213. *The Structure of Products Derived from Tazettine with Acetic Anhydride and Sulphuric Acid.*

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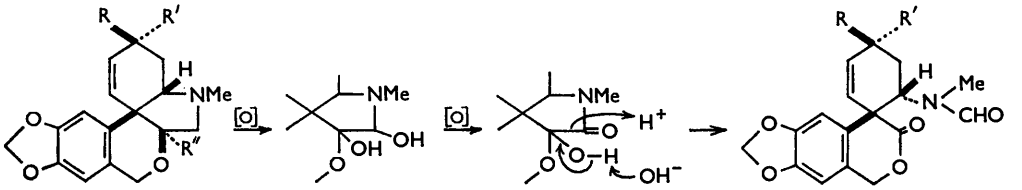
The structure (VII; R = H, R′ = OAc) is advanced for the " neutral compound " obtained by Kondo when tazettine was treated with acetic anhydride in the presence of sulphuric acid. The generality of this reaction was tested upon dihydrotazettine, dihydroisotazettinol, dihydrotazettinol, and demethoxydihydrotazettine.

Tazettine, an alkaloid occurring in the Amaryllidaceæ, has been shown [1,2] to have structure (I; R = OMe, R′ = H, R″ = OH), containing a β-oxopyrrolidine moiety in which the carbonyl group is masked by forming a hemiketal arrangement.* It is conceivable that the methylene group between the nitrogen atom and the masked carbonyl group should be vulnerable to oxidation. However, oxidative studies of tazettine

* That the configuration of the hemiketal hydroxyl group is as formulated will be conclusively shown in a forthcoming paper.

[1] Ikeda, Taylor, Tsuda, Uyeo, and Yajima, *J.*, 1956, 4749.
[2] Irie, Tsuda, and Uyeo, *J.*, 1959, 1446.

have led to a complex of products except where manganese dioxide was used as a reagent.[1,3] The main product in that case was the amido-lactone (IV; R = OMe, R′ = H) which was presumably produced by cleavage of an intermediate (III), which was formed in turn from the primary product, the carbinolamine (II).



(I) (II) (III) (IV)

The free hydroxyl group of the hemiketal moiety must play an important rôle in accelerating this type of oxidation because when it was masked by acetylation, as in acetyltazettine (I; R = OMe, R′ = H, R″ = OAc), diacetyltazettinol (I; R = R″ = OAc, R′ = H), or diacetylisotazettinol (I; R = H, R′ = R″ = OAc), the oxidation did not proceed to any noticeable extent.

This paper is concerned with tazettine derivatives containing an oxygenated methylene grouping at the above-mentioned position.

Späth and Kahovec [4] prepared acetyltazettine (I; R = OMe, R′ = H, R″ = OAc) by the action of acetic anhydride on tazettine in the presence of a few drops of sulphuric acid in the cold. Kondo and Ikeda [5] later reported failure to reproduce this preparation, as they obtained a neutral compound, m. p. 254° (decomp.), together with a diacetate, m. p. 137—140°, but no acetyltazettine. In view of this discrepancy, acetylation of tazettine with acetic anhydride and sulphuric acid was re-investigated. It was found that when the quantity of the sulphuric acid added to a mixture of tazettine and acetic anhydride was so small that some starting material was recovered unchanged, the only isolable product was the normal acetylation product of tazettine (Späth's product). When the amounts of sulphuric acid were increased, however, acetyltazettine was no longer isolable and Kondo and Ikeda's products were obtained. The yield of the neutral fraction could be increased to a maximum of 90% by increasing the proportion of sulphuric acid to anhydride. The " diacetate " was found to be a mixture of diacetyltazettinol [1] (I; R = R″ = OAc, R′ = H) and diacetylisotazettinol [1] (I; R = H, R′ = R″ = OAc) in a ratio of about 1 : 2, and these were separated by chromatography. The neutral fraction was also found to be a mixture which consisted principally of a crystalline compound, acetyldehydroisotazettinol, m. p. 263° (decomp.). Identity of this with the " neutral compound " of Kondo and Ikeda was confirmed by comparison of the infrared spectra. From the mother-liquors another compound, acetyldehydrotazettinol, m. p. 204—207°, was isolated in pure crystalline form only after chromatography over alumina and silica gel.

Analyses indicated that acetyldehydroisotazettinol has the empirical formula $C_{19}H_{19}O_6N$ rather than $C_{20}H_{25}O_6N$ attributed to it by the earlier authors.[5] It contained one acetyl but no methoxyl group. The infrared spectrum did not show the presense of a hydroxyl group, and the ultraviolet absorption spectrum revealed that no double bond was conjugated with the benzene ring. Alkaline hydrolysis yielded a deacetyl compound, dehydroisotazettinol, m. p. 264° (decomp.), which was also very weakly basic and exhibited a sharp hydroxyl band in the infrared spectrum at 3425 cm.$^{-1}$, but neither acetyl nor other carbonyl bands in the carbonyl region. Dehydroisotazettinol did not depress

[3] Highet and Wildman, *Chem. and Ind.*, 1955, 1159.
[4] Späth and Kahovec, *Ber.*, 1934, **67**, 1501.
[5] Kondo and Ikeda, *Ann. Report ITSUU Lab.*, 1951, **2**, 55.

the melting point of acetyldehydroisotazettinol. This and the similarity of the melting points of these two compounds would probably have led the earlier workers [5] to assume erroneously that acetyldehydroisotazettinol could not be hydrolysed with alkali. We have shown, however, by the infrared spectra that reacetylation of dehydroisotazettinol regenerated the parent compound.

Treatment of acetyldehydroisotazettinol with lithium aluminium hydride furnished isotazettatriol (V) which was characterised, after being heated with dilute sulphuric acid, as its derivative deoxyisotazettinol [1] (I; R = R'' = H, R' = OH). Prolonged refluxing of acetyldehydroisotazettinol with sodium borohydride in methanol afforded isotazettinol [1] (I; R = H, R' = R'' = OH) while catalytic hydrogenation of dehydroisotazettinol with platinum oxide gave rise to dihydroisotazettinol [2] (VI; R = H, R' = R'' = OH). Whereas acetyldehydroisotazettinol was unaffected by manganese dioxide in chloroform, dehydroisotazettinol gave an αβ-unsaturated ketone, dehydrotazettinone, with the same reagent.

