Chemistry of Natural Compounds, Vol. 30, No. 1, 1994

PHENOLIC COMPOUNDS OF SUMAC

Sh. Yu. Islambekov, S. M. Mavlyanov, F. G. Kamaev, and A. I. Ismailov

UDC 547.982/83/84

*The main component of the polyphenolic complex has been isolated from the leaves of two species of sumac (*Rhus glabra *and* Rh. typhina*) and for this we have proposed the structure of 3,6-bis-O-digalloyl-1,2,4-tri-O-galloyl-β-D-glucose. A high interferon-inducing and antitumoral activity of the substance isolated has been found. In addition to catechins, 3',4',6-trihydroxyaurone has been tisolated from sumac stems.*

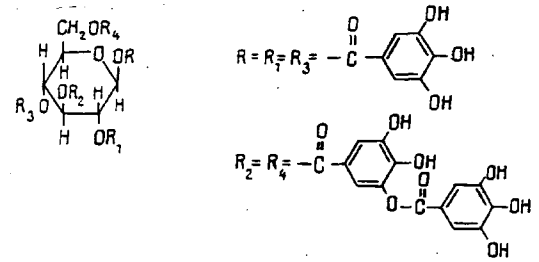
Recently, in the field of the chemistry of natural compounds the search for new biologically active substances from wild and cultivated plants that have been little studied from the chemical point of view has been extremely promising.

We have studied phenolic compounds of two species of sumac cultivated in Uzbekistan as promising tannin-bearing and decorative plants with the aim of creating new drugs with an antiviral, interferon-inducing, and antitumoral action.

We have studied the phenolic compounds of these species of sumac previously [1, 2]. In the present communication we give the results of investigations of the polyphenols of the leaves and stems of the sumac. With the aid of column chromatography on silica gel we have isolated the main component from a total preparation of the polyphenols of sumac leaves, which consisted of a light-colored amorphous powder with a faint yellowish tinge having mp 216°C (decomp.), readily soluble in water, alcohol, ethanol, and acetone, less readily in ethyl acetate, and insoluble in hexane, chloroform, and benzene.

In the products of complete acid hydrolysis of 150 mg of the substance, performed with the aid of 2N HCl in a current of nitrogen, we detected glucose and gallic acid. Glucose was determined in the hydrolysate by the micro method for the determination of sugars [3] and gallic acid photocolorimetrically from a calibration graph drawn up with a standard sample of gallic acid. The amount of glucose was 18.45 mg and that of gallic acid 128 mg. It follows from the figures obtained that the ratio of glucose and gallic acid in the molecule was 1:7, which was also confirmed by the PMR spectrum. In the spectrum we observed a broad structureless singlet at 8.43 ppm belonging to the protons of phenol groups. The aromatic protons of gallic acid residues resonated in the 6.9-7.7 ppm region in the form of a set of narrow signals. The signals of the glucose protons were located at 6.36, 6.05, and 5.68 ppm and in the 4.25-4.70 ppm region with integral intensities of 1:1:2:3, respectively. According to the integral curve, for each glucose residue there were seven gallic acid residues. From the low-field position of the signals of the protons of the glucose residue it followed that all the hydroxy groups of the glucose were substituted and, consequently, there were two digallic acid residues in the molecule of the substance.

The existence of several isomers of pentagalloylglucose with two digalloyl residues is known in the literature (2,3-bis-O-digalloyl-1,4,6-tri-O-galloyl-β-D-glucose;3,4-bis-O-digalloyl-1,2,6-tri-O-galloyl-β-D-glucose;and2,4-bis-O-digalloyl-1,3,6-tri-O-galloyl-β-D-glucose [5]). From its angle of rotation (+25.88°, *c* 0.85; acetone) the substance that we had isolated was close to 2,3-bis-O-digalloyl-1,4,6-tri-O-galloyl-β-D-glucose (+24.4°, *c* 1.35; acetone). However, the absence from the literature of other physicochemical characteristics for the other isomers of bis-digalloyl-trigalloylglucose does not exclude the possibility of the existence of the compounds that we isolated in the form of 3,6-bis-o-digalloyl-1,2,4-tri-O-galloyl-β-D-glucose.



A. S. Sadykov Institute of Bioorganic Chemistry, Academy of Sciences of the Republic of Uzbekistan, Tashkent, fax 627071. Translated from Khimiya Prirodnykh Soedinenii, No. 1, pp. 45-48, January-February, 1994. Original article submitted July 13, 1993.

0009-3130/94/3001-0037$12.50

From the total polyphenols isolated from the ether fraction of an aqueous acetone (70%) extract of the stems of *Rh. glabra* with the aid of column chromatography on silica gel and cellulose, in addition to catechins, we isolated in the individual form a yellow substance with R_f 0.70 on paper in solvent system 1. The amount of this substance considerably exceeded that of catechins. From qualitative reactions, R_f values, in various solvent systems, the results of UV spectroscopy with additives and without them, and its PMR spectrum, it was identified as 3′,4′,6-trihydroxyaurone, which is known under the name of sulphuretin. The two sumac species differed slightly in their sulphuretin contents.

Together with catechins and sulphuretin, in the polyphenol complex of the stems of both species of sumac we detected a substance appearing on a chromatogram when it was sprayed with revealing agent 2 in the form of a band with R_f from 0. 20 to 0.40. In all probability, this substance was a proanthocyanidin; however, because of its small amount we did not study it further.

