The mixture of this substance with the substance of the same m.p. but lower rotation ([α]D +33°) obtained in the PtO_2 reduction showed no depression in melting point.

The monoxime of XVIII was prepared with hydroxylamine acetate in methanol: m.p. 221–224°, λ_{max}^{alc} 300 mμ (ε 79).

Anal. Calcd. for $C_{21}H_{31}O_4N$: N, 3.88. Found: N, 4.20.

Reduction of $\Delta^{5,13(17a)}$-Etiojervadiene-3β-ol-11,17-dione Acetate (VI) with Palladium-on-charcoal in Dioxane.—$\Delta^{5,13(17a)}$-Etiojervadiene-3β-ol-11,17-dione acetate (VI) (700 mg.) was hydrogenated with pre-reduced 5% Pd-on-charcoal (500 mg.) in pure dioxane (25 ml.) until 80 ml. of hydrogen had been taken up, which required *ca.* 24 hours. The crude reduction product (660 mg.) was dissolved in benzene (5 ml.)–hexane (5 ml.) and chromatographed on alumina (13 g.). Benzene (1000 ml. in 28 fractions) eluted essentially a single substance (242 mg.), which after two crystallizations from ether–hexane melted at 135–136°, $[\alpha]^{23}D$ −12° (*c* 1.27 in $CHCl_3$), λ_{max}^{alc} 300 mμ (ε 110), λ_{max}^{Nujol} 5.81 μ (composite of saturated keto- and ester-carbonyls), 6.05 μ (isolated double bond), no OH-bands. This product appears to be Δ^{5}-13ξ,17aξ-etiojervene-3β-ol-11,17-dione acetate (XIX).

Anal. Calcd. for $C_{21}H_{28}O_4$: C, 73.22; H, 8.19. Found: C, 73.55; H, 7.94.

Elution with benzene (360 ml.) gave only difficultly separable mixtures. Benzene–ether (8:2) afforded in the first 70 ml. 75 mg. of $\Delta^{13,15,17}$-5-ξetiojervatriene-3β,17-diol-11-one 3-acetate (XXII) which after recrystallization from acetone–hexane melted at 225–230° dec., $[\alpha]^{23}D$ −210° (*c* 1.26 in $CHCl_3$); λ_{max}^{alc} 224 mμ (ε 15,500), 254 mμ (ε 10,400), 327 mμ (ε 2,050).

Anal. Calcd. for $C_{21}H_{26}O_4$: C, 73.66; H, 7.66. Found: C, 74.05; H, 7.48.

This substance was followed by amorphous material (30 mg. in 75 ml. of benzene–ether 8:2) and eventually by a crystalline substance (50 mg. in 275 ml. of the same solvent mixture). Recrystallization from acetone–hexane afforded material melting at 143–144.5°, $[\alpha]^{24}D$ −109° (*c* 1.20 in $CHCl_3$), λ_{max}^{alc} 300 mμ (ε 80); λ_{max}^{Nujol} 2.89 μ (OH), 5.77 μ (composite saturated keto and ester carbonyls), 6.05 μ (isolated double bond). This substance appears to be Δ^{5}-13ξ,17aξ-etiojervene-3β,17ξ-diol-11-one 3-acetate (XX).

Anal. Calcd. for $C_{21}H_{30}O_4$: C, 72.80; H, 8.73. Found: C, 72.59; H, 8.23.

Benzene–ether 1:1 (175 ml.) eluted a small amount of crystalline material (25 mg.), which could not be crystallized to sufficient purity.

Benzene–acetone 3:1 (125 ml.) eluted 100 mg. of crystalline material, which was readily purified by crystallization from methanol, m.p. 204–205.5°, $[\alpha]^{23}D$ −118° (*c* 1.28 in $CHCl_3$); λ_{max}^{alc} 243 mμ (ε 9000), 318 mμ (ε 45); λ_{max}^{Nujol} 2.93 μ (OH), 5.92 and 5.97 μ (conj. carbonyl); 6.15 μ (conj. double bond). This substance probably represents Δ^{13}-5ξ,17aξ-etiojervene-3β,17ξ-diol-11-one 3-acetate (XXI).

Acknowledgment.—The authors are happy to acknowledge the many informal discussions with Dr. O. Wintersteiner which have helped to evolve the concepts presented in this paper. We are indebted also to Mr. E. A. Paredes for technical assistance, to Mr. J. F. Alicino and his associates for the microanalyses, and to Dr. Nettie H. Coy and her associates for the ultraviolet and infrared spectra.

NEW BRUNSWICK, N. J.

[CONTRIBUTION FROM THE SQUIBB INSTITUTE FOR MEDICAL RESEARCH]

The Structure of Jervine. IV. The Sulfuric Acid-catalyzed Acetolysis of O,N-Diacetyljervine

BY O. WINTERSTEINER AND M. MOORE

RECEIVED JUNE 1, 1953

O,N-Diacetyljervine on acetolysis with acetic anhydride–acetic acid containing several molar equivalents of sulfuric acid yielded besides an amorphous sulfonic acid two neutral compounds melting at 240° and 194°, respectively. In their formation the oxidic ring of jervine has been opened with the establishment of an acetoxy group at position 23 in the piperidine ring. The compound m.p. 240° is assigned the indanone structure III on the basis of its ultraviolet absorption spectrum, its reducibility to a product exhibiting benzenoid absorption characteristics, the inertness of its keto group, and the fact that the N-acetate IV prepared from it by O-desacetylation on Oppenauer oxidation gave the α,β-unsaturated ketone V. The formulation of the compound m.p. 194° as VII rests mainly on the similarity of its absorption spectrum to that of jervine and the presence of a new, non-acylatable hydroxyl group. On hydrolysis with alkali VII suffers not only the loss of its two O-acetyl groups, but also a rearrangement resulting in the formation of a weak tertiary base which is best expressed as IX.

In the preceding paper of this series[1] Fried and Klingsberg have described the acetolysis of jervine by means of boiling acetic anhydride and zinc chloride. In this reaction the piperidine ring and the oxidic oxygen atom are eliminated with the formation of a C_{21}-fragment retaining only two carbon atoms of the original side chain. In contrast, the sulfuric acid-catalyzed acetolysis of acetylated jervine derivatives on which we wish to report in this and the following paper does not result in degradation. The side chain moiety including the nitrogenous ring remains intact except for the anticipated opening of the oxidic bridge linking that ring with ring D of the tetracyclic nucleus (*cf.* formulas I and II, representing, respectively, the structure proposed for jervine by Jacobs and Sato[2] in 1949, and the new structure advanced by us). However, in the case of jervine itself the reaction brings about an additional change in ring D which is highly significant in relation to the structure assigned by Fried and Klingsberg to the "yellow diketone" resulting from further degradation of their acetolysis product. The substance of these investigations already has been communicated in preliminary form.[3,4] In this paper we present in detail the work with jervine amplified by some results obtained more recently.