These results showed that dehydroisotazettinol was closely related in structure and configuration to isotazettinol.

Although acetyldehydroisotazettinol was almost neutral or so weakly basic as to be insoluble in dilute mineral acids and did not give a quaternary salt with methyl iodide, it was readily soluble in cold concentrated hydrochloric acid. When dry hydrogen chloride was passed into a chloroform–benzene solution of acetyldehydroisotazettinol, a quantitative yield of a deliquescent amorphous hydrochloride was obtained which exhibited infrared absorptions at 3330—3570 (OH and H_2O), 2500—2740 ($\equiv N^+-H$), 1620—1660 (H_2O), and 1739 cm.$^{-1}$ (OAc), but no unconjugated $\rangle C=N^+\langle$ band in the region 1670—1700 cm.$^{-1}$. The hydrochloride did not give reproducible analytical values owing to high solvation, but regenerated acetyldehydroisotazettinol on basification of its aqueous solution with sodium hydrogen carbonate.

Reduction of the hydrochloride with sodium borohydride in methanol took place far more readily than in the case of the free base, and the product isolated was the known monoacetylisotazettinol [2] (I; R = H, R' = OAc, R'' = OH) in which the acetyl group is attached to the hydroxyl allylic to the double bond in ring B of isotazettinol, as shown by conversion with manganese dioxide into the amido-lactone (IV; R = H, R' = OAc) containing still an acetyl group.

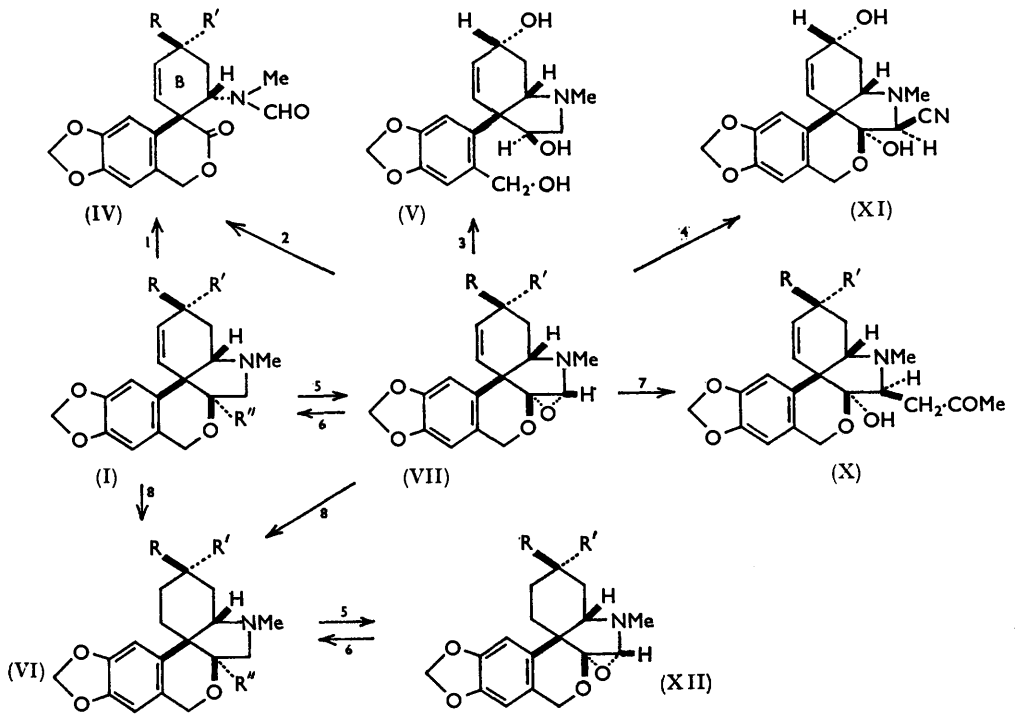
Since the arrangement in ring B of the monoacetylisotazettinol cannot be considered to have been newly formed as a result of borohydride treatment of the hydrochloride or conversion of acetyldehydroisotazettinol into the hydrochloride, there appeared to be no doubt that acetyldehydroisotazettinol and its hydrochloride contained the same moiety as that of ring B in the monoacetylisotazettinol. Evidence in support of this was provided by the catalytic hydrogenation of the hydrochloride with palladium–carbon in water, which gave a deacetoxy-compound, demethoxytazettine [2] (I; R = R' = H, R'' = OH), as expected.

The facts that acetyldehydroisotazettinol contains no hydroxyl group, and two hydrogen atoms less than monoacetylisotazettinol (I; R = H, R' = OAc, R'' = OH), but is readily reduced to the latter through its hydrochloride, led us to conclude that the former compound was an epoxide, formulated as (VII; R = H, R' = OAc), a structure which was also supported by its weak basicity.[6]

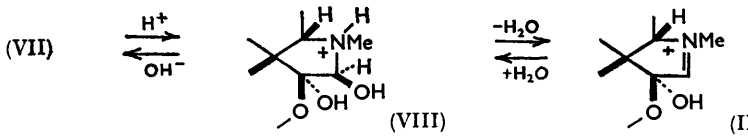
For the hydrochloride a carbinolamine structure (VIII) rather than that of an ordinary amine hydrochloride is preferable because of its highly solvated form and its ready reducibility to monoacetylisotazettinol. In support of this view, the hydrochloride readily formed an acetonyl compound [7] (X; R = H, R' = OAc) when refluxed in acetone for a short time. Although the infrared spectrum of the hydrochloride did not indicate

[6] Bloom and Briggs, *J.*, 1952, 3591.
[7] Small and Lutz, "Chemistry of the Opium Alkaloids," U.S. Treasury Dept. Public Health Service, No. 103, 1932, p. 49.

any C=N$^+$ absorption band in a solid state, it is probable that (VIII) is in equilibrium in solution with the anhydrous form (IX).



Dehydroisotazettinol gave likewise a hydrochloride which also gave an acetonyl compound (X; R = H, R′ = OH), acetylation of which in cold pyridine with acetic anhydride afforded a monoacetyl derivative identical with the acetonyl compound derived from acetyldehydroisotazettinol hydrochloride. Further, a reaction characteristic of



carbinolamines was observed in the formation of a cyanide (XI) when the hydrochloride was treated with sodium cyanide in boiling methanol.

