# THE ERGOT ALKALOIDS

## III. ON LYSERGIC ACID

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The production of an acid $C_{16}H_{16}O_2N_2$, lysergic acid, by the alkaline cleavage of ergotinine has been described in a previous communication.[1] At the time we had commented on the failure to recover from the reaction mixture the base, ergine, which had been previously isolated by Smith and Timmis[2] by the action of methyl alcoholic alkali on ergotinine. We stated that this failure

> "might have been due to the fact that although it [ergine] could have been formed during the reaction in aqueous alkali it might have been further degraded to lysergic acid. In order to determine this point we have replaced ergotinine in the above procedure by ergine. Although a crystalline acid was obtained in small yield it appeared on analysis to be definitely different from lysergic acid. With the amount available it was possible to give it but preliminary study."

At the time we had also noted that ammonia was formed by the action of alkali on ergine. Since then we have found that the substance which we had obtained was a sparingly soluble sulfate of lysergic acid and the analytical figures secured with this material naturally caused confusion in the original attempt to demonstrate the formation of lysergic acid from ergine.

In the meantime, Smith has written us that since their original communication on ergine, Timmis and he[3] have found that ergine does not possess the formula $C_{17}H_{21}ON_3$ originally derived by them, but $C_{16}H_{17}ON_3$ and that it is the amide of an acid $C_{16}H_{16}O_2N_2$ doubtless identical with lysergic acid. Our recent results

[1] Jacobs, W. A., and Craig, L. C., *J. Biol. Chem.*, **104**, 547 (1934).
[2] Smith, S., and Timmis, G. M., *J. Chem. Soc.*, 763 (1932).
[3] Smith, S., and Timmis, G. M., *Nature*, **133**, 579 (1934).

confirm this view and have demonstrated definitely that the acids obtained by the action of aqueous alkali on ergotinine and ergine are identical.

In an earlier communication[4] we have reported the formation of *p*-nitrobenzoic acid and an acid $C_{14}H_9O_8N$ during the oxidation of ergotinine with nitric acid. Since the latter contains still the N-methyl group of ergotinine, it was important to determine whether lysergic acid would yield the same acid. On treatment with nitric acid under conditions similar to those formerly employed, no *p*-nitrobenzoic acid could be obtained and none of the above acid could be detected. However, a new acid has been isolated in exceedingly poor yield which on analysis gave figures from which a formula $C_{13}H_8O_8N_2$ has been derived. It still contains the N-methyl group, and the formation of a deep red color in alkaline solution, which again changed back to a pale yellow on acidifying, suggests the presence of a nitro group. The costliness of the material has prevented its further investigation.

It would appear that this acid may be related to that first obtained from ergotinine by the replacement of a carboxyl by a nitro group, and that in the formation of lysergic acid the bridge joining its precursor to the rest of the molecule in ergotinine may be ruptured in a different way under the influence of alkali from that which occurs on direct oxidation with nitric acid and which leads to a carboxyl group. At this point in lysergic acid a nitro group, instead of a carboxyl group, could be introduced. It is not excluded that the isobutyryl formic acid also produced by alkali may be involved in these considerations. We shall attempt to check such a possibility in a further study.

Since lysergic acid is a nitrogen heterocyclic derivative, it was of interest to study its behavior towards sodium in amyl alcohol. This procedure resulted in the formation of a new crystalline substance which still possesses acid and basic properties and is more stable than lysergic acid itself. From the analysis it appears to be a *dihydrolysergic acid*, $C_{16}H_{18}O_2N_2$. This formula was confirmed by the formation of a *methyl ester* by the use of methyl alcoholic hydrogen chloride.

At this point it should be mentioned that the usual tests for a

[4] Jacobs, W. A., *J. Biol. Chem.*, **97**, 739 (1932).

primary or secondary amine grouping made on the ester of lysergic acid have failed. Attempts to acetylate the ester or combine it with phenyl isothiocyanate were negative. When boiled in toluene solution with metallic sodium, the compound did not appear to form a sodium derivative. It seemed possible that in the formation of the dihydro derivative reduction of one of the double bonds in the ester might produce a secondary amine, but this did not prove to be the case. The ester of dihydrolysergic acid does not react with phenyl isothiocyanate or acetyl chloride, and boiling with acetic anhydride gave only unchanged ester. However, the methyl ester of lysergic acid gave methane easily in the Zerewitinoff test for active hydrogen.