The reaction was carried out by dissolving O,N-diacetyljervine[5] in the acetolysis mixture (acetic

(1) J. Fried and A. Klingsberg, THIS JOURNAL, **75**, 4929 (1953).

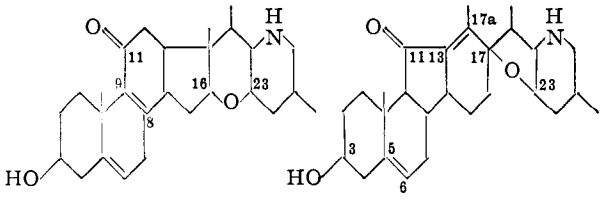
(2) W. A. Jacobs and Y. Sato, *J. Biol. Chem.*, **131**, 55 (1949).

(3) J. Fried, O. Wintersteiner, A. Klingsberg, M. Moore and B. M. Iselin, THIS JOURNAL, **73**, 2970 (1951).

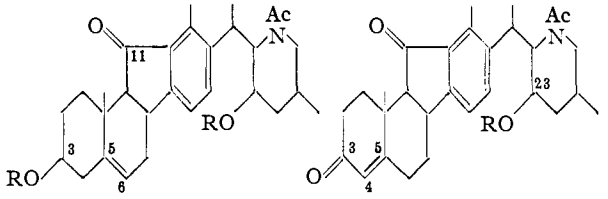
(4) O. Wintersteiner, B. M. Iselin and M. Moore, Abstracts, XIIth Internat. Congress of Chemistry, September 10–13, 1951. Medicinal Chemistry, p. 292.

(5) It was necessary to use the acetylated derivatives as starting materials for this reaction as the free alkaloidal bases combine with the sulfuric acid to form the sulfates which are practically insoluble in the acetolysis mixture.

anhydride–acetic acid 7:3 containing 0.5–1% of concentrated sulfuric acid) and allowing the intensely yellow solution to stand at room temperature for 6 hours or longer. After decomposi-

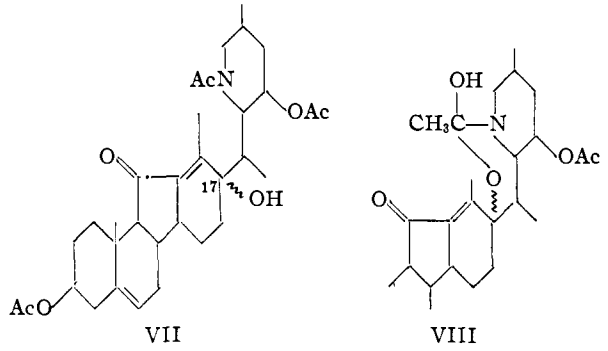


I II

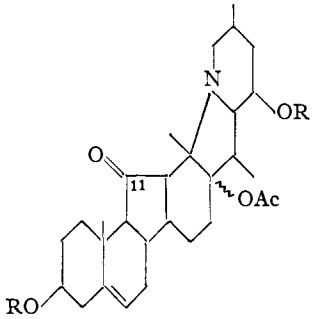


III, R = Ac
IV, R = H

V, R = H
VI, R = Ac



VII VIII



IX, R = H
X, R = Ac

tion of the acetic anhydride with ice-water and sodium bicarbonate the products were extracted into chloroform. Chromatographic fractionation of the chloroform residue yielded two neutral crystalline compounds (III and VII), and an amorphous substance which proved to be the sodium salt of a sulfonic acid.

Indanone III.—The structurally most significant of these products is a neutral compound, m.p. 239–240°, $[\alpha]^{23}D$ − 29°, on which we have already reported in our preliminary communications.[3,4] It was assigned the indanone structure III on the basis of the following facts: Its elementary composition $C_{33}H_{43}O_6N$ (diacetyljervine + C_2H_2O − 2H) and its acetyl content indicated that the oxidic ring of jervine had been opened with the formation of a new acetoxy group. Its ultraviolet absorption spectrum showed two maxima (λ_{max}^{alc} 251 mμ, ε 12,200 and 300 mμ, ε 2200) such as are characteristic for 1-indanones and 1-tetralones. Catalytic hydrogenation with platinum oxide in acetic acid proceeded with the consumption of 2 moles of hydrogen, of which the second mole was taken up sluggishly. The resulting amorphous product (probably a mixture of stereoisomeric indanols) exhibited benzenoid ultraviolet absorption (λ_{max}^{alc} 268 mμ, ε 500) similar to that of neoergosterol and veratramine.[6] Treatment of the reduction with chromic acid reestablished the original spectrum. Since the ketonic carbonyl is the only functional group present which could be involved in this reversible change, it followed that this group was adjacent to a six-membered ring which had been aromatized in the acetolysis reaction and further, from prior knowledge as to the general location of the oxidic bridge, that this ring was ring D, and that the ketonic group consequently occupied the adjoining position in ring C. The postulate that the latter ring is five-membered has its basis in the fact that the acetolysis product is completely unreactive to ketone reagents, as is jervine itself. This should not be the case if the carbonyl were part of a 1-tetralone grouping representing rings C and D of the alternative perhydrochrysene structure.

The properties of III so far discussed are thus quite in line with those of the phenolic monoketone formed under alkaline conditions from the "yellow diketone" of Fried and Klingsberg, and provide independent support for the structures assigned to the products of that series. Conversely, for placing the methyl group and the side chain in III we had to lean on some of the facts ascertained by these authors (ultraviolet characteristics of "yellow diketone," and 1,4-relationship of the keto groups in this compound). The new acetoxy group was assigned position 23 in the piperidine ring on the strength of the demonstration by Jacobs and Sato,[2] that the base $C_8H_{11}ON$ produced in the selenium dehydrogenation of jervine[7] is in all probability 3-methyl-5-hydroxy-6-ethylpyridine.

