[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, UNIVERSITY OF CALIFORNIA, LOS ANGELES]

Amine Oxides. I. Gramine Oxide[1]

BY DAVID W. HENRY AND EDWARD LEETE[2]

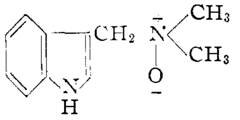
RECEIVED FEBRUARY 15, 1957

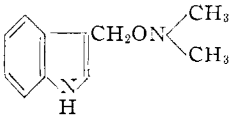
Treatment of gramine with hydrogen peroxide yields gramine oxide (I). On heating, gramine oxide rearranges to O-skatyl-N,N-dimethylhydroxylamine (II). Nucleophilic reagents (amines, alkoxides, hydroxide, cyanide and the nitromethyl anion) react with gramine oxide yielding the corresponding skatyl derivatives and N,N-dimethylhydroxylamine. Gramine oxide decomposes in water at 100° yielding 3,3'-diindolylmethane, formaldehyde and polymeric material. Acids decompose the oxide yielding amorphous material and N,N-dimethylhydroxylamine.

Gramine (3-dimethylaminomethylindole) is found in the leaves of germinating barley, but one month after germination all the gramine has disappeared from the plant.[3] We considered that gramine oxide (I) would be a plausible metabolite of gramine. This communication is an account of the synthesis and properties of gramine oxide.

Aliphatic amine oxides are usually hygroscopic oils[4,5]; however, we found that the treatment of gramine in ethanol solution with hydrogen peroxide yielded non-hygroscopic crystals of gramine oxide which crystallized with one mole of hydrogen peroxide.[6] When the excess hydrogen peroxide was decomposed after oxidation of gramine, the gramine oxide did not crystallize out and removal of the ethanol *in vacuo* yielded a viscous oil readily soluble in water.

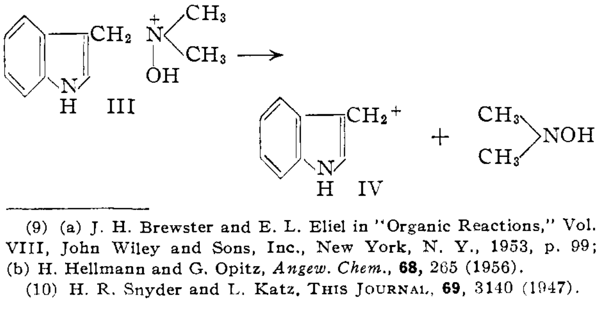
On heating gramine oxide to 125° vigorous decomposition occurred and an isomer, insoluble in water, was isolated from the residue. Cope and Towle[7] showed that N,N-dimethylbenzylamine oxide rearranged on heating to O-benzyl-N,N-dimethylhydroxylamine. By analogy, the rearrangement product of gramine oxide was considered to be O-skatyl-N,N-dimethylhydroxylamine (II). The ultraviolet spectrum of the rearrangement product was consistent with this structure, being almost identical with the spectrum of 3-methoxymethylindole. Furthermore, treatment of the rearrangement product with lithium aluminum hydride yielded skatole. It has been shown previously[8a,b] that 3-alkoxymethylindoles undergo ready hydrogenolysis to skatole with lithium aluminum hydride. A high yield (61%) of the rearrangement product was obtained on refluxing gramine oxide in acetonitrile containing a trace of pyridine. Synthesis of II was attempted by treating gramine methiodide with a solution of sodium in N,N-dimethylhydroxylamine. Trimethylamine was displaced but the product was gramine oxide, a C–N bond being formed rather than the desired C–O bond. The reaction mixture also contained gramine which was presumably produced by reduction of gramine oxide with the excess N,N-dimethylhydroxylamine.