Finally, since lysergic acid contains one carboxyl group, it was of interest to investigate its behavior on dry distillation. When heated in a sublimation apparatus at 0.2 mm., a volatile base began to sublime when the bath reached 200° and attained a maximum at 250°. This base formed yellow leaflets and proved to be very unstable, so that during the attempts to recrystallize it decomposition interfered. Although the analytical figures were not conclusive, the formation of the base by loss of $CO_2$ was suggested. Since the amount of material available is so limited, a further study of this substance has been deferred.

Continued investigation of the degradation of lysergic acid is in progress.

Thus far it is apparent that the ergotinine molecule is made up of distinct portions, and it is probable that in the interconnection of some of them amide linkages play a rôle. These portions are represented by lysergic acid, isobutyryl formic acid, and perhaps by a benzyl or related grouping and a still undetermined nitrogen heterocyclic group. The fact that these alkaloids on decomposition with alkali liberate apparently only 1 mole of ammonia (Soltys[5]) raises the question whether the inferred labile amide linkage occurs in the alkaloid itself as a $CO \cdot NH_2$ group or a —$CO \cdot N \cdot H \cdot CO$— group. Two different amides have been obtained by different procedures from ergotinine, namely ergine and isobutyryl formamide. It is obvious that the $CO \cdot NH_2$ groups in both of these cannot occur as $CO \cdot NH_2$ groups together in the alkaloid molecule.

[5] Soltys, A., *Ber. chem. Ges.*, **65**, 553 (1932).

## EXPERIMENTAL

*Ergine*—Ergine was prepared according to Smith and Timmis[2] and recrystallized from methyl alcohol.

The analysis was made on the air-dried substance.

| | | |
|---|---|---|
| $C_{16}H_{17}ON_3 \cdot CH_3OH$. | Calculated. | C 68.18, H 7.07, N 14.04 |
| | Found. | " 68.40, " 6.99 |
| | " | " 68.49, " 6.88 |
| | " | N 14.12 |
| | " | " 14.20 |

*Hydrolysis of Ergine*—100 mg. of ergine were treated with 3 cc. of N sodium hydroxide and the mixture was heated on the steam bath in a nitrogen atmosphere for 80 minutes. The nitrogen gas from the reaction was passed through 10 cc. of 0.1 N sulfuric acid. On back titration 3.64 mg. of liberated ammonia were determined. The ammonia formed was identified by acylation with *p*-nitrobenzoyl chloride. The *p*-nitrobenzamide obtained melted at 198°.

The aqueous alkaline hydrolysate contained 20 mg. of unchanged ergine as a suspension which was filtered off through a sintered glass filter. The filtrate was neutralized to Congo red with sulfuric acid. After chilling and filtering, 80 mg. of dark colored crystalline material were obtained. This was suspended in 2 cc. of methyl alcohol and treated with 2 drops of ammonium hydroxide. The filtrate on evaporation to dryness under diminished pressure yielded a residue which was digested a short time with 1 cc. of methyl alcohol and filtered. The undissolved portion was boiled with 7 cc. of water and filtered hot. The filtrate on cooling in ice gave 20 mg. of lysergic acid which melted at 238°.

| | | |
|---|---|---|
| $C_{16}H_{16}O_2N_2$. | Calculated. | C 71.69, H 6.00, N 10.45 |
| | Found. | " 71.66, " 6.07 |
| | " | " 71.82, " 6.07 |
| | " | N 10.58 |
| | " | " 10.63 |

For confirmation of its identity the methyl ester was prepared with diazomethane. After recrystallization from benzene it melted at 168°.

*Lysergic Acid Hydrochloride*—100 mg. of lysergic acid were dissolved in 4 cc. of dilute hydrochloric acid. After cooling the crystals which separated were collected with dilute HCl. When

recrystallized from methyl alcohol the product melted with decomposition at 208–210°, depending somewhat on the rate of heating. It was dried for analysis at 120° and 2 mm.

| | | |
|---|---|---|
| $C_{16}H_{16}O_2N_2 \cdot HCl$. | Calculated. | C 63.03, H 5.63, N 9.19, Cl 11.62 |
| | Found. | " 62.93, " 5.48, " 9.31, " 11.15 |

*Lysergic Acid Sulfate*—100 mg. of lysergic acid were dissolved in 8 cc. of hot water and a slight excess of dilute $H_2SO_4$ was added. Upon cooling the sulfate separated as leaflets. It was recrystallized from 6 cc. of hot water. It melts with decomposition at about 220°, depending somewhat upon the rate of heating. It was dried for analysis at 120° and 2 mm.

| | | |
|---|---|---|
| $(C_{16}H_{16}O_2N_2)_2 \cdot H_2SO_4$. | Calculated. | C 60.54, H 5.40, N 8.82 |
| | Found. | " 60.97, " 5.47, " 8.76 |

*Oxidation of Lysergic Acid with Nitric Acid*—A number of experiments were performed but the following gave the best yield of the crystalline product.

