MO Calculations.—In the simple LCAO–MO calculations of π-bond orders, the recommended figures for the coulomb and the exchange integral parameters were used, and a factor of 0.10 was employed for the "auxiliary inductive parameter" of the carbon atoms vicinal to heteroatoms.[16] Actual calculations were made with a Model NEAC-2230 electronic computor at the Computor Center of Tohoku University.

(16) A. Streitwieser, Jr., "Molecular Orbital Theory for Organic Chemists," John Wiley and Sons, Inc., New York, 1961, p. 135.

Purine N-Oxides. XVII. The Oxidation of Guanine at Position 7[1]

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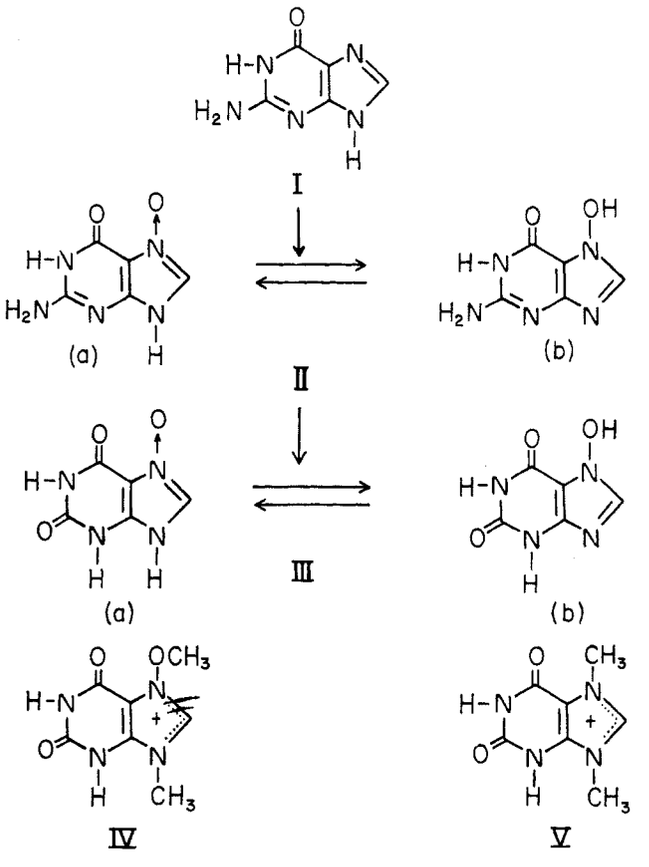
The 7-nitrogen of guanine was oxidized by trifluoroperoxyacetic acid. Upon hydrolysis only the corresponding xanthine derivative results. The position of the oxygen atom was proven by hydrolysis to N-hydroxyglycine.

The oxidation of a nitrogen of guanine was accomplished[2] and the product, designated there as guanine x,N-oxide, and its hydrolysis product, designated as xanthine x,N-oxide, were found to be potent carcinogenic agents.[2] We present here the synthesis, structure, and the characterization of several derivatives of the oxidized guanine.

A mixture of trifluoroacetic acid and hydrogen peroxide was used to oxidize guanine (I). Oxidations of purines with acetic acid–hydrogen peroxide mixtures have been successful when amino[3] or methyl substituents,[4] but not oxygen, are present on the 2- or 6-carbons of the pyrimidine ring, although the presence of oxygen on the imidazole carbon does not interfere.[4,5] Many unsuccessful attempts have been made, including at higher temperatures and with the presence of minimal amounts of water, to oxidize guanine, xanthine, and similar purines. The failures could be attributed to either the existence of the oxypurines in the lactam form, or to their insolubility in acetic acid–hydrogen peroxide mixtures. However, in mineral acid solutions guanine is readily oxidized by hydrogen peroxide to products without a specific absorption in the ultraviolet region.

Despite its insolubility in acetic acid, guanine is quite soluble in trifluoroacetic acid. From a hot solution complexes with trifluoroacetic acid can be obtained; there is no evidence that these are acyl derivatives. With hydrogen peroxide present there results a facile oxidation to one major product. Upon recrystallization from hydrochloric acid, this product is obtained as a hemihydrochloride, and from sodium hydroxide the anhydrous free base, $C_5H_5N_5O_2$ (II), is precipitated by acetic acid. With Raney nickel II consumed 1 mole of hydrogen to yield guanine.[2]

This guanine oxide is stable in moderate concentrations of hydrochloric acid, in contrast to adenine 1-N-oxide which is easily hydrolyzed in 0.1 *N* HCl with the loss of C-2.[6] With hot 6 *N* HCl overnight, conditions comparable to the hydrolysis of guanine to xanthine,[7] II yields a xanthine oxide (III). This separates as a hydrochloride hydrate from 6 *N* HCl and as a hemihydrochloride from 2 *N* HCl. That the xanthine N-oxide is more basic than xanthine is shown by the fact that the latter forms a hydrochloride only in concentrated hydrochloric acid, whereas the xanthine hydrochloride yields free xanthine when recrystallized from 2 *N* HCl. A mono- and a dihydrate of the xanthine N-oxide are obtained from either neutral or acetic acid solutions.