I


II

Gramine and its quaternary salts are well known as alkylating agents[9a,b] eliminating dimethylamine or a tertiary amine, respectively, on reaction with nucleophilic reagents. Gramine oxide reacted similarly eliminating N,N-dimethylhydroxylamine. Thus treatment of gramine oxide with piperidine yielded 3-N-piperidinomethylindole. Other secondary amines reacted similarly. Gramine oxide reacted with sodium alkoxides, sodium hydroxide and sodium cyanide to yield 3-alkoxy-, 3-hydroxy- and 3-cyano-methylindole, respectively. Gramine oxide reacted with nitromethane in the presence of sodium ethoxide yielding 3-(2-nitroethyl)-indole. Snyder and Katz[10] obtained only di-(3-indolylmethyl)-nitromethane by reaction of gramine with nitromethane in the presence of sodium hydroxide. The 3-alkoxymethylindoles were also obtained by refluxing gramine oxide in the appropriate alcohol. Some of the rearrangement product II was produced at the same time.

On warming a solution of gramine oxide in water at 100° polymeric material, 3,3'-diindolylmethane and formaldehyde were produced. The initial product of the reaction was probably 3-hydroxymethylindole which undergoes self condensation and elimination of formaldehyde on heating.[8a] Dilute hydrochloric acid decomposed gramine oxide yielding N,N-dimethylhydroxylamine and amorphous material. The protonated gramine oxide (III) presumably eliminates N,N-dimethylhydroxylamine yielding the skatylcarbonium ion (IV) which then reacts with water or undergoes self condensation.


III IV

---

[1] This work was supported in part by a grant from the Research Corporation, New York.

[2] To whom correspondence concerning this article should be addressed.

[3] K. Brandt, H. v. Euler, H. Hellström and N. Löfgren, *Z. physiol. Chem.*, **235**, 37 (1935).

[4] A. C. Cope and H. H. Lee, THIS JOURNAL, **79**, 964 (1957).

[5] M. S. Fish, N.M. Johnson and E. C. Horning, *ibid.*, **77**, 5892 (1955).

[6] There are several reported examples of the crystallization of amine oxides with hydrogen peroxide; *cf.* G. M. Bennett and E. Glynn, *J. Chem. Soc.*, 211 (1950); M. Oesterlin, *Ber.*, **76**, 224 (1943); M. Polonovski and M. Polonovski, *Bull. soc. chim.*, **39**, 1147 (1926).

[7] A. C. Cope and P. H. Towle, THIS JOURNAL, **71**, 3423 (1949).

[8] (a) E. Leete and L. Marion, *Can. J. Chem.*, **31**, 775 (1953); (b) E. D. Rossiter and J. E. Saxton, *J. Chem. Soc.*, 3654 (1953).

[9] (a) J. H. Brewster and E. L. Eliel in "Organic Reactions," Vol. VIII, John Wiley and Sons, Inc., New York, N. Y., 1953, p. 99; (b) H. Hellmann and G. Opitz, *Angew. Chem.*, **68**, 265 (1956).

[10] H. R. Snyder and L. Katz, THIS JOURNAL, **69**, 3140 (1947).

Experimental[11]

Gramine Oxide (I). (a).—A suspension of gramine (17.4 g., 0.1 mole) in ethanol (40 ml.) was treated with 28.2 ml. of 30% aqueous hydrogen peroxide (0.25 mole). Heat was evolved and all the gramine dissolved. On cooling, the pale brown solution deposited colorless prismatic needles of gramine oxide (18.5 g.) containing one mole of hydrogen peroxide of crystallization, m.p. 135–136° (rapid heating), 121–122° (slow heating) with decomposition in both cases. Recrystallization from ethanol did not remove the hydrogen peroxide of crystallization. If the excess hydrogen peroxide was decomposed by addition of a small amount of 10% platinum-on-charcoal no crystallization occurred. However the addition of hydrogen peroxide to this solution caused separation of the gramine oxide–hydrogen peroxide derivative.

Anal. Calcd. for $C_{11}H_{14}N_2O{\cdot}H_2O_2$: C, 58.91; H, 7.19; N, 12.49; H_2O_2, 15.20. Found: C, 59.13; H, 6.98; N, 12.56; H_2O_2 (by iodometric titration), 14.9.

Specimens several months old were pale brown and contained a slightly lower percentage of hydrogen peroxide.

