Summary

The persulfate oxidation of 20-keto-*allo*-pregnane compounds gave a mixture of androstanols-17 and *etio*-*allo*-cholanic acid.

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[CONTRIBUTION FROM THE CHEMICAL LABORATORY OF THE OHIO STATE UNIVERSITY]

Osage Orange Pigments. III. Fractionation and Oxidation

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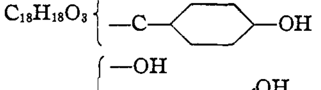
In continuation of our studies[1] on the pigments of the fruit of the osage orange tree (*Maclura pomifera* Raf.) we have made a rather exhaustive fractionation of the naturally occurring mixture of the two pigments, the results of which indicate that the two are probably present in about equal amounts.

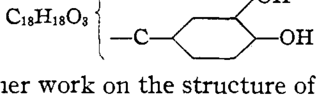
The melting point–composition diagram obtained from mixtures of the two pure substances is shown in Fig. 1. The melting point behavior obtained on mixing the pure compounds is probably slightly different from the behavior of mixtures of equal composition which have been formed by co-crystallization.

The trimethyl ether of pomiferin has been reported previously.[1b] We have now obtained the corresponding dimethyl ether of osajin. When the former is subjected to oxidation, in acetone solution, with alkaline hydrogen peroxide, veratric (3,4-dimethoxybenzoic) acid is produced. Under the same conditions, osajin dimethyl ether yields anisic (*p*-methoxybenzoic) acid. These two acids were identified by analysis, by preparation of the anilide and amide of each, and by comparison with authentic specimens in all cases. The two acids have practically identical melting points. This is also true for their amides and anilides. Mixed melting points, however, give definite depressions.

The formulas of osajin (I) and pomiferin (II), both of which are methoxyl-free, may now be developed as follows



(I)



(II)

Further work on the structure of these two substances is in progress in this Laboratory.

Experimental

Fractionation.—The whole fruit of the osage orange tree (*Maclura pomifera* Raf.) was cut, dried and crushed to a coarse meal. The meal (1500 g.) was extracted successively with petroleum ether and ether. From the latter extract was obtained a yellow solid; yield 47.8 g., m. p. 180–182°. This material was recrystallized from ten parts of hot xylene, the crop being removed by filtration when the temperature of the solution had reached approximately 40°. On repeated recrystallization in this manner pure pomiferin was obtained; yield 10.0 g., m. p. 200.5°. The combined mother liquors were concentrated to give a product melting at 173–175° and after repeated recrystallization from 95% ethanol pure osajin was obtained; yield 5.6 g., m. p. 189°.. By employing a method of crystallizing the xylene mother liquor material from 95% alcohol, and then the alcohol mother liquor material from xylene again, and continuing this cycle, a total of 14.1 g. of high purity osajin and 15.8 g. of high purity pomiferin was obtained.

Although not all the material was accounted for, the above results would seem to indicate that the two substances are present in the crude extract in very nearly equal amounts. The total yield of crude extract in this lot of material was not as high as in the crops of previous seasons. The melting point–composition diagram obtained from mixtures of the two pure substances is shown in Fig. 1. The melting point behavior obtained on mixing the two pure substances is probably slightly different from the behavior of mixtures of equal composition which have been formed by co-crystallization.

Osajin Dimethyl Ether.—To a boiling solution of osajin (2 g., m. p. 189°) in acetone (130 cc.) were added under reflux the following portions of reagents over a period of forty minutes: 24 cc. of dimethyl sulfate, 14 cc. of 50% aqueous potassium hydroxide, then six alternate portions of 6 cc. each of dimethyl sulfate and 8 cc. each of 50% aqueous potassium hydroxide. The refluxing was continued for two hours longer and then the reaction mixture was poured on 1.5 l. of crushed ice and allowed to stand until the ice had melted before filtration of the precipitated solid; yield 2.16 g., m. p. 109°. Pure material was obtained on several recrystallizations from ethanol (95%); m. p. 118.5° unchanged on recrystallization from methanol, propanol-1 and benzene–petroleum ether. The substance crystallized in long, colorless needles and gave a negative ferric chloride–alcohol phenol test.

Anal. Calcd. for $C_{25}H_{22}O_3(OCH_3)_2$: C, 74.98; H, 6.53; OCH_3, 14.35. Found: C, 74.91; H, 6.53; OCH_3, 14.3.

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[1] (a) E. D. Walter, M. L. Wolfrom and W. W. Hess, THIS JOURNAL, **60**, 574 (1938); (b) M. L. Wolfrom, F. L. Benton, A. S. Gregory, W. W. Hess, J. E. Mahan and P. W. Morgan, *ibid.*, **61**, 2832 (1939).

