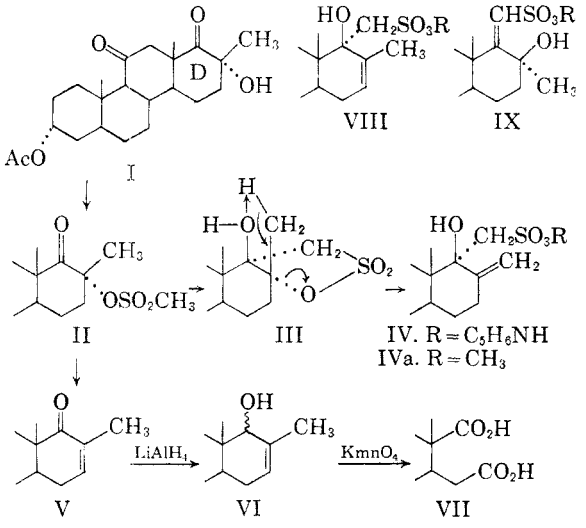
This compound consequently has been formulated as the inner aldol (III).[2] On refluxing in pyridine (conditions which convert II→V[1a]), III was transformed to the pyridinium salt of a sulfonic acid, assigned structure IV on the basis of the following evidence:

The pyridinium salt was water soluble and could be titrated with perchloric acid to give an equivalent weight in excellent agreement with that calculated for IV or an isomer. Treatment of the pyridinium salt in methanol with ethereal diazomethane converted it to the corresponding methyl ester IVa. This ester exhibited no maximum in the ultraviolet, but possessed OH absorption at 2.83 μ as well as intense double bond absorption at 6.04 μ in the infrared. Although this ester was essentially inert to neutral permanganate, it did react slowly with osmium tetroxide thereby chemically confirming the presence of a double bond.



The lack of ultraviolet absorption is inconsistent with the isomeric possibility IX; the data are incompatible as well with the alternate structure VIII as this double bonded type, *e.g.* VI, exhibits no double bond absorption in the infrared under comparable conditions of measurement (see Experimental) and is readily oxidized by neutral permanganate to the etiobilianic acid VII.[1] Finally, the NMR spectrum was consistent with IVa and, by establishing the absence of any D-ring methyl group, clearly eliminated structures VIII and IX.[3]

The formation of the exomethylenic system IV would appear to constitute an example of the Arnold-Schinz mechanism of cyclicly assisted dehydration.[4]

EXPERIMENTAL[5]

Reaction of 3α-acetoxy-17α-hydroxy-17β-methyl-D-homo-5β-androstane-11,17a-dione (I) *with methanesulfonyl chloride.* A solution of 4.1 g. of I in 15 cc. of anhydrous pyridine was treated at 0° with 4 cc. of methanesulfonyl chloride and allowed to stand at 0° for 16 hr. The reaction product was treated with ice and ether. The ether extract was washed successively with dilute aqueous hydrochloric acid, potassium bicarbonate, and sodium chloride solution. The washed ether solution was dried over magnesium sulfate, filtered, and evaporated to the point of incipient turbidity. This solution deposited 1.5 g. of III over a period of several days. Recrystallization from acetone-hexane afforded III as slender prisms, m.p. 142–144° dec., λ_{max}^{Nujol} 3 μ (OH); 5.85 μ (C=O); 5.8 μ, 8 μ (OAc), 7.4 μ, 8.6 μ (OSO_2).

Anal. Calcd. for $C_{24}H_{36}O_7S$: C, 61.54; H, 7.70; S, 6.84. Found: C, 61.65; H, 7.70; S, 6.49.

Pyridinium salt (IV). A solution of 3 g. of III in 50 cc. of pyridine was refluxed for 2 hr. and evaporated to dryness *in vacuo.* The residue was crystallized from acetone-ether, 2.8 g., m.p. 145–150° λ_{max}^{Nujol} 2.97 μ (OH/NH); 5.89 μ (C=O), 5.79, 8 μ (OAc), 6.1, 6.42 μ (Pyridine).

Anal. Calcd. for $C_{29}H_{41}O_7NS$: C, 63.62; H, 7.49; N, 2.74; S, 5.86; Eq. wt., 547. Found: C, 63.88; H, 7.17; N, 2.57; S, 6.01; Eq. wt., 533.

