

readily reduced the latter after mild acid hydrolysis and appeared to be the methyl acetal of an aldehyde or ketone. This in turn was possibly a fragment of a still larger chemical unit which was oxidized with the starch. No conclusive evidence was obtained to indicate whether the crystals were derived from an integral part of the starch macromolecule or from an associated impurity.

The crystalline material was obtained in 0.7 to 0.9% yield from high grade commercial specimens of corn, wheat, potato and arrowroot starches. It was also isolated from soluble potato starch² which after being oxidized was dialyzed against water for four days. Preliminary experiments with xylan gave none of the substance while with cellulose the trace of apparently crystalline material observed was too minute to be identified with certainty.

This note is published because only two of us (J. H. M. and C. B. P.) intend to continue the investigation.

Experimental

Sodium periodate, 34.0 g. or 0.116 mole of $\text{Na}_3\text{H}_2\text{IO}_6$ was shaken at room temperature with 500 cc. of water and 50 cc. of glacial acetic acid until as much as possible had been dissolved. Starch, 20.25 g. air dry or 0.114 mole dry, was then added and the mixture, *pH* about 4.2, gently agitated until the next day, when titration³ showed that all of the periodate had reacted. The oxy-starch was filtered off, washed free of iodate, completely dried in a desiccator and suspended in dry methanol containing 10% of hydrogen chloride. Five hours of heating under a reflux condenser completed the degradation and after the removal of acid as the lead salt the solvent was evaporated. The viscid, levorotatory residue was dissolved in 70 cc. of 0.2 *N* aqueous caustic soda, the solution was almost saturated with sodium chloride and extracted with a total volume of 1500 cc. of diethyl ether. After drying, the extract yielded a pale yellow oil which partly crystallized when alternately cooled in dry-ice and allowed to thaw. The same crystals also separated from the high-boiling fraction of the oil (b. p. 170–180° at 3–4 mm.). Two recrystallizations from ether left the m. p. constant at 150–150.5° (corr.) and not depressed by admixture with specimens derived from other starches. The yield of pure material was 0.15 to 0.17 g. for the different starches and either sodium or potassium periodate, buffered to *pH* 4.2 with acetate or phthalate, was used in the preparation.

Anal. Calcd. for $\text{C}_{13}\text{H}_{18}\text{O}_8(\text{OCH}_3)_4$: C, 48.11; H, 6.60; OCH_3 , 29.24; mol. wt., 424. Found for two independent preparations: C, 48.21, 47.77, 47.75; H, 6.61, 6.59, 6.63; OCH_3 , 28.87, 29.09, 29.20; mol. wt. (Rast), 419, 425, 428, 432 for 3–10% solutions.

The substance was sparingly soluble in water and petroleum ether but dissolved more readily in other solvents.

(2) Baird, Haworth and Hirst, *J. Chem. Soc.*, 1201 (1935).

(3) Treadwell and Hall, "Analytical Chemistry," 8th ed., Vol. II, John Wiley and Sons, Inc., New York, p. 616.

The specific rotation at 26° in dioxane was -7.1° (sodium light, *C*, 0.911).

CONTRIBUTION No. 188

RESEARCH LABORATORY OF ORGANIC CHEMISTRY
MASSACHUSETTS INSTITUTE OF TECHNOLOGY
CAMBRIDGE, MASS.

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The Dissociation Constant of Hypobromous Acid

BY MANFRED KIESE AND A. BAIRD HASTINGS

In experiments on the catalytic effect of certain acids upon the hydration of carbon dioxide, it became necessary to know the dissociation constant of hypobromous acid. No data on this constant were found in the literature.

Hypobromous acid was prepared by allowing bromine in aqueous solution to react with mercuric oxide. The acid was purified by distillation in vacuum. Since hypobromous acid is not very stable, the experiments were carried out immediately after the preparation of the acid, in a constant temperature room, at a solution temperature of 5°.

The dissociation constant was estimated from data obtained by the titration of 0.07 *M* solutions of hypobromous acid with 0.7 *N* sodium hydroxide; *pH* was measured potentiometrically, using the glass electrode and a saturated calomel cell. The standard of reference used in the calculation of the *pH* values was "standard" acetate assuming its *pH* = 4.65.¹ Three titration curves were determined (obtaining from 15 to 20 points for each curve), and the dissociation constants calculated by application of the mass law equation. The average value of the apparent dissociation constant was found to be 1.0×10^{-9} , or expressed logarithmically $pK' = 9.0 \pm 0.06$.

It may be noted that the value of *pK* found for hypobromous acid lies between that reported for hypochlorous acid, *pK* = 7.6,² and that for hypiodous acid, *pK* = 10.6.³

(1) D. I. Hitchcock and A. C. Taylor, *THIS JOURNAL*, **59**, 1812 (1937).

(2) References in Gmelin, *Handbuch der anorg. Chemie*, Vol. VI, 1927, p. 255.

(3) F. Fürth, *Z. Elektrochem.*, **26**, 57 (1922).

DEPARTMENT OF BIOLOGICAL CHEMISTRY
HARVARD MEDICAL SCHOOL

BOSTON, MASS.