Since the composition $C_{33}H_{43}O_6N$ indicated by the elementary analysis differs by two hydrogen atoms less from that corresponding to the addition of the elements of ketene to diacetyljervine, it seemed that the third double bond necessary for the completion of the aromatic nucleus had been acquired by a dehydrogenation reaction rather than by migration of the 5,6-double bond into ring D. In order to gain clarity on this point, the N-acetate IV (m.p. 227–229°, $[\alpha]^{29}D$ − 31.5°) was prepared from III by hydrolysis with alkali and subjected to Oppenauer oxidation. The α,β-unsaturated ketone nature of the resulting product (V, m.p. 275–280°, $[\alpha]^{23}D$ + 126°) was evidenced by its ultraviolet absorption characteristics (Fig. 1, curve 2) which clearly represent the combined absorptions of the indanone and unsaturated ketone chromophores. The abnormally high positive molecular rotation change accompanying this reaction (+733°) ac-

(6) W. A. Jacobs and L. C. Craig, *J. Biol. Chem.*, **160**, 155 (1945).
(7) W. A. Jacobs, L. C. Craig and G. I. Lavin, *ibid.*, **141**, 51 (1941).

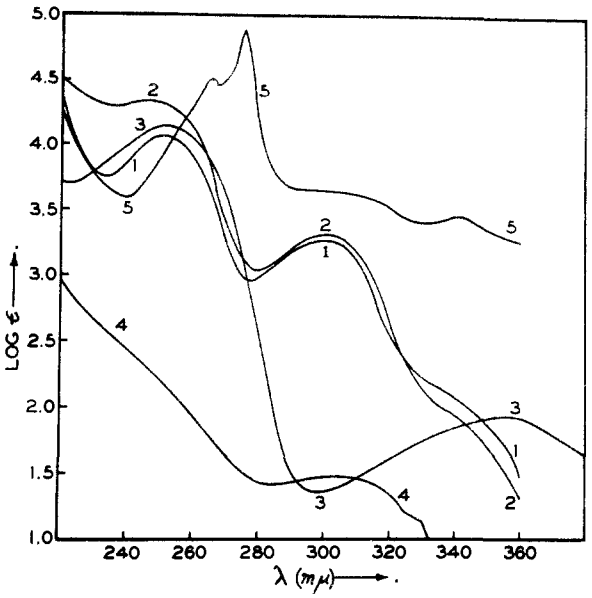


Fig. 1.—Ultraviolet absorption spectra (abs. ethanol): curve 1, III; curve 2, V; curve 3, VII; curve 4, IX; curve 5; sulfonic acid (Na salt) from acetolysis.

cords well with the similarly high values observed in the oxidation of jervine (+742°) and of the 3-hydroxy-11,17-diketone $C_{19}H_{24}O_3$ of Fried and Klingsberg[1] (+684°) to the corresponding Δ^4-3-ketones. That the 23-hydroxyl group had remained unoxidized followed from the presence in the infrared spectrum of a band at 2.98 μ, and from the formation of a *mono*oxime (m.p. 248–251°) and of an O,N-diacetate (VI, m.p. 230–232°, $[\alpha]^{23}D$ + 128°) on acetylation. Significantly, the preformed hydroxyl group in the piperidine ring of veratramine exhibits similar resistance to oxidation by aluminum *t*-butylate and acetone,[8,9] and since this group has been conclusively shown to occupy the 23-position,[8] the above result supports the assignment of this position to the new acetoxy group in III.[10]

The mechanism by which the additional double bond needed for the completion of the benzenoid ring arises must remain a matter of conjecture. Autoxidation of the dihydrobenzene derivative which may be assumed to be the primary acetolysis product is not likely to occur under the acidic conditions prevailing during the reaction. That it does not occur during the work-up is evidenced by the fact that in one experiment the indanone could be isolated in small yield directly from the reaction mixture when the latter was concentrated *in vacuo*. A more likely event would be disproportionation of the hypothetical dihydrobenzene precursor. Finally there is the possibility that the aromatic ring is completed by the elimination of sulfurous acid from the sulfonic acid mentioned earlier, provided that the latter possesses the appropriate structure, namely, that of $\Delta^{13(17a),16}$-diene-11-one-15-sulfonic acid. Elimination reactions of this kind resulting in aromatization are known to occur under conditions somewhat similar to those used here, a case in point being the formation of 3,4,5-trimethylphenol from 3,3,5-trimethylcyclohex-2-enone-4-sulfonic acid under the action of boiling acetic anhydride.[11] However, attempts to convert the sulfonic acid or its sodium salt to the indanone III by prolonged heating with acetic anhydride or by pyrolysis were unsuccessful.

The other neutral acetolysis product (m.p. 193–194°, $[\alpha]^{22}D$ − 113°) had the composition $C_{33}H_{47}O_7N$ (diacetyljervine + CH_3COOH) and exhibited an ultraviolet absorption spectrum very similar to that of jervine (λ_{max}^{alc} 252 mμ, ϵ 14,000; 355 mμ, ϵ 85). The infrared spectrum revealed the presence of hydroxyl, O-acetyl, ketonic carbonyl, conjugated C=C, and N-acetyl (bands at 2.96, 5.77, 5.86, 6.14 and 6.26 μ). A reasonable structure compatible with these data would be VII, except perhaps insofar as one would expect the oxidic ring to be opened with the formation of a tertiary acetoxy instead of a hydroxyl group at C_{17}. On the other hand, it is conceivable that after the addition of a proton derived from the sulfuric acid to the oxygen atom the bond to C_{23} is broken, so that this carbon atom acquires an acetoxy group from the reagent, while the hydroxyl group at C_{17} resulting from the equalization of charges, since it is tertiary, resists subsequent acetylation. Alternatively the ortho ester-amide structure VIII could be considered, which would afford an explanation for the fact that all three acetyl groups were liberated in the Kuhn–Roth acetyl determination (intermediary formation of 17-O-acetate), whereas in our experience fully or N-acetylated derivatives of jervine and veratramine generally yield but a fraction of the N-acetyl group in this procedure. However, this formulation fails to account for the infrared band at 6.26 μ, provided that this band is correctly interpreted as representing an N-acetyl group.[12]

More puzzling than this anomaly was the behavior of the triacetate on hydrolysis with methanolic potassium hydroxide either at room or reflux temperature. Comparable derivatives of jervine (and veratramine) merely suffer O-desacetylation when so treated, and hence yield the corresponding N-acetates. The hydrolysis product obtained from VII (m.p. 243–251.5°, $[\alpha]^{23}D$ − 133°) showed indeed the composition $C_{29}H_{43}O_5N$ of the expected N-acetate. The fact that the remaining acetyl group was fully determinable by the Kuhn–Roth procedure was not surprising in view of the behavior of the parent triacetate. However, it became immediately clear from the ultraviolet and

(8) W. A. Jacobs and Y. Sato, *J. Biol. Chem.*, **191**, 77 (1951).