Methyl ester (IVa). Treatment of the pyridinium salt (IV) in methanol solution with an excess of ethereal diazomethane afforded the methyl ester (VIa) crystallized from ether, m.p. 180.5–182°, $\lambda_{max}^{CHCl_3}$ 2.83 μ (OH); 5.8–5.85 μ, 8 μ (C=O, OAc); 7.3 μ, 8.5–8.6 μ (OSO_2), 6.04 μ (C=C).

Anal. Calcd. for $C_{25}H_{38}O_7S$: C, 62.24; H, 7.88; S, 6.64. Found: C, 62.46; H, 7.85; S, 6.71.

The above ester (100 mg.) was recovered essentially unchanged after oxidation with potassium permanganate (200 mg.) in acetone (15 cc.) at 25° for 2 hr. or after refluxing for 1 hr.

Δ^{16}-17-Methyl-D-homo-5β-androstene-3α,11β,17a-triol (VI). A solution of 100 mg. of the $\Delta^{\alpha,\beta}$ ketone (V)[1] in 20 cc. of ether was reduced with 200 mg. of lithium aluminum hydride at room temperature for 5 hr. The isolated triol (VI) crystallized from ether, m.p. *ca.* 280° λ_{max}^{Nujol} 2.81, 3.03 μ (OH).

Anal. Calcd. for $C_{21}H_{34}O_3$: C, 75.45; H, 10.18. Found: C, 75.49; H, 9.87.

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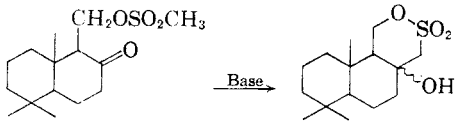
(2) E. Romann, A. J. Frey, P. A. Stadler, and A. Eschenmoser, [*Helv. Chim. Acta.*, **40**, 1900 (1957) Footnote 13] have reported a similar reaction:



We are grateful to Professor R. B. Woodward for calling this reference to our attention.

(3) The authors are grateful to N. R. Trenner and B. Arison for the NMR determination.

(4) R. T. Arnold, *Helv. Chim. Acta.*, **32**, 134 (1949); H. Schinz and G. Schäppi, *Helv. Chim. Acta.* **30**, 1483 (1947).

(5) Melting points were taken on a micro hot stage and are corrected. Infrared spectra were determined on a Baird Associates Infrared Spectrophotometer.

Configuration and Conformation of 3-Bromohesperetin Triacetate. Dimorphs of Hesperetin Triacetate[1,2]

J. H. LOOKER AND MYRON J. HOLM[3]

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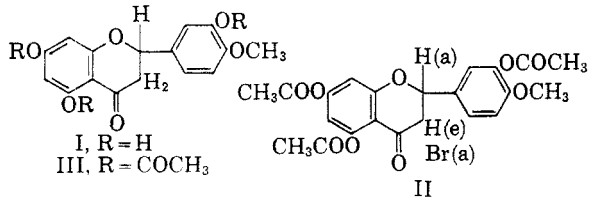
Hesperetin (I), the aglycon of the important glycoside hesperidin, has been investigated rather

intensively recently. Of particular interest stereochemically is a study which establishes the absolute configuration of (—)-hesperetin.[4] The present note describes spectral and chemical data which establish the configuration and conformation of 3-bromohesperetin triacetate (II).



II

Hesperetin (I) was obtained from hesperidin by a new, simple method involving heating of the glycoside for 60 to 72 hours in 2% sulfuric acid in 50% ethanol. Acetylation of racemic I gave the triacetate III, which is dimorphous, m.p. 126.5–127° and 143.5–144.5°. Carbon–hydrogen analyses for both high and low-melting forms were in agreement with theory for the triacetate. Neither form lost weight at 100° *in vacuo*, thus indicating that solvation is not responsible for the different melting points. Interconversion of high and low-melting forms was readily effected (Experimental). Thus III is considered dimorphous. Confirmation was obtained from infrared spectral data[5] (Experimental). Solution spectra of the two forms are identical, but solid state spectra of the individual dimorphs differ.