With the structure of acetyldehydroisotazettinol and the hydrochloride solved, it was desirable to oxidise acetyldehydroisotazettinol (with alkaline potassium ferricyanide) to the lactam (III) The product isolated was, however, not the expected compound, but the amido-lactone (IV; R = H, R′ = OH) identical with one of the products [2] from the oxidation of isotazettinol with manganese dioxide. Presumably the lactam initially formed rearranged to the amido-lactone by a process similar to that in the manganese dioxide oxidation of tazettine mentioned above (II ⟶ III ⟶ IV).

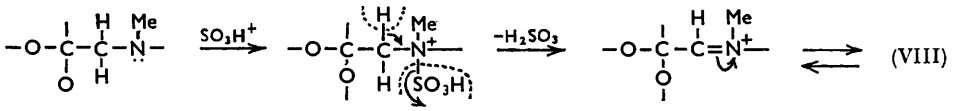
Acetyldehydrotazettinol was isomeric with acetyldehydroisotazettinol and resembled it in being almost neutral to acid and in containing one acetyl but no hydroxyl group (infrared spectrum). The hydrolysis product, dehydrotazettinol, afforded with manganese

dioxide the ketone, dehydrotazettinone (VII; R, R′ = O), obtained similarly from dehydroisotazettinol, suggesting that they were epimers in respect to the allylic hydroxyl group. Since dehydrotazettinol yielded tazettinol [1] (I; R = R″ = OH, R′ = H) on treatment of its hydrochloride with sodium borohydride, acetyldehydrotazettinol has been assigned the formula (VII; R = OAc, R′ = H) in analogy with acetyldehydroisotazettinol.

Formation of acetyldehydrotazettinol and acetyldehydroisotazettinol is considered to be the result of dehydrogenation of the intermediates, acetyltazettinol and acetylisotazettinol, derived from tazettine probably by replacement of the methoxyl group by an acetoxyl anion originating from acetic anhydride and sulphuric acid. The fact that both acetyldehydrotazettinol and acetyldehydroisotazettinol were invariably obtained when either tazettinol or isotazettinol was treated with acetic anhydride and sulphuric acid would also be accounted for by assuming that the same type of replacement took place, at least partly, during the conversion of the hydroxyl group in tazettinol or isotazettinol into the acetoxyl group.

The dehydrogenation is thought to have been caused by the sulphuric acid, acetic anhydride playing the rôle of a diluent, since the reaction mixture contained sulphurous acid as shown by a positive Malachite Green test,[8] although it was not known to us that sulphuric acid could have acted as an oxidising agent at room temperature.* In support of this, we have confirmed that other acids, such as perchloric acid and boron trifluoride, do not convert tazettine into dehydro-derivatives, the only isolable products being a mixture of diacetyltazettinol and diacetylisotazettinol.

A possible mechanism for this reaction might involve initial addition of SO_3H^+ cation to the nitrogen atom followed by elimination of the elements of sulphurous acid to give an anhydronium base, as shown.†



The unusual nature of the reaction prompted us to investigate other derivatives of tazettine containing the hemiketal moiety. Dihydroisotazettinol (VI; R = H, R′ = R″ = OH) gave an analogous product (XII; R = H, R′ = OAc) under the same conditions in fair yield. This acetyldehydrodihydroisotazettinol was not soluble in cold dilute mineral acids and contained one acetyl but no hydroxyl group, as shown in its infrared spectrum. Alkaline hydrolysis afforded dehydrodihydroisotazettinol (XII; R = H, R′ = OH) which gave back dihydroisotazettinol on treatment of its hydrochloride with sodium borohydride.

Dihydrotazettinol (VI; R = R″ = OH, R′ = H) similarly afforded acetyldehydrodihydrotazettinol (XII; R = OAc, R′ = H) which did not crystallise but was characterised as its saponification product, dehydrodihydrotazettinol (XII; R = OH, R′ = H). Analogous properties of the compound and reduction of its hydrochloride to dihydrotazettinol by sodium borohydride confirmed its structure.

In expectation that a methoxyl group attached to a saturated ring might be more stable than when it was allylic, an analogous reaction was carried out with dihydrotazettine (VI; R = OMe, R′ = H, R″ = OH). With the acetic anhydride–sulphuric acid complex, it gave unexpectedly a mixture from which only a small amount of acetyldehydrodihydroisotazettinol was isolated as crystals. Saponification of the mixture

* There may be some analogy between this reaction and the oxidative phosphorylation reported by Sir Alexander Todd and his collaborators (Clark, Kirby, and Todd, *Nature*, 1958, **181**, 1650).

† It is possible that the acetic anhydride formed some pyrosulphuric acid which then dissociated to yield the active species, SO_3H^+.

[8] Feigl, " Spot Tests," Elsevier Publishing Co., Amsterdam, Vol. I, 1954, p. 286.

furnished, however, both dehydrodihydroisotazettinol (XII; R = H, R′ = OH) and dehydrodihydrotazettinol (XII; R = OH, R′ = H).

Demethoxydihydrotazettine [2] (VI; R = R′ = H, R″ = OH) represents the simplest compound among those which leave the skeleton and the hemiketal moiety of tazettine unaffected, and it contains no functional groups in ring B which might be attacked by acetic anhydride and sulphuric acid. The product isolated from a neutral fraction of the reaction mixture gave analyses for $C_{17}H_{19}O_4N,\frac{1}{2}H_2O$, and showed a weak absorption at about 3333 cm.$^{-1}$ (H_2O) but no bands in the carbonyl region, indicating the absence of an acetyl group. Along with these results, the fact that its hydrochloride, prepared in benzene, reverted to demethoxydihydrotazettine (VI; R = R′ = H, R″ = OH) on treatment with either sodium borohydride or hydrogen and palladium–carbon, proved the structure of this product as dehydrodemethoxydihydrotazettine (XII; R = R′ = H).

Confirming the view that acetic anhydride–sulphuric acid complex acted upon tazettine and its derivatives as a mild oxidising agent, we have shown that dehydrodemethoxydihydrotazettine (XII; R = R′ = H) is also obtained in a low yield on oxidation of demethoxydihydrotazettine with mercuric acetate. An analogous reaction with tazettine was not successful, the only product isolated containing the metal.