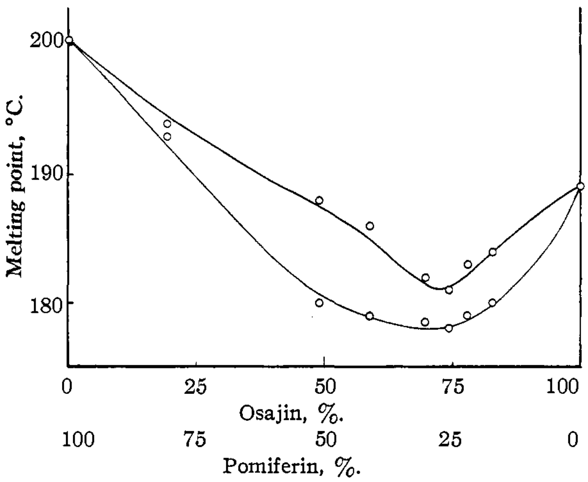

Fig. 1.—Melting point–composition diagram for mixtures of osajin and pomiferin. Lower curve represents beginning of melting and upper curve represents complete fusion.

Isolation of Anisic Acid from the Oxidation of Osajin Dimethyl Ether with Alkaline Peroxide.—Osajin dimethyl ether (1.7 g., m. p. 118°) dissolved in acetone (150 cc.) was treated under mechanical stirring with 1.5 cc. of 50% aqueous potassium hydroxide, followed by the dropwise addition of 20 cc. of 30% aqueous hydrogen peroxide. The oxidation was continued for forty-eight hours at room temperature, whereupon the solution tested only faintly oxidizing to potassium iodide paper. Saturated aqueous sodium bisulfite solution (5 cc.) was added slowly with cooling and the mixture then was neutral to potassium iodide paper and basic to litmus. The acetone was removed under reduced pressure, 50 cc. of 1% hydrochloric acid added and the solution extracted with ether. The concentrated ethereal extract (75 cc.) was extracted with several portions of 5% ammonium carbonate solution. This carbonate solution was concentrated to 50 cc., some tarry material removed by filtration, and the white precipitate that formed on acidification with hydrochloric acid was recrystallized from hot water; yield 0.10 g., m. p. 178°. After two further recrystallizations from water (decolorizing charcoal) the product appeared as fine white needles melting at 180°, which formed an anilide of melting point 167–168° and an amide of melting point 163–164°. Authentic specimens of anisic acid (m. p. 180°), anisic acid anilide (m. p. 167–168°) and anisic acid amide (m. p. 163–164°) were prepared and showed no depression in melting point with the corresponding products obtained from the oxidation.

Anal. Calcd. for $C_8H_8O_3$ (anisic acid): OCH_3, 20.4; neutral equivalent, 152. Found: OCH_3, 21.0; neutral equivalent, 157. Calcd. for $C_8H_9O_2N$ (anisic acid amide): OCH_3, 20.5. Found: OCH_3, 20.3.

Isolation of Veratric Acid from the Oxidation of Pomiferin Trimethyl Ether with Alkaline Peroxide.—In the description of the synthesis of pomiferin trimethyl ether[1b] previously described from this Laboratory, the addition of 14 cc. of 50% aqueous potassium hydroxide should follow the addition of the first amount of dimethyl sulfate. Pomiferin trimethyl ether (1.7 g.) was oxidized with alkaline peroxide exactly as described for osajin dimethyl ether. On the addition of the alkali and hydrogen peroxide a turbidity appeared which cleared slowly on continued stirring. The acid fraction of the oxidation mixture was isolated as described for the osajin dimethyl ether and yielded a crystalline product; yield 0.24 g., m. p. 173–175°. Pure material was obtained on several recrystallizations from hot water (decolorizing charcoal); m. p. 179.5°. The substance was identified as veratric acid by preparation of its anilide (m. p. 166.5–167°) and amide (m. p. 164°). The substance and its two derivatives showed no depression in melting point on admixture with authentic specimens of veratric acid, veratric acid anilide and veratric acid amide. Mixed melting points of (1) veratric acid (179.5°) with anisic acid (180°), (2) veratric anilide (166.5°) with anisic anilide (167–168°) and (3) veratric amide (164°) with anisic amide (163–164°) were, respectively: (1) 146–162°, (2) 138–142° and (3) 132–140°. The isolated substance also showed the sublimation characteristic of veratric acid.

Anal. Calcd. for $C_9H_{10}O_4$ (veratric acid): OCH_3, 34.1; neutral equivalent, 182. Found: OCH_3, 33.5; neutral equivalent, 182.

We are indebted to Mr. John Walker (W. P. A. Project 18062) for assistance rendered in the analytical determinations and to Mr. Joseph Tracht (N. Y. A. Project O. S. U. 181) for assistance in preparing the plant material.

Summary

1. It is shown that the two pigments of the fruit of the osage orange (*Maclura pomifera* Raf.) are present in approximately equal amounts.

2. Osajin dimethyl ether has been prepared.

3. Anisic (*p*-methoxybenzoic) acid has been obtained from the oxidation of osajin dimethyl ether with alkaline hydrogen peroxide.

4. Veratric (3,4-dimethoxybenzoic) acid has been obtained from the oxidation of pomiferin trimethyl ether with alkaline hydrogen peroxide.

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