(9) Ch. Tamm and O. Wintersteiner, THIS JOURNAL, **74**, 3842 (1952).

(10) The indanone III has meanwhile been shown to be triacetyl-11-ketoveratramine by the identification of a ketonic product obtained in the chromic acid oxidation of triacetyldihydroveratramine as the 5,6-dihydroderivative of III (O. Wintersteiner and H. Hosansky, THIS JOURNAL, **74**, 4474 (1952)). Furthermore, the degradation of veratramine to benzene-1,2,3,4-tetracarboxylic acid has made secure the assignment in all these compounds of position 17a to the methyl group representing carbon atom 18 (O. Wintersteiner, M. Moore and N. Hosansky, THIS JOURNAL, **75**, 2781 (1953)).

(11) W. von E. Doering and F. M. Beringer, *ibid.*, **71**, 2221 (1949); *cf.* also M. Beringer and I. Kuntz, *ibid.*, **73**, 364 (1951).

(12) We have measured the infrared spectrum of a variety of N-acetylated derivatives of jervine and veratramine, and find that the N-acetyl band may be located anywhere between 6.06 and 6.26 μ. We were unable to correlate the position of this band with constitutional factors.

infrared data that this compound was not simply O-desacetylated VII, and further study showed it to be an isomer which owes its formation to a rearrangement hitherto not encountered in the jervine series. The ultraviolet spectrum was identical with that of dihydrojervine (λ_{max} 306 mμ, ϵ 29, and end absorption, Fig. 1, curve 4) indicating disappearance of the conjugated 13,17a-double bond. Of the four bands in the double bond region of the infrared spectrum of VII, only that at 5.77 μ had survived. This band has to be interpreted as a composite band originating in the now unconjugated ketonic group in the five-membered ring C (dihydrojervine, 5.80 μ) and in the acetoxy group, which, as will be shown presently, accounts for the remaining, analytically demonstrable acetyl. The absence of the N-acetyl band made the hydrolysis product suspect of being a base, and this was indeed found to be the case. Its basic group was titrable with perchloric acid in acetic acid solution, while the parent triacetate VII, as expected, showed no consumption of this reagent. It was then clear that the treatment with cold methanolic potassium hydroxide had effected, contrary to all previous experience, the removal of the N-acetyl group, and that consequently the remaining acetyl group was bound to oxygen. The basic group liberated in this facile manner differed in essential respects from that of jervine. In the first place, as will be shown below, the hydrolysis product is not a secondary but a tertiary amine. Moreover, it is a much weaker base than N-methyljervine or the naturally occurring tertiary veratrum alkaloids such as rubijervine, germine, *etc.* Unlike the latter, the monoacetate was insoluble in 10% acetic acid and did not form stable salts with hydrochloric acid or perchloric acid, though it could be brought into aqueous solution with excess mineral acid and therefrom precipitated with base reagents such as picric acid, phosphotungstic acid and Mayer reagent.

On acetylation with acetic anhydride and pyridine the monoacetate m.p. 252.5° yielded a triacetate, m.p. 165–167°, $[\alpha]^{22}D$ −134°, which had the composition $C_{33}H_{47}O_7N$ and hence was isomeric with the acetolysis product VII. The three acetyl groups were readily demonstrable analytically. However, the isomeric triacetate likewise behaved as a weak base (perchloric acid titration), showing that the basic group was not acetylable and hence tertiary, and this was borne out by the absence in the infrared spectrum of bands in the 3 μ region referable to >NH or -OH. The double bond region was devoid of any bands except that at 5.77 μ; the ultraviolet spectrum was identical with that of the monoacetate. As with the latter, no stable salts could be prepared. Treatment with methyl iodide yielded an amorphous product which, however, reverted to the crystalline tertiary base on manipulation in organic solvents.

As regards the structural interpretation of these observations, it should be noted first of all that the acetolysis product VII is the only "open" derivative of jervine so far known in which the original α,β-unsaturated ketone grouping is still present, in other words, in which interaction between this group and the nitrogen atom in the piperidine ring is possible, since due to the opening of the oxidic bridge the latter ring is now free to approach sterically favored sites in the tetracyclic nucleus. We assume that actually two such sites become involved when VII is exposed to alkali: First the N-acetyl group is transposed to the tertiary hydroxyl at C_{17} by N → O migration, possibly *via* the intermediate cyclic ortho-ester-amide VIII. The free imino group then adds to the nuclear double bond in the 17a-position, that is β to the ketonic carbonyl. The end product of this sequence (which of course includes the simultaneous saponification of the two secondary acetoxy groups at C_3 and C_{23}) is the tertiary base IX, yielding on acetylation the O-triacetate X. This formulation also provides an explanation for the weakly basic character of the rearranged products in that the methyl group at C_{17a} may be assumed to exert a strong shielding effect on the basic group, impeding the approach of protons to the latter. The tertiary 17-acetoxy group in IX conforms to the requirement that the saponification product of VII contain O-bound acetyl not removable by alkali. In the sense that IX is the only expression which logically accounts for all the observed facts, it also constitutes proof for the attachment at C_{17} of the oxidic bridge in our formulation of jervine (II), a concept hitherto supported by circumstantial evidence only.[1]