Experimental

Ultraviolet absorption spectra were determined for 95% ethanol solutions, and infrared spectra for Nujol mulls unless otherwise stated. Identities were confirmed by infrared comparisons.

Acetylation of Tazettine with Acetic Anhydride and Sulphuric Acid.—(*a*) Tazettine (0·3 g.) in acetic anhydride (10 ml.) containing concentrated sulphuric acid (2 drops) was kept at about 5° for 2 days. Basification with aqueous ammonia and extraction with ether afforded a mixture (0·3 g.) which was separated by chromatography in benzene over alumina. The benzene eluate gave acetyltazettine (I; R = OMe, R′ = H, R″ = OAc) (0·17 g.), plates, m. p. and mixed m. p. 124—125° (from ethanol). Further elution with chloroform yielded unchanged tazettine (0·13 g.).

(*b*) Tazettine (0·21 g.), acetic anhydride (10 ml.), and concentrated sulphuric acid (5 drops) were kept overnight at room temperature. The mixture was poured into ice-water (60 ml.), basified with sodium hydrogen carbonate, and extracted with ether. The ethereal layer was extracted with 2% sulphuric acid, and the aqueous acidic solution basified with sodium hydrogen carbonate and extracted with ether. Evaporation of the ether gave a solid (0·15 g.), m. p. 137—140°, apparently identical with Kondo's diacetate.[5] This was chromatographed in benzene over alumina; the first benzene eluate gave diacetyltazettinol (I; R = R″ = OAc, R′ = H) (40 mg.), m. p. and mixed m. p. 199—200° (from ethanol). Further elution with benzene gave diacetylisotazettinol (I; R = H, R′ = R″ = OAc) (75 mg.), m. p. and mixed m. p. 148—150°. The ethereal layer from which the basic product had been removed as mentioned above gave, on evaporation, a solid from which acetyldehydroisotazettinol, m. p. 263° (decomp.) (10 mg.), was obtained after crystallisation from benzene. The infrared spectrum of this sample was identical with that of a sample, m. p. 254° (decomp.), previously obtained by Kondo and Ikeda.[5]

(*c*) Tazettine (1·2 g.) in acetic anhydride (10 ml.) and concentrated sulphuric acid (2 ml.) was set aside at room temperature for 2 days. The mixture was poured into ice-water, basified with sodium hydrogen carbonate, and extracted with chloroform. The water layer decolorized a dilute solution of Malachite Green. The chloroform layer was washed with 3% hydrochloric acid to remove some basic substances (0·1 g.), then washed with water, dried, and evaporated to dryness to yield a solid, which was treated with ether to separate it into a soluble and an insoluble fraction. The latter crystallised from benzene to give *acetyldehydroisotazettinol* (VII; R = H, R′ = OAc) (0·7 g.), needles, m. p. 263° (decomp.), $[\alpha]_D$ +126° (*c* 3·0 in $CHCl_3$) (lit.,[5] $[\alpha]_D$ +128·4° in $CHCl_3$), $\lambda_{max.}$ 240 and 292 mμ (log ε 3·62 and 3·57), $\nu_{max.}$ 1739 and 1724 cm.$^{-1}$ (OAc) in Nujol mull, 1724 cm.$^{-1}$ (OAc) in $CHCl_3$ (Found: C, 63·7; H, 5·1; N, 3·7; OMe, 0; *C*-Me, 4·0; Ac, 12·9. $C_{19}H_{19}NO_6$ requires C, 63·9; H, 5·4; N, 3·9; 1*C*-Me, 3·9; 1Ac, 12·0%). The former fraction was chromatographed in benzene over alumina. The first benzene eluate gave an amorphous

material (0·25 g.), and the second gave an additional crop of acetyldehydroisotazettinol (50 mg.). Re-chromatography of the above amorphous material in chloroform over silica gel and elution with chloroform gave *acetyldehydrotazettinol* (VII; R = OAc, R′ = H) (0·2 g.), m. p. 204—207° (from ligroin), $[\alpha]_D$ −26° (*c* 0·2 in $CHCl_3$), $\nu_{max.}$ 1730 cm.$^{-1}$ (OAc) (in a KBr disc) (Found: C, 64·3; H, 5·6; N, 3·5%).

Attempted Oxidation of Acetyltazettine (I; R = OMe, R′ = H, R″ = OAc), *Diacetyltazettinol* (I; R = R″ = OAc, R′ = H), *Diacetylisotazettinol* (I; R = H, R′ = R″ = OAc), *and Acetyldehydroisotazettinol* (VII; R = H, R′ = OAc) *by Manganese Dioxide.*—Acetyltazettine, diacetyltazettinol, diacetylisotazettinol, and acetyldehydroisotazettinol (0·1 g.) were severally stirred in chloroform (10—12 ml.) with manganese dioxide (1 g.) at room temperature for 24 hr. The starting materials were recovered in not less than 90% yield.

Dehydroisotazettinol.—(*a*) Acetyldehydroisotazettinol (50 mg.) was refluxed in tetrahydrofuran (10 ml.), 3% ethanolic potassium hydroxide (4 ml.), and water (2 ml.) for 1 hr. The mixture was evaporated under reduced pressure, then diluted with water, and the precipitate was collected, to give *dehydroisotazettinol* (VII; R = H, R′ = OH) (40 mg.), needles, m. p. 264° (decomp.) (from benzene), $[\alpha]_D$ +95·4° (*c* 2·4 in $CHCl_3$) $\nu_{max.}$ 3425 cm.$^{-1}$ (OH) (Found: C, 64·7; H, 5·5; N, 4·3. $C_{17}H_{17}NO_5$ requires C, 64·8; H, 5·4; N, 4·4%).

When acetyldehydroisotazettinol (50 mg.) was heated with potassium hydroxide (0·4 g.) in diethylene glycol (6 ml.) at 150° for 1 hr. 6-phenylpiperonyl alcohol (5 mg.), m. p. and mixed m. p. 101—102°, was isolated from the mixture.