The ultraviolet spectrum of the gramine oxide–hydrogen peroxide derivative is almost identical with that of gramine, with three maxima at 272 mμ (ε 6500), 280 mμ (ε 6720) and 287.5 mμ (ε 5950). The infrared spectrum has a broad band at 2500 cm.$^{-1}$ which is absent in the infrared spectrum of gramine. This band is possibly assignable to the O–H stretching of $\overset{+}{N}$–O–H which may exist in the hydrogen peroxide derivative. Strychnine oxide monohydrate[12] has no absorption in this region of the spectrum, but has a broad band at 3260 cm.$^{-1}$. N,N-Dimethylhydroxylamine picrate has an absorption band at 2700 cm.$^{-1}$.

(b).—Gramine methiodide[13] (1.0 g.) was added to a solution of sodium (80 mg.) in 3.3 g. of N,N-dimethylhydroxylamine[14] at room temperature. After 19 hours the excess N,N-dimethylhydroxylamine was removed *in vacuo* and the semi-solid residue extracted with 20 ml. of water. The material insoluble in water was gramine (0.24 g., 44%). The aqueous filtrate was shown to contain gramine oxide by paper chromatography. The solutions used for development of the paper chromatograms were (numbers in parentheses are parts by volume): 1, 15% aqueous ammonia; 2, acetone (6), 7.5% aqueous ammonia (5); 3, triethylamine (2), *n*-propyl alcohol (1.3), water (1). Gramine oxide had R_f values of 0.78, 0.79 and 0.89; gramine had R_f values of 0.72, 0.76 and 1.0; and gramine methiodide had R_f values of 0.56, 0.61 and 1.0, in solutions 1, 2 and 3, respectively. The indole derivatives were detected on the paper chromatograms by spraying with Millon reagent.

Reduction of Gramine Oxide.—In this and all subsequent reactions of gramine oxide the hydrogen peroxide of crystallization was decomposed by the addition of 10% platinum-on-charcoal to a solution of a weighed amount of the gramine oxide–hydrogen peroxide derivative. When effervescence of oxygen had ceased, the solution was filtered and reactions were carried out on the filtrate. All weights of gramine oxide refer to the hydrogen peroxide derivative. Gramine oxide (1.0 g.) in 20 ml. of water was treated with 5.0 g. of zinc dust and 10 ml. of glacial acetic acid. After stirring for 30 minutes the suspension was filtered and the filtrate added to potassium hydroxide solution. The white precipitate which separated was extracted with ether. On evaporation of the ether solution colorless plates of gramine (0.72 g., 92%), m.p. 133°, were obtained.

Rearrangement of Gramine Oxide. (a) With No Solvent.—Gramine oxide (1.0 g.) in 20 ml. of methanol was evaporated at 10° *in vacuo* to leave a pale brown viscous sirup. On warming to 125° vigorous decomposition took place. After 10 minutes the reaction ceased and the tarry residue was extracted with ether. The ether extract was concentrated to 10 ml., diluted with 20 ml. of pentane and chromatographed on alumina of low activity. The column was eluted first with a mixture of ether–pentane (1:2 by vol.), then with 1:1 ether–pentane and finally with pure ether. The fractions from the 1:1 eluent yielded 0.15 g. of crystalline material. This was recrystallized from pentane yielding colorless rhombic crystals of O-skatyl-N,N-dimethylhydroxylamine (II), m.p. 93–94°.

Anal. Calcd. for $C_{11}H_{14}N_2O$: C, 69.44; H, 7.42; N, 14.73. Found: C, 69.32; H, 7.56; N, 14.28.

The ultraviolet spectrum of II has maxima at 272 mμ (ε 6000), 280 mμ (ε 6100) and 288.5 mμ (ε 5300). The pure ether eluent yielded 0.40 g. of crystals which after sublimation did not depress the melting point of gramine.

(b) **In Organic Solvents.**—Gramine oxide was refluxed in a variety of solvents for one hour. The solutions were filtered and the solvents removed *in vacuo*. The residues obtained were chromatographed on alumina as before. The percentage yield of II obtained in the following solvents is denoted in parentheses: toluene (16), dimethylformamide (25), dioxane (37), acetonitrile containing a trace of pyridine (61).