In subsequently studying the perchloric acid-catalyzed acetolysis of diacetyljervine we have discovered a more convenient route than the one described above for preparing the isomerized triacetate X. The main product of this reaction is a crystalline perchlorate (m.p. 220–222°, $[\alpha]^{23}D$ − 43°) of what seems to be a secondary base related to VII. This salt shows the remarkable property of instantaneously rearranging to X merely on treatment with cold aqueous methanolic sodium carbonate or with pyridine. Its ultraviolet spectrum resembled, but was not identical with, that of jervine, the main maximum having undergone a shift from 251 to 243 mμ while the low intensity peak appeared at 365 mμ instead of at 360 mμ. The analyses fitted best for the perchlorate of a base $C_{33}H_{45}O_6N$ (diacetyljervine + C_2H_2O), but this composition is difficult to accept for the following reasons: The salt character of the compound would seem to preclude the presence of acetyl on nitrogen; therefore, the three acetyl groups revealed by the analysis and by the facile conversion to the triacetate X should be all O-bonded, and this requires the composition $C_{33}H_{47}O_7N$ (that of X) for the basic component. Since, on account of its spectral properties, the latter cannot be X itself, a secondary base of type VII but differing from it by having the third acetyl group attached to oxygen at C_{17} instead of to the basic group, would seem to be indicated. The infrared spectrum contained bands at 2.98, 5.76, 5.88, 6.10 and 8.04 μ. The band at 6.10 μ was of medium intensity and probably corresponds to that at 6.14 μ in the spectrum of VII (C=C conjugated to CO), while that at 2.98 μ must be interpreted as arising in the NH group, since it was

also exhibited by piperidine perchlorate (but not by potassium perchlorate). The only observation (aside from the unsatisfactory analysis) which was difficult to reconcile with the suggested structure was that the perchlorate, unlike VII and X, yielded only little more than two of its three acetyl groups in the Kuhn–Roth acetyl determination. Attempts to replace the perchloric acid in the salt by other strong acids, and to demonstrate the presence of a secondary amino group by the preparation of N-acetyl and N-nitroso derivatives led either to the recovery of unchanged starting material or to amorphous, unidentifiable products.

The sulfonic acid, which under the conditions used is the main reaction product of the acetolysis reaction, passes after the neutralization step into the chloroform extract as the sodium salt, and is eluted in this form from the chromatographic column with methanol. It was obtained as a yellow amorphous powder which could not be crystallized either as such or after conversion to the free acid. The analyses were inconclusive, and revealed little more than that one sulfonic acid group had entered the molecule. However, from the demonstration (*vide infra*) that VII is an intermediate in the formation of the acid it is clear that the latter must be likewise an "open" derivative possessing the additional 23-acetoxy group. The most characteristic and least variable property of these amorphous products was their ultraviolet absorption spectrum (Fig. 1, curve 5) which showed an intense narrow band at 275 mμ, ε 56,500,[13] and subsidiary maxima at 265 mμ, ε 32,600, and 340 mμ, ε 2900. We have found no data in the literature which would provide a clue to the chromophore giving rise to this type of spectrum, nor does the latter seem to fit the structures which suggest themselves on chemical grounds (indanone sulfonic acid, $\Delta^{13(17a),16}$-diene-11-one-15-sulfonic acid). The acid was not further investigated.

In order to gain some insight into the mode and order of formation of the three acetolysis products, each of them was subjected to treatment with the acetolysis mixture. The indanone III was recovered unchanged, as expected. On the other hand, the triacetate VII gave a small amount of III and for the rest was converted to the sulfonic acid. It must therefore be regarded as the primary acetolysis product, a fact which accords well with the assigned structure. The sulfonic acid did not appear to undergo any marked changes on further exposure to the acetolysis conditions. Prolonged refluxing with acetic anhydride resulted in the obliteration of the high maximum at 275 mμ, and the emergence of a much lower and less well-defined peak at 258 mμ, but on the whole the spectrum gave no indication that III had been formed, nor could this compound be isolated from the chromatographed mixture.

Experimental

The melting points were taken in the open capillary and are corrected for stem exposure. The rotation measurements were taken on chloroform solution of the substance in a 1-dm. semi-micro tube. The ultraviolet spectra were measured in a quartz Beckman spectrophotometer, Model DU, and those of the more important compounds were later checked in a Cary self-recording instrument. The infrared spectra were determined in nujol suspensions in a Perkin–Elmer, Model 12-B, spectrophotometer. The analytical samples were dried over phosphorus pentoxide at 110° (1 mm.). The alumina used for chromatography (Harshaw) was washed with dilute sulfuric acid and water to *p*H 4.5 and reactivated by heating at 150° for 48 hours.

Acetolysis of Diacetyljervine.—Diacetyljervine (2.04 g.) was dissolved in a mixture of acetic anhydride (70 cc.), acetic acid (30 cc.) and concentrated sulfuric acid (1.0 cc.). The solution, which soon turned yellow and finally a deep orange, was allowed to stand at room temperature for 6 hours. It was then concentrated *in vacuo* (bath temp. 35°) to about 20 cc., and poured onto crushed ice and water. Solid sodium bicarbonate was added in portions with mechanical stirring till the reaction was alkaline. The resulting oily product was extracted into chloroform (2 × 300 cc.); the chloroform extract was washed with water, dried and freed from solvent *in vacuo*. The orange-colored, glassy residue (1.92 g.) was dissolved in benzene (25 cc.) and chromatographed on a column of sulfuric acid-washed alumina (32 × 80 mm.). Elution was effected with 200-cc. portions of benzene (fractions 1–23), benzene–ether 2:1 (fractions 24–33), ether (fractions 34–37), and methanol (fractions 38–44). Fractions 1–13, together 312 mg., and 24–35, together 185 mg., were crystalline. The former on recrystallization afforded the indanone III, and the latter its precursor VII (see below). The combined methanol eluates, 1.11 g., were lyophilized from water, yielding a yellow powder which consisted essentially of the sodium salt of the sulfonic acid.

Slightly better yields of the two neutral products could be obtained by omitting the concentration of the acetolysis mixture prior to treating it with water and bicarbonate. It was also found that most of the sulfonic acid could be removed from the chloroform residue by dissolving the latter in ethanol, adding water and sulfuric acid and extracting the acid solution with ether. The residue of the ether solution consisted essentially of a crystalline mixture of the two neutral acetolysis products, but their separation by fractional crystallization is not profitable, so that the chromatographic step cannot be dispensed with.

22,26-Imino-5,13,15,17(17a)-jervatetraene-3(β),23-diol-11-one Triacetate (III).[14]—The crystalline fractions eluted with benzene were combined and recrystallized three times from 1:2 ethyl acetate–hexane, yielding small, slightly yellow needles melting at 238–240° after softening at 232°, with $[\alpha]^{24}D$ −27 ± 1° (*c* 0.925).

Anal. Calcd. for $C_{33}H_{43}O_6N$ (549.7): C, 72.10; H, 7.89; 3 $COCH_3$, 23.5. Found: C, 71.82; H, 7.98; $COCH_3$, 17.0.