(*b*) The acetate (50 mg.) in 20% hydrochloric acid (10 ml.) was heated at 100° for 10 min. Basification and extraction with chloroform afforded dehydroisotazettinol (35 mg.), m. p. 264° (decomp.), identical with the product obtained as in (*a*). Acetylation of dehydroisotazettinol with sodium acetate and acetic anhydride on a water-bath gave acetyldehydroisotazettinol (VII; R = H, R′ = OAc), m. p. 263° (decomp.).

Reduction of Acetyldehydroisotazettinol.—(*a*) Treatment of the acetate (0·12 g.) with lithium aluminium hydride (0·1 g.) in refluxing tetrahydrofuran (10 ml.) gave, in 3 hr., a triol (V) which when heated in 3% sulphuric acid (10 ml.) on a water-bath for 1 hr. afforded deoxyisotazettinol (I; R = R″ = H, R′ = OH) (0·1 g.), m. p. and mixed m. p. 120—122° (after chromatography in benzene on alumina followed by crystallisation from ether).

(*b*) Sodium borohydride (0·12 g.) was added in portions to the acetate (50 mg.) in boiling methanol (10 ml.) during 30 hr. After removal of the solvent, water was added, and the solution saturated with ammonium chloride and extracted with chloroform, to give, after concentration, isotazettinol (I; R = H, R′ = R″ = OH) (30 mg.), m. p. and mixed m. p. 204—206° (from ethanol).

Hydrogenation of Dehydroisotazettinol.—Dehydroisotazettinol (15 mg.) and platinum oxide (50 mg.) in ethanol (10 ml.) were stirred in hydrogen at room temperature. After 6 hr. the mixture was filtered and evaporated to dryness, and the residue taken up into chloroform and extracted with 3% hydrochloric acid. The acidic layer afforded, after basification and extraction with chloroform, dihydroisotazettinol (VI; R = H, R′ = R″ = OH), m. p. and mixed m. p. 250°.

Oxidation of Dehydroisotazettinol by Manganese Dioxide.—Dehydroisotazettinol (60 mg.) in chloroform (8 ml.) was stirred with manganese dioxide (0·7 g.) at room temperature for 22 hr. The product isolated with chloroform gave *dehydrotazettinone* (VII; R, R′ = O), needles, m. p. 271° (decomp.) (from acetone), $\nu_{max.}$ 1684 cm.$^{-1}$ (conjugated ketone) (Found: C, 61·9; H, 5·4. $C_{17}H_{15}NO_5,H_2O$ requires C, 61·6; H, 5·2%).

Acetyldehydroisotazettinol Hydrochloride.—Dry hydrogen chloride was passed into a solution of acetyldehydroisotazettinol (0·1 g.) in chloroform (1 ml.) and benzene (20 ml.) with cooling. This gave the hydrochloride (0·1 g.) as a white deliquescent powder, which was washed with benzene, then with light petroleum (b. p. 60—80°), and dried over P_2O_5 *in vacuo*. An aqueous solution of the hydrochloride regenerated acetyldehydroisotazettinol on neutralisation with sodium hydrogen carbonate.

Hydrogenation of the hydrochloride (90 mg.) in water (20 ml.) with 5% palladium–carbon (0·1 g.) at room temperature for 1·5 hr. gave demethoxytazettine (I; R = R′ = H, R″ = OH) (70 mg.), m. p. and mixed m. p. 193—194° (from ether).

The hydrochloride (0·1 g.) dissolved in boiling acetone (10 ml.) in a few minutes. After 1 hr., the mixture was evaporated to give a residue which was taken up in water, washed with chloroform, basified with sodium carbonate, and extracted with chloroform. Concentration

of the chloroform extract yielded the *acetonyl derivative* (X; R = H, R′ = OAc), m. p. 156—157°, prisms from methanol, $[\alpha]_D$ +250° (*c* 0·96 in $CHCl_3$), $\nu_{max.}$ 3289 (OH) and 1720 cm.$^{-1}$ (CO, OAc) (Found: C, 63·9; H, 6·1. $C_{22}H_{25}NO_7$ requires C, 63·6; H, 6·1%).

Sodium borohydride (0·15 g.) in methanol (10 ml.) was added dropwise to a stirred solution of the foregoing hydrochloride (80 mg.) in methanol (20 ml.) at 5°. After 1 hr., the mixture was acidified with acetic acid and evaporated to dryness; the residue was redissolved in water and basified with aqueous ammonia. Extraction with ether and evaporation of the extract afforded monoacetylisotazettinol (I; R = H, R′ = OAc, R″ = OH) (50 mg.), m. p. and mixed m. p. 193—194° (from ethanol).

Oxidation of Monoacetylisotazettinol by Manganese Dioxide.—Acetylisotazettinol (35 mg.) in chloroform (8 ml.) was stirred with manganese dioxide (0·5 g.) for 26 hr. The oxide was filtered off and washed with chloroform, and the combined filtrate and washings were evaporated to give the *amido-acetate* (IV; R = H, R′ = OAc), leaflets (from methanol), m. p. 266—267°, $\nu_{max.}$ 1736 (OAc), 1718 (lactone), and 1672 cm.$^{-1}$ (amide) in Nujol mull, and 1737 (OAc), 1724 (lactone), and 1675 cm.$^{-1}$ (amide) in $CHCl_3$ (Found: C, 61·5; H, 5·0. $C_{19}H_{19}NO_7$ requires C, 61·1; H, 5·1%). Hydrolysis of this (5 mg.) by 1% methanolic potassium hydroxide on a water-bath for 3 min., followed by acidification with dilute hydrochloric acid, gave the amide (IV; R = H, R′ = OH) (2 mg.), m. p. and mixed m. p. 207—209°. Reacetylation of the latter with acetic anhydride in pyridine gave the foregoing amido-acetate (IV; R = H, R′ = OAc).

Dehydroisotazettinol Hydrochloride.—The hydrochloride was obtained as a deliquescent white powder by the procedure described for acetyldehydroisotazettinol hydrochloride. The infrared spectrum of this showed absorptions at 3200 (OH), 2740—2500 ($\equiv N^+H$) and 1626 cm.$^{-1}$ (H_2O). The hydrochloride regenerated the dehydro-base (VII; R = H, R′ = OH) on neutralisation of its aqueous solution with sodium hydrogen carbonate.