Investigations performed in the Institute of Virology and Microbiology of the Russian Ministry of Health to which we passed the tannin from sumac leaves, together with other preparations, have shown that it is a highly active inductor of interferon with a very low toxicity both in vitro and in vivo. The high solubility of the preparation in water makes it possible to introduce it into the organism both perorally and parenterally. In addition, investigations of sumac tannin performed in the Scientific-Research Institute of Experimental Diagnosis and Tumor Therapy of the All-Russian Oncological Scientific Center have shown a high antitumoral activity. Thus, the average index of the inhibition of the incorporation of labeled thymidine into the DNA of cells for sumac tannin is 96.3%.

EXPERIMENTAL

For column chromatography we used silica gel of types L40/100 and Silpearl, and cellulose. For PC we used Filtrak paper. The following solvent systems were employed: 1) *n*-butanol–acetic acid–water (40:12:28); 2) a 6% solution of acetic acid; 3) moist diethyl ether; 4) triethyl ether–ethyl acetate (1:1); and 5) ethyl acetate. Revealing agents: 1) mixture of 1% aqueous solutions of $FeCl_3$ and $K_3Fe(CN)_6$ (1:1) and 2) 1% solution of vanillin in concentrated hydrochloric acid.

Isolation of the Main Component of the Polyphenolic Complex of Sumac Leaves. Comminuted air-dry leaves (300 g) were extracted with hot 70% aqueous acetone (at a liquor ratio of 1:10) three times. The extracts were filtered and dried, and the aqueous residue was treated successively with chloroform (three times) and with ethyl acetate (five times). The ethyl acetate extracts, dried with freshly calcined sodium sulfate, were concentrated, the total polyphenols were precipitated with hexane, and the precipitate was filtered off and dried. This gave 33.0 g of total polyphenols (11% on the weight of the leaves).

To isolate the main component of the polyphenolic complex of sumac leaves, a solution of 4 g of the total substances isolated in 25 ml of acetone was mixed with 30 g of silica gel, the acetone was evaporated off, and the resulting powder was transferred to a column of silica gel (120 g). Solvent systems 3 and 4 were used as eluents. The fractions obtained when the column was washed with system 4, which contained only one substance, with R_f 0.53, were combined, dried with freshly calcined sodium sulfate, and concentrated, and the substance was precipitated with a fivefold volume of hexane. The precipitate that had deposited was filtered off, washed with hexane, and dried in a vacuum-drying chamber.

This gave 1. 84 g of a light cream-colored amorphous powder with a burning taste. The substance was readily soluble in water, ethanol, methanol, and acetone, soluble in ethyl acetate, sparingly soluble in diethyl ether, and insoluble in chloroform, hexane, benzene, and petroleum ether, R_f 0.53 (PC, solvent system 1); mp 61°C (decomp.). UV spectrum, λ_{max}^{MeOH}, nm: 217, 280; $[\alpha]_D^{22}$ 25.88° (*c* 0.85; acetone). PMR spectrum (taken on Varian XL-200 instrument with a working frequency of 200 MHz): 8.43, 6.36, 6.90–7.70, 6.05, 5.68, 4.25-4.70 ppm.

The analysis of the substance corresponded to the calculated figures.

Isolation of an Aurone from Sumac Stems. Comminuted air-dry stems (branches) (500 g) were extracted three time with hot 70% aqueous acetone (liquor ratio 1:5). The combined acetone extract was filtered and concentrated, and the concentrate (~1.0 liter) was treated with chloroform (3 × 300 ml), and then with diethyl ether (seven times). The combined ether extracts (~2.0 liters) were dried with sodium sulfate, concentrated to a volume of 120 ml, and chromatographed on a column of silica gel by M. N. Zaprometov's method [4]. The eluent was moist diethyl ether.

The course of the separation was monitored by PC and from the reaction with reagent 2. As a result, we obtained fraction 1, containing a yellow substance with R_f 0.70 and (+)-catechin (R_f 0.64); fraction 2, containing (+)-catechin and (−)-epicatechin; and fraction 3, containing (−)-epicatechin and (−)-epigallocatechin).

Fraction 1 was concentrated, and the residue was dissolved in a small volume of distilled water and chromatographed on cellulose powder (3 × 50 cm). The eluent was solvent system 2. The fractions of eluate containing only one yellow substance with R_f 0.70 were combined, washed with water in a separatory funnel, and then treated with ethyl acetate (5 × 50 ml), and the ethyl acetate extracts were combined, dried with anhydrous sodium sulfate, filtered, and concentrated under vacuum at 35-40°C to small volume. The substance was precipitated by the addition of a fourfold volume of chloroform. The precipitate was filtered off on a Schott No. 3 funnel, washed with chloroform, and dried in a vacuum-drying chamber at 45°C for 5 h. This gave 0.85 g of a yellow pulverulent substance with mp 280°C. UV spectrum, λ_{max}^{MeOH}: 272, 398 nm.

The analyses of the substance corresponded to the calculated figures.

REFERENCES

1. S. M. Mavlyanov, Sh. Yu. Islambekov, A. K. Karimov, A. K. Karimdzhanov, A. I. Ismailov, and E. V. Gnatchenko, in: Abstracts of Lectures at the Vth All-Union Symposium on Phenolic Compounds. Chemistry Section [in Russian], Tallinn (1987), p. 56.
2. Sh. Yu. Islambekov, S. M. Mavlyanov, A. K. Karimov, A. R. Bakhidova, and A. K. Karimdzhanov, Khim. Prir. Soedin., 787 (1988).
3. A. I. Ermakov, Methods for the Biochemical Investigation of Plants [in Russian], Agropromizdat, Leningrad (1987), p. 430.
4. M. N. Zaprometov, The Biochemistry of Catechins [in Russian], Nauka, Moscow (1964).
5. Makato Nishisawa and Takashi Yamagishi, J. Chem. Soc., Perkin Trans. I, 2963 (1982).