides. It can be seen from the constituent monosaccharides that the readily hydrolyzable polysaccharides were present mainly in the form of a xylan, and the difficultly hydrolyzable polysaccharides in the form of a glucan.

Thus, the septa of the fruit of *Juglans regia* are a rich raw material containing various natural compounds: lipids, low-molecular-mass phenolic carboxylic acids, phenolic aldehydes, catechins, proanthocyanidins, oligomeric and polymeric phenolic substances, and other extractive and nonextractive compounds.

EXPERIMENTAL

The ether-soluble and alcohol-soluble fractions were isolated by the exhaustive extraction of the raw material under investigation in a Soxhlet apparatus. The walnut septa, in 10-g batches, were extracted successively with 300 ml of diethyl ether and with 96% ethanol. The extracts were evaporated in a rotary evaporator at 40°C.

The phenolic compounds were investigated with the use of the alcoholic extract. The hexane-extracted raw material (10 g) was extracted with 96% ethanol (1:10) in a Soxhlet apparatus.

Qualitative analysis of the lipids was conducted by TLC on Silufol plates in the solvent system hexane–diethyl ether–acetic acid (85:14:1), and the chromatograms were revealed with iodine vapor [1]. Fatty acid compositions were determined by GLC.

Low-molecular-mass aromatic compounds were determined by HPLC [2].

Catechins and the products of the transformation of proanthocyanidins were determined qualitatively by paper chromatography in the solvent system butan-1-ol–acetic acid–water (4:1:5). To detect catechins, the chromatograms were revealed with the vanillin reagent. Quantitative analysis of the catechins and proanthocyanidins was conducted by the method of Swain and Hillis [4].

The monosaccharides formed by hydrolysis of the polysaccharides were determined by GLC under the following conditions: column, 3 mm; carrier gas, helium; liquid phase, OV-225, 3%; rate of flow 35.5 ml/min; temperature, 200°C; flame-ionization detector.

Polysaccharides were determined from the monosaccharides formed on their hydrolysis [5].

Lignins were determined by Klasons's method in Komarov's modification [6].

REFERENCES

1. E. Stahl, Thin Layer Chromatography, A Laboratory Handbook, 1st English ed., Springer/Academic Press, New York (1965).
2. V. D. Nemirovskii, Ya. D. Rakhmilevich, and V. G. Kostenko, Khim. Drev., No. 3, 22 (1987).
3. S. V. Durmishidze, A. G. Shalashvili, V. V. Mzhavanadze, and G. Ch. Tsiklauri, The Flavonoids and Hydroxycinnamic Acids of Some Representatives of the Georgian Flora [in Russian], Metsniereba, Tbilisi (1981), p. 139.
4. T. Swain and W. E. Hillis, J. Sci. Food Agric., **10**, 63 (1959).
5. A. V. Obolenskaya, V. P. Shchegolev, G. A. Akim, É. L. Akim, N. L. Kossovich, and I. Z. Emel'yanova, Practical Work in the Chemistry of Wood and Cellulose [in Russian], Lesnaya Prom-st', Moscow (1969), p. 134.
6. I. Z. Emel'yanova, The Chemical and Technological Control of Hydrolysis Factories [in Russian], Lesnaya Prom-st', Moscow (1976), p. 72.