An aqueous solution (20 ml.) of the hydrochloride (50 mg.) was hydrogenated over 5% palladium–carbon (0·1 g.) for 4 hr. Removal of the catalyst followed by basification of the filtrate and extraction with chloroform gave isotazettinol (I; R = H, R′ = R″ = OH) (24 mg.), m. p. and mixed m. p. 204—206° (from ethanol).

The above hydrochloride was heated under reflux in acetone for 2 hr., during which it gradually changed to colourless prisms. The resulting *acetonyl derivative hydrochloride* was crystallised from acetone, to give prisms, m. p. 187—188° (decomp.), $\nu_{max.}$ 3330—3030 (OH), 2700—2500 ($\equiv N^+H$), 1706 (CO), and 1640—1620 cm.$^{-1}$ (H_2O) (Found: C, 58·7; H, 6·5. $C_{20}H_{23}NO_6$,HCl requires C, 58·9; H, 5·9%). The hydrochloride thus obtained was dissolved in water, basified with sodium carbonate, and extracted with chloroform. Concentration of the chloroform solution yielded the *acetonyl derivative* (X; R = H, R′ = OH), m. p. 180—181°, prisms from methanol, $[\alpha]_D$ +193° (*c* 0·97 in $CHCl_3$), $\nu_{max.}$ 3185 (OH) and 1712 cm.$^{-1}$ (CO) (Found: C, 64·4; H, 6·2. $C_{20}H_{23}NO_6$ requires C, 64·3; H, 6·2%). Acetylation of this (30 mg.) with acetic anhydride (0·5 ml.) and pyridine (1 ml.) at room temperature overnight gave the acetate (X; R = H, R′ = OAc), m. p. 156—157°, identical with the acetonyl compound derived from acetyldehydroisotazettinol.

The hydrochloride (100 mg.) and sodium cyanide (200 mg.) in methanol (10 ml.) were heated under reflux for 3 hr. The precipitated sodium chloride was filtered off and the filtrate was evaporated to dryness, to give a residue which was extracted with chloroform. The extract was washed with water, dried, and evaporated, affording the *cyano-compound* (XI) (50 mg.) as colourless needles, m. p. 225—227° (decomp.) (from ethanol), $\nu_{max.}$ 3257 (OH) and 2232 cm.$^{-1}$ (CN) (Found: C, 63·0; H, 5·4; N, 8·1. $C_{18}H_{18}N_2O_5$ requires C, 63·2; H, 5·3; N, 8·2%).

Ferricyanide Oxidation of Acetyldehydroisotazettinol.—Potassium ferricyanide (0·5 g.) and potassium hydroxide (0·3 g.) in water (10 ml.) were added to acetyldehydroisotazettinol (VII; R = H, R′ = OAc) (70 mg.) in hot dioxan (10 ml.), and the mixture was heated on a water-bath for 1 hr. After dilution with water, the mixture was washed with chloroform, acidified with dilute sulphuric acid, and extracted with chloroform. Concentration of the extract gave the amide (IV; R = H, R′ = OH) (5 mg.), m. p. and mixed m. p. 216—218° after crystallisation from methanol and drying over P_2O_5.

Dehydrotazettinol.—Acetyldehydrotazettinol (VII; R = OAc, R′ = H) (50 mg.) was refluxed in 3% ethanolic potassium hydroxide (10 ml.) for 1 hr. The mixture was concentrated to half-volume, diluted with water, and extracted with chloroform, to give *dehydrotazettinol* (VII; R = OH, R′ = H) (32 mg.), prisms, m. p. 244—245° (decomp.) (from benzene), $[\alpha]_D$

+28° (*c* 0·7 in $CHCl_3$), $\nu_{max.}$ 3213 cm.$^{-1}$ (OH) (Found: C, 67·3; H, 5·7. $C_{17}H_{17}NO_5,\frac{1}{2}C_6H_6$ requires C, 67·8; H, 5·7%).

Oxidation of Dehydrotazettinol by Manganese Dioxide.—Dehydrotazettinol (25 mg.) in chloroform (10 ml.) was stirred with manganese dioxide (0·5 g.) at room temperature for 1 hr. After removal of the manganese dioxide and concentration of the filtrate to dryness, the residue was crystallised from acetone to furnish dehydrotazettinone (VII; R, R′ = O), m. p. and mixed m. p. 271° (decomp.).

Reduction of Dehydrotazettinol Hydrochloride by Sodium Borohydride.—Dehydrotazettinol (20 mg.) was dissolved in methanol (2 ml.) containing concentrated hydrochloric acid (2 drops), then evaporated to dryness, and the resulting hydrochloride, redissolved in methanol (2 ml.), was treated with sodium borohydride (0·1 g.) at room temperature for 1 hr. The mixture was concentrated and taken up into water which was saturated with ammonium chloride and extracted with chloroform. The chloroform extract was dried and evaporated, to yield tazettinol (I; R = R″ = OH, R′ = H) (15 mg.), m. p. and mixed m. p. 185—187°.

Conversion of Tazettinol or Isotazettinol into Dehydrotazettinol and Dehydroisotazettinol.—(*a*) Tazettinol (I; R = R″ = OH, R′ = H) (100 mg.) in acetic anhydride (5 ml.) and concentrated sulphuric acid (1 ml.) was kept overnight at room temperature. The mixture was poured into ice-water, basified with sodium hydrogen carbonate, and extracted with chloroform. The chloroform extract was washed with 3% hydrochloric acid, dried, and evaporated to dryness and the residue hydrolysed in 10% methanolic potassium carbonate (20 ml.) under reflux for 3 hr. After removal of methanol, water was added and the whole extracted with chloroform. Concentration of the chloroform gave a gum which was chromatographed in benzene over alumina. The benzene eluate gave dehydroisotazettinol (VII; R = H, R′ = OH) (40 mg.), m. p. 262—263° (decomp.); elution with acetone gave dehydrotazettinol (VII; R = OH, R′ = H) (30 mg.), m. p. 238—241° (decomp.).

(*b*) Isotazettinol (I; R = H, R′ = R″ = OH) (110 mg.) was treated with acetic anhydride (5 ml.) and concentrated sulphuric acid (1 ml.) for 12 hr. at room temperature and worked up as described in (*a*), whereupon there were obtained dehydroisotazettinol (VII; R = H, R′ = OH) (55 mg.) and dehydrotazettinol (VII: R = OH, R′ = H) (10 mg.).