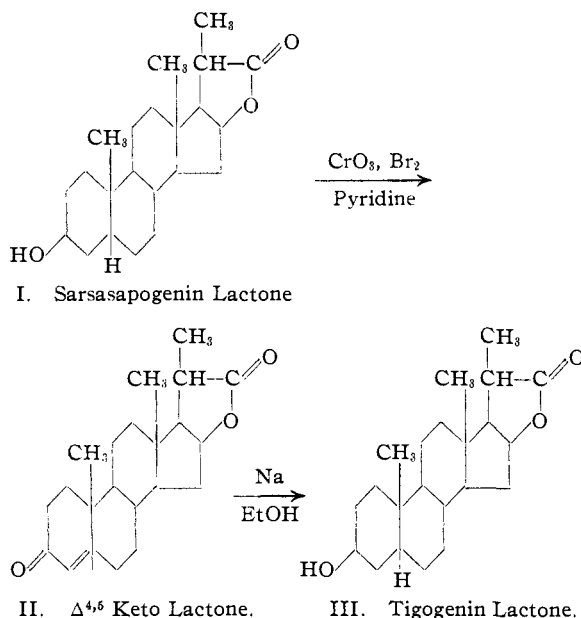
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Sterols. LXII. Position of the Hydroxyl Group in Tigogenin and Sarsasapogenin

BY RUSSELL E. MARKER AND EWALD ROHRMANN

Until the present time no substantial chemical evidence has been given which would distinguish

between positions at C-2 and C-3 for the hydroxyl group of tigogenin and sarsasapogenin. Likewise, it has not been shown that the hydroxyl group occupies the same position in both compounds. We have converted the hydroxy sarsasapogenin lactone into its *allo* form and find that this is identical with the hydroxy tigogenin lactone. This transformation indicates that the sarsasapogenin lactone differs from the tigogenin lactone only in regard to the configuration at C₅ and that the hydroxyl groups are located at C-3.



The keto lactone of sarsasapogenin first described by Farmer and Kon¹ was caused to react with one mole of bromine to give a bromo keto lactone, m. p. 194–195°. *Anal.* Calcd. for $\text{C}_{22}\text{H}_{31}\text{O}_3\text{Br}$: C, 62.4; H, 7.4. Found: C, 62.3; H, 7.5. Treatment of this compound with boiling pyridine gave an unsaturated keto lactone, m. p. 213–214°. *Anal.* Calcd. for $\text{C}_{22}\text{H}_{30}\text{O}_3$: C, 77.15; H, 8.8. Found: C, 77.1; H, 8.9. This was reduced with sodium and ethanol and the epimeric forms separated with digitonin. Decomposition of the insoluble digitonide yielded a hydroxy lactone, m. p. 234–235°. *Anal.* Calcd. for $\text{C}_{22}\text{H}_{34}\text{O}_3$: C, 76.25; H, 9.9. Found: C, 76.3; H, 10.0. This formed an acetate, m. p. 217–218°, which gave no depression with an authentic sample of tigogenin lactone acetate, m. p. 217–218°, first described by Tschesche and Hagedorn.² *Anal.* Calcd. for $\text{C}_{24}\text{H}_{36}\text{O}_4$: C, 74.15; H, 9.4. Found: C, 73.9; H, 9.3. Mild oxidation of the lactone with chromic anhydride gave a keto lactone, m. p. 252–254°. *Anal.* Calcd. for $\text{C}_{22}\text{H}_{32}\text{O}_3$: C, 76.7; H, 9.4. Found: C, 76.7; H, 9.6.

We wish to thank Parke, Davis and Company for their

(1) Farmer and Kon, *J. Chem. Soc.*, 414 (1937).

(2) Tschesche and Hagedorn, *Ber.*, 68, 1412 (1935).

generous help and assistance in the various phases of this work.

SCHOOL OF CHEMISTRY AND PHYSICS
PENNSYLVANIA STATE COLLEGE
STATE COLLEGE, PENNA.

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On the Combination of Electrophoretic and Chromatographic Adsorption Methods

BY HAROLD H. STRAIN

Resolution of mixtures of water-soluble compounds on Tswett adsorption columns¹ is facilitated by application of electrical potential to the ends of the columns. This combination of chromatographic and electrophoretic methods extends and enhances the utility of both methods for the isolation of pure substances from mixtures.

Colored substances are separated upon adsorption columns by electrophoresis in the following manner. An electrode of coiled wire covered loosely with cotton is placed in the constricted end of a glass tube and the tube is filled with the adsorbent, usually cotton, Hyflo Super Cel (heat-treated siliceous earth) or mixtures of this with talc. Before use, the adsorption column is attached to a suction flask and filled with water. A little of the mixture of charged or ionized colored substances to be separated is then drawn into the upper portion of the column where it forms a rather homogeneous colored band. The suction is released; a second electrode is placed at the top of the column and covered with water; and potential is applied to the two electrodes. This causes the pigments to migrate toward the electrodes of opposite charge and gives rise to a series of colored bands or zones each of which contains a single pigment.

Compounds separated upon adsorption columns by electrophoresis may be isolated in two ways. The bands of colored substances may be removed separately from the column and the pigments themselves eluted from the adsorbent with additional water, ethanol, alkalis or acids, or the pigments may be washed through the columns and collected separately in successive portions of the percolate.

In striking contrast to the effect obtained in the usual electrophoretic experiments where the bands of colored substances migrate with little or no widening, the materials on Tswett columns spread out rapidly until rather homogeneous colored zones are obtained. The pigments in

(1) M. Tswett, *Ber. deut. botan. Ges.*, 24, 384 (1906).