Our melting point data confirm Perkin's observation[6] of a melting point in the vicinity of 127° for III. However, as other workers[7] have assigned a chalcone tetraacetate structure to a hesperetin acetylation product, m.p. 127°, the structure of this substance has been reinvestigated. Acetyl determination under acidic conditions revealed the presence of only three acetoxyl groups. A ferric chloride test was negative, precluding the possibility of a hydroxytriacetoxychalcone structure. The infrared spectrum in carbon tetrachloride showed a band at 1691 cm.$^{-1}$, indicative of a carbonyl group of a flavanone.[8] The ultraviolet spectrum also indicates a flavanone, as the intensity of the broad band at 3140 Å is not as great as that expected for a chalcone.[9] The product gives a positive magnesium-hydrochloric acid test for a flavonoid,[10] although the color develops rather slowly. We conclude that the acetylation product of hesperetin, m.p. 127°, is the flavanone derivative (III).

3-Bromohesperetin triacetate (II), first described by Zemplen and Bognar,[11] was prepared by their general procedure, as recently described.[12] The 3-bromo derivative (II) loses hydrogen bromide in presence of silver acetate–acetic anhydride,[12] and in pyridine, even at −5°, with formation of diosmetin triacetate. The latter reaction in particular involves ready elimination of hydrogen bromide, and thus the hydrogen at carbon-2 is very probably *trans* to the bromine at carbon-3.[13] Infrared spectral data for methylene chloride solutions of II and III show that the keto carbonyl bands are at 1698 and 1693 cm.$^{-1}$, respectively. The small spectral shift of 5 cm.$^{-1}$ indicates that the bromine *alpha* to the carbonyl group probably is in an axial position.[14] Accordingly, the *trans*-hydrogen at carbon-2 also must be axial, as indicated in formula II. The conformation of II is the expected one if the Corey rule[15] pertaining to bromination of ketosteroids under kinetic conditions is extended to the flavanone (III). However, the detailed mechanism may be different in the present case, as the bromination is effected under irradiation with ultraviolet light. These conditions could lead to homolytic steps in the early stages of the bromination. However, the hydrogen bromide subsequently produced may catalyze enolization, and the enol usually postulated then could participate in an ionic bromination process. In view of the apparent conformational stability of II, it is likely that the hydrogen atom at carbon-2 in I and III also is axial.

EXPERIMENTAL[16]

Hydrolysis of hesperidin. A 5.0-g. quantity of hesperidin was suspended in a solution of 2.5 ml. of concd. sulfuric acid in 250 ml. of 50% aqueous ethanol, heated until solu-

(1) From the M.S. (1956) and Ph.D. (1958) theses of Myron James Holm.

(2) This investigation was supported in part by a research grant (E-1703) from the National Institute of Allergic and Infectious Diseases, Public Health Service.

(3) DuPont Postgraduate Teaching Assistant, 1956–1957; Standard Oil of Indiana Foundation Fellow, 1957–1958.

(4) H. Arakawa and M. Nakazaki, *Chem. & Ind. (London)*, 1960, 73.

(5) F. A. Miller, in *Organic Chemistry*, Vol. III, edited by H. Gilman, John Wiley and Sons, New York, N. Y. (1953), p. 139.

(6) A. G. Perkin, *J. Chem. Soc.*, 1031 (1898).

(7) F. Tutin, *J. Chem. Soc.*, 2054 (1910); O. A. Oesterle and R. Kueny, *Arch. Pharm.*, **253**, 383 (1915); Y. Asahina, J. Shinoda, and M. Inubuse, *J. Pharm. Soc. Japan*, **48**, 207 (1928). The procedure of Tutin was identical with that of Perkin, except for reaction period. Tutin characterized his product by acetyl determination in basic medium. The other workers cited herein utilized acetic anhydride and sodium acetate for acetylation. In our hands, the latter procedure gave hesperetin triacetate.

(8) H. L. Hergert and E. F. Kurth, *J. Am. Chem. Soc.*, **75**, 1622 (1953).

(9) Recent data supporting this statement are presented by M. Shimokoriyama, *J. Am. Chem. Soc.*, **79**, 214 (1957).

(10) For leading references, see S. Rangaswami and T. R. Seshadri, *Proc. Indian Acad. Sci.*, **16A**, 129 (1942).