Action of Acetic Anhydride and Boron Trifluoride on Tazettine.—Tazettine (280 mg.) in acetic anhydride (5 ml.) and boron trifluoride–ether complex (1·5 ml.) was set aside for 12 hr. The mixture was poured into water (50 ml.) and basified with sodium carbonate, and the deposited precipitate collected, washed with water, dried (290 mg.; m. p. 139—142°), and crystallised from ether, to give diacetylisotazettinol (I; R = H, R′ = R″ = OAc) (180 mg.), m. p. and mixed m. p. 148—150°. The mother-liquor from this was dissolved in ether and extracted with 3% hydrochloric acid. Basification and extraction of the acidic solution yielded an oil which was chromatographed in benzene over alumina. From the first benzene eluate, diacetyltazettinol (I; R = R″ = OAc, R′ = H) (40 mg.), m. p. and mixed m. p. 199—200°, was isolated and the second benzene eluate yielded an additional crop of diacetylisotazettinol (10 mg.). The ethereal extract gave only a trace of gum, no dehydro-base being isolated.

Action of Acetic Anhydride and Perchloric Acid on Tazettine.—60% Perchloric acid (20 drops) was added dropwise with cooling to tazettine (0·2 g.) in acetic anhydride (5 ml.), and the whole kept at 10° for 24 hr. The mixture was poured into water (50 ml.), filtered, basified with sodium carbonate, and extracted with chloroform, and the chloroform solution was extracted with 1% hydrochloric acid. Basification of the aqueous layer and extraction with ether furnished diacetylisotazettinol (100 mg.), m. p. and mixed m. p. 141—144°. The chloroform layer gave, on concentration, diacetyltazettinol (100 mg.), m. p. and mixed m. p. 199—200°. No neutral substances were isolated from the reaction mixture.

Action of Sulphuric Acid and Acetic Anhydride on Dihydroisotazettinol.—Dihydroisotazettinol (VI; R = H, R′ = R″ = OH) (130 mg.) in acetic anhydride (5 ml.) containing concentrated sulphuric acid (0·5 ml.) was kept at room temperature overnight and worked up as described above. The *acetyldehydrodihydroisotazettinol* (XII; R = H, R′ = OAc) (90 mg.) thus obtained crystallised from chloroform–ethanol as needles, m. p. 240—241°. The infrared spectrum showed a band at 1721 cm.$^{-1}$ (OAc) but no absorptions in the 3 μ region (Found: C, 63·8; H, 6·0. $C_{19}H_{21}NO_6$ requires C, 63·5; H, 5·9%).

Dehydrodihydroisotazettinol.—Acetyldehydrodihydroisotazettinol (50 mg.) in tetrahydrofuran (10 ml.) and 3% methanolic sodium hydroxide (5 ml.) was heated on a water-bath

for 2 hr. After evaporation of the mixture, water was added and the precipitate formed was collected, washed with water and methanol, and crystallised from methanol, to give *dehydrodihydroisotazettinol* (XII; R = H, R′ = OH) as prisms, m. p. 252° (decomp.), $[\alpha]_D$ −77° (*c* 1·06 in $CHCl_3$), $\nu_{max.}$ 3378 cm.$^{-1}$ (OH) (Found: C, 64·6; H, 6·1. $C_{17}H_{19}NO_5$ requires C, 64·3; H, 6·0%). The m. p. was depressed to 230—240° on admixture with dihydroisotazettinol, m. p. 250°.

The hydrochloride, prepared as described above, crystallised from methanol–ether as white needles which showed no distinct m. p., gradually becoming brown at 180—190°; they had $\nu_{max.}$ 3436, 3257 (OH), 2750—2550 (≡N^+H), and 1620 cm.$^{-1}$ (H_2O). This salt regenerated the base (XII; R = H, R′ = OH) on basification of its aqueous solution; extraction with chloroform gave this base.

The hydrochloride (20 mg.) in methanol (3 ml.) was stirred with sodium borohydride (50 mg.) for 1 hr. at room temperature. Working up as usual afforded dihydroisotazettinol (VI; R = H, R′ = R″ = OH), m. p. and mixed m. p. 246—248° (prisms from ethanol), in good yield.

Dihydrotazettinol.—Tazettinol (100 mg.) in ethanol (10 ml.) was hydrogenated in the presence of platinum oxide (100 mg.) for 5 hr. at room temperature. The mixture was filtered and evaporated, to give *dihydrotazettinol* (VI; R = R″ = OH, R′ = H) (100 mg.) as prisms, m. p. 218—220° (from ethanol), $\nu_{max.}$ 3300 and 3155 cm.$^{-1}$ (OH) (Found: C, 64·3; H, 6·6. $C_{17}H_{21}NO_5$ requires C, 63·9; H, 6·6%).

Dehydrodihydrotazettinol.—Dihydrotazettinol (50 mg.) was treated with acetic anhydride (3 ml.) and concentrated sulphuric acid (0·2 ml.) at room temperature overnight and the mixture worked up in the usual manner, to give acetyldehydrodihydrotazettinol (XII; R = OAc, R′ = H) which failed to crystallise. The product showed an infrared absorption at 1727 cm.$^{-1}$ (OAc). Hydrolysis of it in 5% ethanolic sodium hydroxide (10 ml.) under reflux for 2 hr. gave *dehydrodihydrotazettinol* (XII; R = OH, R′ = H) (15 mg.), m. p. 220—221° (from benzene), $[\alpha]_D$ −119° (*c* 0·32 in $CHCl_3$), $\nu_{max.}$ 3413 cm.$^{-1}$ (OH) (Found: C, 64·6; H, 6·1. $C_{17}H_{19}NO_5$ requires C, 64·3; H, 6·0%). The m. p. was depressed to 180—190° on admixture with dihydrotazettinol, m. p. 218—220°.

Dehydrodihydrotazettinol (50 mg.) was converted into its hydrochloride by adding concentrated hydrochloric acid to its methanolic solution and evaporating the mixture under reduced pressure. The hydrochloride thus obtained was treated with sodium borohydride (100 mg.) in methanol (2 ml.) for 1 hr. at room temperature; this yielded dihydrotazettinol (VI; R = R″ = OH, R′ = H) (35 mg.), m. p. and mixed m. p. 218—220°.