In order to remove the yellow pigment adsorbed on the crystals it was found necessary to saponify the two acetoxy groups and to reacetylate the resulting N-acetate IV (see below). The compound then melted at 239–240° and showed $[\alpha]^{23}D$ −28.7 ± 1° (*c* 0.900); ultraviolet spectrum, Fig. 1, curve 1; infrared spectrum, 5.75, 5.88, 6.09, 6.34, 7.88 μ. Found: C, 72.20; H, 8.21.

Reduction.—A solution of III (77 mg.) in acetic acid (3 cc.) was shaken with hydrogen at atmospheric pressure in the presence of platinum oxide catalyst (40 mg.). While the first mole was consumed in 45 minutes, it required 5 more hours till the reaction came to a standstill with the uptake of somewhat less than an additional mole. The ultraviolet spectrum of the amorphous reduction product showed that it still contained 18% of unchanged indanone. Chromatography in hexane–benzene 3:1 on alumina effected removal of the unreduced portion in the early benzene eluates. The remainder of the material was recovered in two main fractions (A, eluted with benzene, and B, eluted with benzene–ether 9:1). Both were amorphous, highly dextrorotatory ($[\alpha]D$ + 85° and +99°) and exhibited benzenoid ultraviolet absorption (λ_{max}^{alc} 268 mμ, ε 530 and 420, respectively, calculated on molecular weight of 5,6-dihydroindanol corresponding to III). A portion of fraction B was lyophilized from benzene for the analysis.

Anal. Calcd. for $C_{33}H_{47}O_6$ (553.7): C, 71.58; H, 8.56; $COCH_3$, 23.4. Found: C, 70.70; H, 8.60; $COCH_3$, 16.4.

(13) Calculated on mol. wt. 653, which is that of III + HSO_3Na; the ε values are those of curve 5 in Fig. 1; other preparations gave lower figures (ε 43,000, 27,000 and 2350, respectively).

(14) For nomenclature *cf.* references 29 in foregoing paper by Fried and Klingsberg, III of this series.

Acetylation of the remainder of fraction B failed to yield crystalline material. The acetylated product was hydrolyzed in the usual manner and then subjected to a mild oxidation with chromium trioxide (3 atom equivalents of oxygen). The ultraviolet characteristics of the reoxidized material were those of III, (λ_{max}^{alc} 253 mμ, $E_{1\,cm}^{1\%}$ 204; 300 mμ, $E_{1\,cm}^{1\%}$ 37).

22,26-Imino-5,13,15,17(17a)-jervatetraene-3(β),23-diol-11-one N-Acetate (IV).—To a solution of the triacetate III (339 mg.) in methanol (8 cc.) 10% methanolic potassium hydroxide (8 cc.) was added. The orange colored solution, which soon turned green, was allowed to stand for 24 hours at room temperature, and then worked up in the usual manner (chloroform extraction). The crude product, a greenish resin, was freed from the pigment by passing its solution in benzene-ether 1:1 (25 cc.) through a short column of alumina (5 g.). The residue from the combined filtrate and washings, on recrystallization from ethyl acetate–hexane, afforded 187 mg. of small colorless needles melting at 226–228°, $[\alpha]^{23}D$ −31.9° (*c* 0.954). The ultraviolet spectrum was identical with that of III.

Anal. Calcd. for $C_{29}H_{39}O_4N$ (465.5): C, 74.80; H, 8.44; $COCH_3$, 9.20. Found: C, 74.69; H, 8.84; $COCH_3$, 4.3.

22,26-Imino-4,13,15,17(17a)-jervatetraene-23-ol-3,11-dione N-Acetate (V).—To a solution of the N-acetate IV (123 mg.) in dry acetone (3.0 cc.) aluminum *t*-butoxide (507 mg.) dissolved in dry benzene (6 cc.) was added. The turbid solution was boiled under reflux for 22 hours. After cooling, 1 *N* sulfuric acid (about 5 cc.) was added slowly till the precipitate was dissolved. After withdrawal of the benzene phase the acid solution was extracted with several portions of benzene, and the combined extracts were washed with sodium bicarbonate solution and water, dried and evaporated. The residue, together with Girard reagent T (125 mg.), was dissolved in absolute ethanol (2.0 cc.) and acetic acid (0.2 cc.), and the solution was boiled under reflux for one hour, whereupon the separation into ketonic and non-ketonic fractions was carried out in the usual way. The non-ketonic fraction (51 mg.) consisted largely of starting product. The ketonic material (48 mg.) was recrystallized three times from ethyl acetate, from which it formed small, elongated plates melting at 250–253°. Further recrystallization from dilute ethanol raised the melting point to 268–271°. The analytical sample was dried at 110° (0.1 mm.) to constant weight (loss 0.3%) and then melted at 275–280°, $[\alpha]^{23}D$ +126 ± 2° (*c* 0.462); ultraviolet spectrum, Fig. 1, curve 2: λ_{max}^{alc} 245 mμ (20,700), 300 mμ (2080); infrared spectrum, 2.98, 5.88, 6.04, 6.23 μ.

Anal. Calcd. for $C_{29}H_{37}O_4N$ (463.6): C, 75.13; H, 8.05. Found: C, 75.38; H, 8.17.

The oxime was prepared in the usual manner with excess ethanolic hydroxylamine acetate (room temperature, 3 days). Recrystallized from aqueous ethanol it formed small needles which melted, after drying at 210° (0.1 mm.), at 248–251.5° dec.

Anal. Calcd. for $C_{29}H_{38}O_4N_2$ (478.6): N, 5.85. Found: N, 5.74.

The 23,N-diacetate VI, obtained from IV with acetic anhydrine–pyridine, was recrystallized from ether and melted at 230–232°, $[\alpha]^{23}D$ +128 ± 2° (*c* 0.738).

Anal. Calcd. for $C_{31}H_{39}O_5N$ (505.6): C, 73.64; H, 7.78. Found: C, 74.07; H, 7.96.

22,26-Imino-5,13(17a)-jervadiene-3(β),17,23-triol-11-one 3,23,N-Triacetate (VII).—The crystalline benzene–ether 9:1 eluates (fraction 24–35) from the chromatographed acetolysis products were combined (185 mg.) and recrystallized repeatedly from aqueous ethanol. While after the first two recrystallizations the melting point was unsharp and remained in the range 140–155°, it subsequently rose to 190.5–193° and finally became constant at 193–194°, though softening in the former range could still be observed. (The corresponding fractions from other acetolysis experiments behaved in the same manner on purification). Only by drying of the pure material at 110° (1 mm.) to constant weight (weight loss 2.9%) could the preliminary softening in the low range be eliminated: $[\alpha]^{22}D$ −113 ± 1° (*c* 0.864): ultraviolet spectrum, Fig. 1, curve 3; infrared spectrum, 2.96, 5.86, 6.14, 6.26, 7.94 μ.