(11) G. Zemplen and R. Bognar, *Ber.*, **76B**, 454 (1943).

(12) J. H. Looker and M. J. Holm, *J. Org. Chem.*, **24**, 1019 (1959).

(13) A review of the four centers in a plane generalization is given by D. H. R. Barton, *J. Chem. Soc.*, 1030 (1953).

(14) R. N. Jones, D. A. Ramsay, F. Herling, and K. Dobriner, *J. Am. Chem. Soc.*, **74**, 2828 (1952).

(15) E. J. Corey, *J. Am. Chem. Soc.*, **76**, 176 (1954).

tion was complete (usually *ca.* 75 hr.) and then an additional 5 hr. The *p*H was adjusted to between 4 and 5. Most of the alcohol was removed by distillation, the residual solution filtered while hot, and the filtrate allowed to cool. The light tan, crude, crystalline hesperetin was collected by filtration and air-dried; yield, 1.78 g. (72%), m.p. 225–227° with softening at 218°. The crude product was recrystallized by solution in hot isopropyl alcohol (10–11 ml.), cooling, and permitting the mixture to stand 12 hr. in a refrigerator. The recrystallized hesperetin (1.15 g.) melted at 228–229.5° (lit.,[6,17] m.p. 226°). The purest hesperetin obtained in this study melted at 228.5–229.2°. By sublimation, a product, m.p. 232.8°, has been previously reported.[18]

Dimorphous forms of hesperetin triacetate. A 7.00-g. quantity of hesperetin, m.p. 226–228°, was dissolved in a mixture of 45 ml. of acetic anhydride in 45 ml. of pyridine and permitted to stand at room temperature for 24 hr. The mixture was poured into 800 ml. of crushed ice-water, and the precipitated product collected and dissolved in 100 ml. of boiling 95% ethanol. Cooling the ethanol solution at 0° for 12 hr. gave 8.00 g. (80%) of the crude triacetate. Recrystallization from 75 ml. of 95% ethanol gave 7.50 g., m.p. 141.5–143.5°, and two additional crystallizations gave 6.68 g. of hesperetin triacetate, m.p. 143.5–144.5°. In another example of this purification, the melting point after the fourth crystallization was 143.5–144.2°. After a fifth crystallization hesperetin triacetate, m.p. 126.5–127°, was obtained.

Anal. Calcd. for $C_{22}H_{20}O_9$: C, 61.67; H, 4.71; CH_3CO, 30.14. Found: (For dimorph, m.p. 126.5–127°): C, 61.89; H, 4.78. (For dimorph, m.p. 143.5–144.5°): C, 61.94; H, 4.98; CH_3CO, 29.6. The residue from the acetyl determination was isolated, recrystallized from ethanol and shown to be hesperetin by melting point and mixed melting point determination.

Hesperetin triacetate, m.p. 143.4–144.2°, was dissolved in hot 95% ethanol, and the resulting solution filtered into a flask containing seed crystals of the dimorph, m.p. 126.5–127°. Upon cooling, the total separated hesperetin triacetate was collected, air-dried, and found to melt 126.5–127°. When the dimorph, m.p. 126.5–127°, was melted on a Kofler hot stage[19] and held at 130–135° for several minutes, crystals grew in the melt and remelted between 145 and 147°. Samples of both dimorphs were weighed, dried in a drying pistol *in vacuo* for 1 hr., and reweighed. No change in weight was observed.

In Nujol mull, the infrared spectrum of the dimorph, m.p. 143.5–144.5°, showed strong or medium absorption bands at 1760, 1685, 1618, 1581, 1520, 1444, 1330, 1279, 1269, 1212, 1190, 1136, 1130, 1076, 1060, 1028, 903, and 809 cm^{-1}. The Nujol spectrum of the dimorph, m.p. 126.5–127°, showed strong or medium absorption bands at 1755, 1670, 1616, 1570, 1515, 1438, 1331, 1282, 1262, 1249, 1210 (broad), 1180, 1129, 1074, 1060, 1025, 899, and 817 cm^{-1}. In carbon tetrachloride solution, the dimorphs gave identical infrared spectra, showing absorption bands at 1775, 1691, 1618, 1437, 1369, 1327, 1188 (broad), 1127, 1073, 1023, and 898 cm^{-1}. The ultraviolet absorption spectrum of the high-melting dimorph in absolute ethanol showed maxima at 220 mμ (ϵ 38.7 × 10^3), 259 mμ (ϵ 11.2 × 10^3), and 314 mμ (ϵ 3.89 × 10^3), and minima at 241 mμ (ϵ 6.22 × 10^3) and 288 mμ (ϵ 1.81 × 10^3).