Action of Sulphuric Acid and Acetic Anhydride on Dihydrotazettine.—Dihydrotazettine (VI; R = OMe, R′ = H, R″ = OH) (0·3 g.) in acetic anhydride (5 ml.) and concentrated sulphuric acid (1 ml.) was kept for 24 hr. at room temperature. The mixture was poured into ice-water, basified with sodium hydrogen carbonate, and extracted with chloroform. The chloroform extracts were concentrated to give a gum (0·25 g.) which was chromatographed in chloroform over silica gel. The chloroform eluate gave a colourless gum which, when triturated with ethanol, gave a small quantity of acetyldehydrodihydroisotazettinol (XII; R = H, R′ = OAc), m. p. and mixed m. p. 241—242°. The mother-liquor was hydrolysed in 3% ethanolic sodium hydroxide (10 ml.), and the product (0·17 g.) was chromatographed in benzene over alumina. The first benzene eluate gave a gum (30 mg.); further elution with benzene and benzene–chloroform (1 : 1) gave dehydrodihydroisotazettinol (XII; R = H, R′ = OH) (40 mg.), m. p. and mixed m. p. 252° (decomp.), and with chloroform and chloroform–acetone (2 : 1) gave dehydrodihydrotazettinol (XII; R = OH, R′ = H) (60 mg.), m. p. and mixed m. p. 219—221°.

Action of Sulphuric Acid and Acetic Anhydride on Demethoxydihydrotazettine.—Concentrated sulphuric acid (0·5 ml.) was added to demethoxydihydrotazettine (VI; R = R′ = H, R″ = OH) (150 mg.) in acetic anhydride (5 ml.). After being kept at room temperature for 12 hr. the mixture was poured into water (20 ml.), basified with sodium hydrogen carbonate, and extracted with chloroform (2 × 20 ml.). The aqueous layer gave a positive Malachite Green test for sulphurous acid. The dried chloroform extracts were concentrated to dryness and the residue was extracted with cold 5% hydrochloric acid and benzene. The organic layer was dried and evaporated, to yield *dehydrodemethoxydihydrotazettine* XII; R = R′ = H) (90 mg.), prisms (from ethanol), m. p. 260° (decomp.), $[\alpha]_D$ −46° (*c* 1·34 in $CHCl_3$) (Found: C, 65·8; H, 6·5. $C_{17}H_{19}NO_4,\frac{1}{2}H_2O$ requires C, 65·8; H, 6·5%). The infrared spectrum showed very weak band at 3·0 μ (water of crystallisation) but no absorptions in the 5 μ region.

Dehydrodemethoxydihydrotazettine Hydrochloride.—Dry hydrogen chloride was passed into a solution of the dehydro-base (0·1 g.) in benzene (10 ml.). The precipitated hydrochloride was collected, washed repeatedly with benzene and ether, and dried over P_2O_5, to give a hygroscopic white powder, $\nu_{max.}$ 3333 and 3125 (OH and H_2O), 2740—2500 ($\equiv N^+H$), and 1626 cm.$^{-1}$ (H_2O). Basification of an aqueous solution of the hydrochloride and extraction with chloroform regenerated the base (XII; R = R′ = H).

The hydrochloride (50 mg.) and excess of sodium borohydride in methanol (10 ml.) were stirred at room temperature for 1 hr. The mixture was acidified with 5% hydrochloric acid, then evaporated to dryness; the residue was redissolved in water, washed with ether, basified with sodium carbonate, and extracted with chloroform, to afford demethoxydihydrotazettine (VI; R = R′ = H, R″ = OH), m. p. and mixed m. p. 164—166° (from ethanol), in nearly quantitative yield.

The hydrochloride (20 mg.) in 2% hydrochloric acid (10 ml.) was hydrogenated over 10% palladium–carbon (100 mg.) for 2 hr. at room temperature. After working up in the usual manner, the mixture gave demethoxydihydrotazettine (VI; R = R′ = H, R″ = OH), m. p. and mixed m. p. 164—166°.

Oxidation of Demethoxydihydrotazettine by Mercuric Acetate.—Demethoxydihydrotazettine (VI; R = R′ = H, R″ = OH) (120 mg.) and mercuric acetate (150 mg.) in 10% acetic acid (5 ml.) were heated on a water-bath for 40 min. The precipitated mercurous acetate was removed and the filtrate saturated with hydrogen sulphide and again filtered. The filtrate was basified with sodium carbonate and extracted with chloroform, which was washed with 5% hydrochloric acid to remove some unchanged starting material (50 mg.). Concentration of the washed chloroform solution gave a gum (40 mg.) which was chromatographed in benzene over neutral alumina. A benzene eluate gave the dehydro-compound (XII; R = R′ = H) (10 mg.) (prisms from ethanol), m. p. and mixed m. p. 260—261° (decomp.). A methanol eluate gave a gum (25 mg.) which was not investigated.

Oxidation of Tazettine with Mercuric Acetate.—Tazettine (200 mg.) and mercuric acetate (400 mg.) in 10% acetic acid (5 ml.) were heated on a water-bath for 10 min. The mercurous acetate which was deposited was removed and the filtrate saturated with hydrogen sulphide, to give mercury sulphide which was removed with the aid of Filter-Cel. The filtrate was basified with aqueous sodium hydroxide and extracted with chloroform. The chloroform was washed with 5% hydrochloric acid to remove some unchanged starting material (50 mg.) and then concentrated to dryness, giving a gum (100 mg.). This was chromatographed in benzene over alumina. A benzene eluate gave a trace of gum; elution with chloroform afforded a solid (60 mg.) which was rechromatographed in chloroform over silica gel. The first eluate therefrom gave a white substance (50 mg.) which crystallised from ethanol as fine needles, m. p. 226—228° (decomp.), $\lambda_{max.}$ 290 mμ, $\nu_{max.}$ 3571—3448 (OH) and 1739 cm.$^{-1}$ (CO) (Found: C, 37·8; H, 3·9; N, 2·6%). This was very soluble in chloroform, benzene, and acetone, sparingly in ethanol, methanol, and ether, and contained mercury.

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