Reduction of O-Skatyl-N,N-dimethylhydroxylamine.—Compound II (42 mg.) was added to an ether solution (20 ml.) of an equal weight of lithium aluminum hydride and refluxed for one hour. Excess hydride was decomposed by the addition of wet ether and evaporation of the filtered ether solution yielded a crystalline residue having the odor of skatole. Addition of 1,3,5-trinitrobenzene to a methanolic solution of the residue yielded orange needles of the 1,3,5-trinitrobenzene derivative of skatole, m.p. 184–185°, (reported[15] 185°), not depressed on admixture with an authentic specimen.

Anal. Calcd. for $C_9H_9N{\cdot}C_6H_3N_3O_6$: C, 52.33; H, 3.51. Found: C, 52.08; H, 3.50.

Reaction of Gramine Oxide with Nucleophiles. (a) **Secondary Amines.**—Gramine oxide (2.0 g.) was refluxed with 40 ml. of piperidine for 3 hours. The residue remaining after removal of the piperidine *in vacuo* was crystallized from ethanol (charcoal) to yield colorless plates of 3-N-piperidinomethylindole (1.59 g., 83%), m.p. 158–159° (reported[16] 161°). Addition of piperidine to an aqueous solution of gramine oxide at room temperature gave no visible reaction. However, on warming to 100° colorless plates of 3-N-piperidinomethylindole separated. This is in contrast to gramine methiodide which gave an immediate precipitate of the piperidino derivative on addition of piperidine to its aqueous solution at room temperature.

Gramine oxide in refluxing diethylamine was converted to 3-diethylaminomethylindole, m.p. 102.5–104° (reported[17] 105–106°. The m.p. of 165° reported by Kühn and Stein[16] erroneously for the free base is apparently that of the hydrochloride). Refluxing gramine oxide in dimethylamine using a Dry Ice-cooled condenser yielded gramine. Treatment of gramine oxide with morpholine at 100° yielded 3-N-morpholinomethylindole, m.p. 119–121° (reported[18] 121°).

N-Methylaniline and gramine oxide at 100° yielded 3-N-methyl-N-phenylaminomethylindole (58%), m.p. 85–86.5° (reported[19] 87–88°).

(b) **Sodium Alkoxides.**—Gramine oxide (1.6 g.) in 20 ml. of methanol was added to a solution of sodium (0.17 g.) in 20 ml. of methanol. The solution was refluxed for one minute and then allowed to cool to room temperature. Sodium bicarbonate (0.65 g.) and 0.2 ml. of water were added and the mixture evaporated *in vacuo*. The vapors were condensed in a Dry Ice trap. Treatment of this distillate with picric acid yielded yellow crystals which after several recrystallizations from ethanol gave yellow needles of N,N-dimethylhydroxylamine picrate, m.p. 160–161°, not depressed on admixture with an authentic specimen.

Anal. Calcd. for $C_2H_7NO{\cdot}C_6H_3N_3O_7$: C, 33.11; H, 3.47. Found: C, 33.33; H, 3.57.

---

[11] All melting points are corrected. Ultraviolet spectra were determined on a Cary recording spectrophotometer, model 11 PMS, in 95% ethanol. Infrared spectra were determined on a Perkin–Elmer recording spectrophotometer, model 21, as dispersions in potassium bromide unless otherwise stated. The spectra were determined by Mr. R. L. Meeker and Mr. H. Uehara. We are indebted to Miss Heather King for analyses.

[12] A. S. Bailey and R. Robinson, *J. Chem. Soc.*, 703 (1948).

[13] T. A. Geissman and A. Armen, THIS JOURNAL, **74**, 3916 (1952).

[14] M. A. T. Rogers, *J. Chem. Soc.*, 769 (1955).

[15] E. Hertel, *Ann.*, **451**, 179 (1927).

[16] H. Kühn and O. Stein, *Ber.*, **70**, 567 (1937).

[17] N. Kühn and O. Stein, German Patent 673,949; *C. A.*, **33**, 6484 (1939).

[18] A. M. Akkerman, D. K. de Jongh and H. Veldstra, *Rec. trav. chim.*, **70**, 899 (1951).