3-Bromohesperetin triacetate. This substance was prepared as previously described.[11,12] In a potassium bromide pellet, the infrared spectrum of 3-bromohesperetin triacetate showed strong or medium absorption bands at 1771, 1692, 1620, 1580, 1517, 1432, 1372, 1273, 1202, 1180, 1127, 1071, 1022, 903, and 810 cm^{-1}.

Diosmetin triacetate from 3-bromohesperetin triacetate and pyridine. A 0.5-g. sample of 3-bromohesperetin triacetate was dissolved in 20 ml. of cold pyridine and stored for 1 week at −5°. The mixture was then placed in a desiccator containing concd. sulfuric acid. The desiccator was evacuated and stored at −5°. When all of the pyridine had been absorbed by the sulfuric acid, the reaction vessel was removed and the residue triturated with cold methanol. After standing at *ca.* 5° overnight, 0.29 g. (70%) of diosmetin triacetate was collected by filtration and recrystallized from methanol; m.p. 195–197° (lit. m.p.[20] 195–196°). The solid state spectrum (potassium bromide pellet) was identical with that of diosmetin triacetate obtained as previously described.[12]

Anal. Calcd. for $C_{22}H_{18}O_9$: C, 61.97; H, 4.26. Found: C, 62.55, 62.28; H, 4.30, 4.27.

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(16) Melting points are uncorrected and were observed in capillary tubes. The infrared measurements were carried out with a Perkin-Elmer Model 21 double-beam recording spectrophotometer. A Cary recording spectrophotometer was employed for ultraviolet spectral measurements.

(17) F. Tiemann and W. Will, *Ber.*, **14**, 951 (1881).

(18) R. Seka and G. Prosche, *Monatsh.*, **69**, 284 (1936).

(19) L. Kofler, *Angew. Chem.*, **51**, 703 (1938).

(20) A. Lovecy, R. Robinson, and S. Sugasawa, *J. Chem. Soc.*, 817 (1930).

Steroidal Esters of 3-Indoleacetic Acid[1-3]

JOHN F. HOFERT AND HAROLD M. SELL

Received March 29, 1960

As alterations of the lyophilic properties of the alcohol portion might enhance the activity for parthenocarpic fruit induction in the tomato,[4,5] steryl esters of 3-indoleacetic acid have been prepared and tested for biological activity.

Attempts to synthesize the esters using 3-indoleacetyl chloride[6,7] and free sterol in pyridine[8] resulted in the formation of a highly insoluble orange-red material. The preparation of the esters was accomplished by using the free sterol, silver carbonate, and approximately twice the theoretical amount of acyl chloride in benzene or petroleum ether. The products were purified by recrystallization to constant melting point followed by removal of the final traces of free sterol with digitonin.[9]

(1) Journal Article No. 2594 of the Michigan Agricultural Experiment Station.

(2) This research was supported by a grant from the National Science Foundation.

(3) From the masters thesis of J. Hofert, Michigan State University.

(4) H. M. Sell, S. H. Wittwer, T. L. Rebstock, and C. T. Redemann, *Plant Physiol.*, **28**, 481 (1953).

(5) L. E. Weller, S. H. Wittwer, and H. M. Sell, *J. Am. Chem. Soc.*, **77**, 4937 (1955).

(6) E. Shaw and D. W. Woolley, *J. Biol. Chem.*, **203**, 979 (1953).

(7) K. N. F. Shaw, A. McMillan, A. G. Gudmundson, and M. D. Armstrong, *J. Org. Chem.*, **23**, 1171 (1958).

(8) J. F. Norris, and G. W. Rigby, *J. Am. Chem. Soc.*, **54**, 2088 (1932).

(9) L. F. Fieser, *Natural Products Related to Phenanthrene*, Reinhold Publishing Corp., New York, 1949, p. 102.