Anal. Calcd. for $C_{33}H_{47}O_7N$ (569.7): C, 69.57; H, 8.32; 3 $COCH_3$, 22.7. Found: C, 69.77; H, 8.46; $COCH_3$, 21.8.

The compound consumed no perchloric acid on titration with this reagent in acetic acid. It was recovered unchanged on acetylation with acetic anhydride and pyridine. Hydrogenation in ethanol with platinum oxide as the catalyst proceeded very slowly and stopped after the uptake of only half the calculated amount for the reduction of one double bond. Addition of new catalyst and of small amounts of potassium hydroxide[15] brought the consumption of hydrogen to near one mole per mole of substance, but the product was a mixture which on chromatographic fractionation yielded only starting material as the sole crystallizable product.

Hydrolysis of Triacetate VII. 17a?,22,26-Nitrilo-jerv-5-ene-3(β),17,23-triol-11-one 17-acetate (IX).—A solution of the triacetate VII (104 mg.) in 5% methanolic potassium hydroxide (4 cc.) was allowed to stand at room temperature for 20 hours, and then after addition of water extracted with several portions of chloroform. The combined chloroform extracts (3 × 25 cc.) were washed once with 1 *N* hydrochloric acid (30 cc.) and water, dried and evaporated. The residue (74 mg.) was dissolved in a few drops of acetone; addition of ether and then pentane resulted in the formation of small, diamond-shaped plates; m.p. 219–223°. The product was recrystallized once in the same manner, and then twice from 50% aqueous ethanol, when the melting point became constant at 243–250° (m.p. sample dried at 110° (0.1 mm.) for 3 hours), $[\alpha]^{23}D$ −133 ± 1° (*c* 0.989). (It should be mentioned, however, that in preparations from other runs melting point constancy was reached at somewhat lower temperatures (238–245°) although their $[\alpha]D$'s checked closely with the above value; chromatographic treatment failed to raise or sharpen the melting point.); ultraviolet spectrum, Fig. 1, curve 4; infrared spectrum, 2.86, 5.77, 8.00 μ.

Anal. Calcd. for $C_{29}H_{43}O_5N$ (485.6): C, 71.71; H, 8.93; N, 2.89; 1 $COCH_3$, 8.89. Found: C, 71.79; H, 8.91; N (perchloric acid titration), 2.93; $COCH_3$, 8.5.

The monoacetate was insoluble in 10% acetic acid, but soluble in glacial acetic and in 1 *N* hydrochloric acid. From the latter solution it was precipitated by picric acid, phosphotungstic acid and Mayer reagent. An attempt to prepare the hydrochloride by bringing a solution of the base in 2 *N* hydrochloric acid to dryness and removing the excess acid by repeated distillation with water resulted in the recovery of the crystalline base.

The hydrochloric acid washings of the original chloroform solution contained some basic material (29 mg.) which was recovered by alkalinization and chloroform extraction. This product could eventually be obtained in crystalline form, but as it proved to be difficult to purify, its examination has been deferred.

Acetylation of Monoacetate IX. 17a,22,26-nitrilo-jerv-5-ene-3(β),17-23-triol-11-ketone 3,17,23-Triacetate (X).—A solution of the monoacetate IX (14.5 mg.) in pyridine (0.5 cc.) and acetic anhydride (0.5 cc.) was allowed to stand at room temperature for 18 hours. The mixture was worked up in the usual way. The product (18.5 mg.) was recrystallized from aqueous ethanol and then from aqueous acetone, from which it formed rosettes of triangular plates melting at 166–167.5°, $[\alpha]^{22}D$ −134 ± 2° (*c* 0.749); ultraviolet spectrum, λ_{max}^{alc} 306 mμ, ε 29, end absorption below 280 mμ; infrared spectrum, 5.77, 7.97 μ.

Anal. Calcd. for $C_{33}H_{47}O_7N$ (569.7): C, 69.57; H, 8.32; N, 2.46; 3 $COCH_3$, 22.7. Found: C, 69.46; H, 8.27; N (perchloric acid titration), 2.44; $COCH_3$, 22.8.

Preparations recrystallized from aqueous ethanol showed the same or a slightly lower melting point (164–166°) and retained 1/2 mole of ethanol even on drying at 110° *in vacuo* as was shown by ethoxyl analysis (see below under perchlorate).

In its solubility properties and behavior toward base precipitants X behaved like the monoacetate IX. It was re-

(15) According to R. Angliker, H. Heusser and O. Jeger (*Helv. Chim. Acta*, **35**, 838 (1952)), the catalytic reduction of jervine to (13, 17a)-dihydrojervine with PtO_2 in ethanol proceeds more readily and affords better yields when carried out in the presence of about 4 molar equivalents of potassium hydroxide. In the present case an excess of alkali could not be employed, since it was ascertained by ultraviolet spectrum measurements that under these conditions an appreciable amount of VII underwent hydrolysis and rearrangement to the tertiary base IX within 24 hours.

covered unchanged when it was recrystallized from aqueous ethanol in the presence of a slight excess of perchloric acid.

In an attempt to prepare the quaternary methyl iodide the triacetate (50 mg.) was treated with 50% methanolic methyl iodide (2 cc.) for 4 days at room temperature. The resulting resinous, yellow product was taken into a few drops of methanol. Addition of ethyl acetate caused the precipitation of a gelatinous product (20 mg.), which was not unchanged X, as the latter is readily soluble in this solvent. However, on manipulation with various warm solvent mixtures it changed its solubility properties, and a portion (8 mg.) could eventually be obtained in crystalline form. The melting point (160–167°) and the absence of iodine identified the crystals as starting product X. Likewise, the material remaining in the orginal ethyl acetate mother liquor, which was readily crystallizable, turned out to be unchanged X.

Since it was conceivable that IX or X might revert to VII under acidic conditions, X was treated with the acetolyzing mixture initially used, but was recovered unchanged. Likewise, prolonged standing in methanol saturated with hydrogen chloride gas produced no change in the ultraviolet spectrum. However, refluxing in 3.5% aqueous ethanolic hydrochloric acid for 5 hours led to the emergence of a high peak at 245 mμ (ε 6500) with a shoulder at 317 mμ (ε 425). This observation will be followed up preparatively.