[19] J. Thesing and H. Mayer, *Ber.*, **87**, 1084 (1954).

The residue remaining after removal of the methanol and N,N-dimethylhydroxylamine was extracted with ether, dried over potassium carbonate and the solution evaporated to yield a crystalline residue. This residue was recrystallized from pentane yielding colorless plates of 3-methoxymethylindole (0.72 g., 63%), m.p. 97–98° (reported[13] 99–100°).

Sodium ethoxide and gramine oxide yielded 3-ethoxymethylindole (59%), m.p. 62–63° (reported[13] 63–64°).

(c) **Alcohols.**—Gramine oxide (1.0 g.) was refluxed with 25 ml. of isobutyl alcohol for 2 hours. Removal of the solvent yielded a pale orange oil which was chromatographed on alumina. The eluting solvents varied from ether–pentane (1:3) to pure ether. The first fractions were liquid and were distilled (120° (0.0005 mm.)) to yield 3-isobutoxymethylindole, a colorless oil, n^{25}_{D} 1.5574.

Anal. Calcd. for $C_{13}H_{17}NO$: C, 76.81; H, 8.43; N, 6.89. Found: C, 76.47; H, 8.50; N, 6.84.

The 1,3,5-trinitrobenzene derivative crystallized in yellow prismatic microscopic needles, m.p. 103–104°.

Anal. Calcd. for $C_{13}H_{17}NO \cdot C_6H_3N_3O_6$: C, 54.80; H, 4.84. Found: C, 54.91 H, 4.89.

3-Isobutoxymethylindole also was obtained in 44% yield by the treatment of gramine with ethyl iodide and sodium isobutoxide. Later fractions from the alumina column were semi-solid and sublimation (80° (0.0002 mm.)) followed by crystallization from pentane yielded colorless rhombic crystals of O-skatyl-N,N-dimethylhydroxylamine, m.p. 93–94°.

On refluxing gramine oxide in ethanol, 3-ethoxymethylindole and II were obtained along with material insoluble in ether, presumably polymeric.

(d) **Sodium Hydroxide.**—Gramine oxide (1.0 g.) in 50 ml. of water was added dropwise to a rapidly stirred, refluxing mixture of 50 ml. of 10% aqueous sodium hydroxide and 50 ml. of ether. After 4 hours the ether layer was separated, dried and evaporated. The residue (0.11 g.) was crystallized from pentane to yield colorless plates of 3-hydroxymethylindole, m.p. 99–100° (reported[20] 100–101°). If the reaction was allowed to proceed overnight, more 3-hydroxymethylindole was obtainable from the ether layer.

(e) **Water.**—Gramine oxide (1.0 g.) in 20 ml. of water was heated at 100° for 20 hours. A white precipitate was produced. An ether extract of this mixture was chromatographed on alumina eluting with ether–pentane (1:4). The main fraction was crystallized from benzene–petroleum ether yielding colorless plates of 3,3′-diindolylmethane (0.10 g.), m.p. 163–164° (reported[8a] 163–164°). There was much material insoluble in ether, presumably polymeric. In a duplicate experiment, the aqueous solution at the end of the reaction was treated with dimedone yielding formaldehyde-dimedone derivative, m.p. 190–191° (58%), not depressed on admixture with an authentic specimen.

(f) **Sodium Cyanide.**—Gramine oxide (1.0 g.) in 25 ml. of water was stirred rapidly with a mixture of 25 ml. of toluene, 10 g. of sodium cyanide and 25 ml. of a saturated solution of sodium cyanide at 100° for 1.5 hours. The toluene layer was separated and dried over potassium carbonate. The liquid residue remaining after removal of the toluene *in vacuo* was dissolved in methanol and treated with 1,3,5-trinitrobenzene yielding orange needles of the 1,3,5-trinitrobenzene derivative of 3-cyanomethylindole (1.36 g., 83%), m.p. 135.5–137°.

Anal. Calcd. for $C_{10}H_8N_2 \cdot C_6H_3N_3O_6$: C, 52.03; H, 3.00. Found: C, 52.01; H, 3.12.