400 mg. of lysergic acid were treated with 16 cc. of $HNO_3$ (sp. gr. 1.4) and the solution was placed on the steam bath for 20 hours. A clear red solution resulted, which was evaporated to dryness on the steam bath under reduced pressure. 10 cc. of water were added and the evaporation was repeated. This procedure was repeated twice in order to remove as much of the nitric acid as possible. The solid residue was boiled with 20 cc. of water and filtered after cooling. The filtrate was concentrated to approximately 1 cc. On long standing, leaflets gradually separated. The substance was collected with water. The yield was 10 to 12 mg.

The substance, which proved to be an acid, was rather slightly soluble in water. Recrystallization was accomplished by dissolving in a rather large volume of boiling water and concentrating to a small volume. It separates in small yellow rhombs which do not melt at 350°. The alkaline solution is of a brownish red color which again becomes pale yellow on acidifying.

In general properties it resembles closely the acid $C_{14}H_9O_8N$ obtained from ergotinine with the exception of the deep red color in alkaline solution.

| | | |
|---|---|---|
| $C_{13}H_8O_8N_2$. | Calculated. | C 48.75, H 2.51, N 8.75, $NCH_3$ 9.05 |
| | Found. | " 48.80, " 2.41 |
| | " | " 48.80, " 2.70 |
| | " | N 9.04 |
| | " | $NCH_3$ 7.94 |

*Dihydrolysergic Acid*—200 mg. of crystalline lysergic acid were suspended in 8 cc. of amyl alcohol and 400 mg. of sodium were added. The mixture was heated to the boiling point of the alcohol and shaken vigorously during the reduction. It was kept at this point until the sodium was dissolved. After cooling, 8 cc. of ether and then 4 cc. of water were added. The mixture was saturated with carbon dioxide. It was evaporated to dryness under reduced pressure and 10 cc. of ethyl alcohol were added. After reconcentration the residue was extracted with 15 cc. of hot alcohol. The alcoholic extract yielded a residue which was dissolved in 5 cc. of water. The solution was carefully treated with acetic acid which caused precipitation of dihydrolysergic acid. After collection with water 120 mg. of material were obtained which melted at 329°.

When recrystallized from water it melts with decomposition at approximately 336°, depending somewhat on the rate of heating. The dihydro acid is a more stable compound than lysergic acid and colors only slightly in the light. It is less soluble in water than lysergic acid.

$$[\alpha]_D^{20} = -88.0° \ (c = 0.5 \text{ in pyridine})$$

| | | |
|---|---|---|
| $C_{16}H_{18}O_2N_2$. | Calculated. | C 71.06, H 6.72, N 10.36 |
| | Found. | " 71.24, " 6.68, " 9.78 |
| | " | " 70.85, " 6.74 |

*Dihydrolysergic Acid Methyl Ester*—When an attempt was made to prepare the ester with diazomethane in the same way in which lysergic acid methyl ester was prepared, the yield was very poor. A considerable amount of an amorphous by-product was formed. Methyl alcoholic hydrochloric acid proved to be the better reagent.

50 mg. of dihydrolysergic acid were dissolved in 10 cc. of 4 per cent methyl alcoholic hydrogen chloride and allowed to stand at room temperature for 2 days. The solution was evaporated to dryness under reduced pressure on the steam bath and the residue was dissolved in 1 cc. of water. After precipitation with am-

monium hydroxide the solid was extracted with ether. The ether residue was recrystallized from benzene. The ester melts at 182°.

It is insoluble in water but soluble in the usual organic solvents. It crystallizes from benzene in broad leaves but can also be recrystallized from dilute alcohol.

| | | |
|---|---|---|
| $C_{17}H_{20}O_3N_2$. | Calculated. | C 71.78, H 7.10, N 9.85 |
| | Found. | " 71.58, " 7.03, " 9.78 |
| | " | " 71.81, " 6.84 |