22,26-Imino-3(β),17,23-triacetoxy-5,13(17a)-jervadiene-11-one (?) Perchlorate.—A solution of diacetyljervine (2.15 g.) in a mixture of acetic anhydride (7.5 cc.), acetic acid (7.5 cc.) and 70% perchloric acid (1.209 g., 2 molar equivalents) was allowed to stand at room temperature for 2.75 hours and then poured onto crushed ice. The resulting precipitate was collected after the ice had melted, thoroughly washed with ice-water, and dried (2.61 g.). Recrystallization from absolute ethanol afforded 1.33 g. of needles melting at 220–222°, $[\alpha]^{23}D$ −42.6 ± 1° (*c* 0.927). The weight loss on drying the analytical sample at 110° (1 mm.) for 3 hours was 0.28%.

Anal. Calcd. for $C_{33}H_{45}O_6N\cdot HClO_4$ (652.2): C, 60.77; H, 7.11; N, 2.15; 3 $COCH_3$, 19.8; 2 $COCH_3$, 12.9; $HClO_4$, 15.4; for $C_{33}H_{47}O_7N\cdot HClO_4$ (670.2): C, 59.14; H, 7.22; N, 2.09; 3 $COCH_3$, 19.3; 2 $COCH_3$, 12.53; $HClO_4$, 15.0. Found: C, 61.02, 60.91; H, 7.08, 7.12; N, 1.97; $COCH_3$, 13.9, 14.6; $HClO_4$ (titration with $NaOCH_3$ in anhydrous methanol–benzene), 14.6; ultraviolet spectrum, λ_{max}^{alc} 243 mμ, ε 16,400; 365 mμ, ε 74; infrared spectrum, 2.98, 5.76, 5.88, 6.10, 8.04 μ.

The perchlorate was also obtained by treating the acetolysis product VII (36 mg.) with 70% perchloric acid (21.4 mg.) in acetic anhydride–acetic acid 1:1 (1.0 cc.) for 2.5 hours. The identity of the recrystallized product (26 mg., m.p. 219–220°, $[\alpha]^{22}D$ −39.8°) with the salt obtained directly from diacetyljervine was confirmed by the ultraviolet data.

The conversion of the perchlorate to the rearranged triacetate X was effected in the following manner: To a solution of the salt (301 mg.) in methanol (30 cc. slight warming) 2 *N* sodium carbonate (35 cc.) was added gradually with mechanical stirring. After the addition of water (30 cc.) the mixture was extracted with several portions of ether. The residue of the water washed and dried ether solution was dissolved in benzene–hexane 1:1 and chromatographed on alumina (16 × 44 mm.). Elution with the same solvent mixture yielded crystalline material (182 mg.) which after recrystallization from absolute ethanol melted at 165–167° after softening at 162° ($[\alpha]^{23}D$ −135 ± 1° (*c* 0.902)). The analytical sample on drying to constant weight (9 hours) at 110° (1 mm.) showed a weight loss of 2.0%.

Anal. Calcd. for $C_{33}H_{47}O_7N\cdot{}^1/_2C_2H_5OH$ (592.7): C, 68.88; H, 8.50; 3 $COCH_3$, 21.8; $^1/_2$ OC_2H_5, 3.98. Found: C, 68.86; H, 8.61; $COCH_3$, 21.2; OC_2H_5, 2.96.

The ultraviolet and infrared spectra were identical with those of the preparation obtained by acetylation of the monoacetate IX.

The triacetate X was also obtained in an early attempt to acetylate the perchlorate with acetic anhydride in pyridine. The amorphous product yielded on chromatographing about 30% of X and for the rest non-crystallizable products.

Hydrolysis of the perchlorate (85 mg.) with methanolic potassum hydroxide at room temperature followed by distribution between chloroform and 1 *N* hydrochloric acid gave 49 mg. of a "neutral" fraction and 18 mg. of basic products. The former on repeated recrystallization from dilute ethanol yielded plates melting at 243–253°, $[\alpha]D$ −133°. which did not depress the melting point of the monoacetate IX obtained in the same manner from VII.

Sulfonic Acid.—The sodium salt was obtained as a yellow amorphous powder by lyophilizing the combined methanol eluates from water. It was readily soluble in the lower alcohols, acetone, chloroform and concentrated or 10% acetic acid, sparingly soluble in warm ethyl acetate, and insoluble in hexane, ether and carbon tetrachloride. Addition of hydrochloric acid to its aqueous solution, which is highly acidic (*p*H 4–5), gave an amorphous precipitate which was soluble in alkali; ultraviolet spectrum, Fig. 1, curve 5.

Anal. Found: C, 54.89; H, 6.56; S, 6.29; Na, 2.58; $COCH_3$, 12.3. Other preparations showed S/Na ratios approximating, 1.0.

Conversion of Acetolysis Product VII to Indanone III and to Sulfonic Acid.—A solution of compound VII (62 mg.) in acetic anhydride (7 cc.), acetic acid (3 cc.) and concentrated sulfuric acid (0.1 cc.) was allowed to stand at room temperature for 24 hours, and was then worked up as described for the acetolysis of diacetyljervine. The residue of the chloroform extract (34 mg.) was chromatographed in the usual manner. The fractions eluted with benzene, together 10.4 mg., were crystalline. On repeated recrystallization from ethyl acetate–hexane they yielded 2.7 mg. of slightly impure indanone VII, m.p. 233.5–235.5°, identified as such by the ultraviolet characteristics (λ_{max}^{alc} 250 mμ, ε 11,300; 300 mμ, ε 1500).

The fractions eluted with benzene–ether 9:1 (A, 6.8 mg.), ether (B, 2.5 mg.), and methanol (*C*, 10.5 mg.) were all amorphous. They were shown by sulfur analysis and ultraviolet measurements (fraction A: S, 2.6%; λ_{max}^{alc} 272 mμ, $E_{1\ cm}^{1\%}$ 655; fraction C: S, 3.3%, λ_{max}^{alc} 274 mμ, $E_{1\ cm}^{1\%}$ 400) to consist largely of the sulfonate discussed above.

Acknowledgments.—The authors are indebted to Mr. Joseph Alicino and his associates for the microanalyses, and to Dr. Nettie H. Coy and her colleagues, Mr. Carl Sabo and Mr. Charles Fairchild, for the ultraviolet and infrared measurements.

NEW BRUNSWICK, N. J.