When gramine oxide was treated with methanolic sodium cyanide a mixture of 3-cyanomethylindole and 3-methoxymethylindole was obtained.

(g) **Nitromethane.**—Gramine oxide (1.0 g.) in 15 ml. of nitromethane was treated with a solution of sodium (0.10 g.) in 2 ml. of ethanol and refluxed with stirring for one hour. Water (0.2 ml.) was then added and the excess nitromethane removed *in vacuo*. The residue was extracted with ether, diluted with pentane and chromatographed on alumina of low activity. The major fraction from the chromatogram was crystallized from pentane, yielding colorless plates of 3-(2-nitroethyl)-indole (0.27 g., 31%), m.p. 53.5–54° (reported[21] as existing in dimorphic forms, m.p. 56.5–57° and 68–68.5°). The infrared spectrum of 3-(2-nitroethyl)-indole in chloroform has absorption bands at 1546 and 1380 cm.$^{-1}$ assignable to the NO_2 group.

(h) **Hydrochloric Acid.**—Gramine oxide (1.0 g.) in 15 ml. of water was treated with 1 ml. of 4 *N* hydrochloric acid and heated at 100° for 2 hours. The white amorphous precipitate which was obtained was filtered off, washed with water and dried in a desiccator over sodium hydroxide.

Anal. Found: C, 79.46; H, 5.47; N, 6.78. Calcd. for C_9H_7N (the empirical formula of the polymer obtained by the action of acid on 3-hydroxymethylindole[8a]): C, 83.69; H, 5.46; N, 10.85.

The aqueous filtrate was evaporated to dryness yielding a colorless sirup which was treated with picric acid giving a yellow precipitate. After several recrystallizations from ethanol, yellow needles of N,N-dimethylhydroxylamine picrate separated, m.p. 160–161°, not depressed on admixture with an authentic specimen.

Attempts to make a picrate of gramine oxide by addition of ethanolic picric acid to an ethanol solution of the oxide resulted in the formation of resinous material.

LOS ANGELES 24, CALIF.

---

(20) J. Thesing, *Ber.*, **87**, 692 (1954).

(21) W. E. Noland and P. J. Hartmann, THIS JOURNAL, **76**, 3227 (1954).

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY OF THE UNIVERSITY OF WISCONSIN]

The Alkaloids of *Rauwolfia*. Structure of Raunescine and Isoraunescine

BY EUGENE E. VAN TAMELEN AND CHARLES W. TAYLOR

RECEIVED MARCH 30, 1957

The technique of gradient-elution chromatography has been applied to a mixture of minor alkaloids derived from *Rauwolfia tetraphylla* L. The generalized structure VII for two of the components, raunescine and isoraunescine, is supported by the conversion of deserpidine to raunescic acid (= isoraunescic acid), supplemented by evidence for the stereochemistry at C-3 and C-16.

Rauwolfia tetraphylla L. serves as an important commercial source of the hypotensive and tranquilizing agent reserpine, which is accompanied in this plant by a profusion of other indole alkaloids.[1,2] There became available to us substantial quantities of the alkaloid mixture[3,4] from which reserpine and certain of the other bases had been largely removed, thus affording the opportunity to scru-

---

(1) R. E. Woodson, Jr., H. W. Youngken, E. Schlittler and J. A. Schneider, "Rauwolfia: Botany, Pharmacognosy, Chemistry and Pharmacology," Little, Brown and Co., Boston, Mass., 1957, pp. 54–55.

(2) (a) C. Djerassi, M. Gorman, S. C. Pakrashi and R. B. Woodward, THIS JOURNAL, **78**, 1259 (1956); (b) M. W. Klohs, F. Keller, R. E. Williams and G. W. Kusserow, *Chemistry & Industry*, 187 (1956).

(3) Reserpine-depleted weakly basic alkaloids, supplied by S. B. Penick and Co., New York, N. Y.

(4) Although *R. canescens* L. and *R. heterophylla* Roem. and Schult. are botanically indistinguishable (ref. 1, p. 11) from *R. tetraphylla* L., the first term has been generally applied to Indian roots and the second, to the American alkaloid source. In this investigation a mixture from material classified as "*heterophylla*" was used.