Xanthine oxide is but slowly reduced to xanthine by Raney nickel and hydrogen,[2] in analogy to the difficult reduction of xanthine 3-N-oxide,[8] and of 1-hydroxy-7-benzylxanthine.[9]

(1) This investigation was supported in part by Public Health Service Research Grant No. CA-03190-08, and from the Atomic Energy Commission, Contract AT(30-1)-910.

(2) G. B. Brown, K. Sugiura, and R. M. Cresswell, *Cancer Res.*, in press.

(3) M. A. Stevens, D. I. Magrath, H. W. Smith, and G. B. Brown, *J. Am. Chem. Soc.*, **80**, 2755 (1958).

(4) M. A. Stevens, A. Giner-Sorolla, H. W. Smith, and G. B. Brown, *J. Org. Chem.*, **27**, 567 (1962).

(5) G. B. Brown, M. A. Stevens, and H. W. Smith, *J. Biol. Chem.*, **233**, 1513 (1958).

(6) M. A. Stevens and G. B. Brown, *J. Am. Chem. Soc.*, **80**, 2759 (1958).

(7) E. Fischer, *Ber.*, **43**, 805 (1910).

(8) R. M. Cresswell, H. K. Maurer, T. Strauss, and G. B. Brown, *J. Org. Chem.*, **30**, 408 (1965).

(9) L. Bauer, *J. Heterocyclic Chem.*, **1**, 275 (1964).

TABLE I

Compd.	λ_{max}, mμ (ε × 10⁻³)			λ_{min}, mμ (ε × 10⁻³)	pH	R_f in solvents[a] A	B	C
Xanthine 7-N-oxide	297 (8.42)	224 (24)		267 (4.1)	10–12	0.65[c]	0.54	0.28
	272 (~6.0)	212 (~18)		237 (~4.3)[b]	7			
	272 (~10.2)			244 (~3.3)	3–4			
Dimethyl derivative from xanthine 7-N-oxide	287 (9.5)	243 (5.1)		261 (2.1)	*Ca.* 14	0.66	0.66	0.47
	294 (11.7)	242 (9.7)		265 (3.2)	7–12			
	287 (12.2)	237 (8.3)		258 (4.0)	2–3.5			
Guanine 7-N-oxide	283 (~9.7)	227 (~31)		243 (~5.7)[b]	12	0.52	0.54[c]	0.24[c]
	292 (~6.6)	254 (~5.2)	226 (~30)	268 (~6.4)[b]	9			
	267 (9.45)	245 (7.8)		251 (7.6)	1			
Xanthine						0.50	0.42	0.30
Guanine						0.44	0.34	0.0

[a] See the Experimental Section for solvents. [b] Sensitive to small changes of pH. [c] Fluorescent.

Methylation of either the guanine or xanthine oxide with dimethyl sulfate and sodium hydroxide yields several products, some transient, as evidenced by paper chromatography of the reaction mixtures. The major final product from the xanthine oxide was more readily obtained with dimethyl sulfate in dimethylformamide. Elemental analysis shows it to be a dimethyl derivative, tentatively assigned structure IV. No further methylation resulted when this was again treated with dimethyl sulfate in dimethylformamide. Another methylated derivative, presumably an intermediate enroute to the foregoing, was obtained in small quantity. Its analysis corresponded to the replacement of one hydrogen by a methyl and of a second by a methylsulfate. Upon melting, this was converted to IV.

Attempts to prepare methylated xanthine N-oxides by direct oxidation of some methylated xanthines have failed. Both caffeine and theophylline were slowly oxidized by trifluoroperoxyacetic acid to N,N′-dimethylparabanic acid. A similar oxidative ring cleavage has been reported for caffeine with boiling nitric acid,[10] chlorine in water,[11] or chromic acid.[12] The ring system of tetramethyluric acid is also degraded by trifluoroperoxyacetic acid and allocaffeine was obtained. This has been previously reported by Fischer[13] with chlorine in water as the oxidant. It appears that derivatives of xanthine which have at least two N-methyl substituents are quite susceptible to the oxidative opening of the ring.

That the guanine was oxidized at the 7-nitrogen was demonstrated by hydrolysis of either the guanine oxide or the xanthine oxide to N-hydroxyglycine. In parallel experiments guanine and xanthine yielded glycine, which is known to be derived from carbons 4 and 5 and nitrogen 7.[14]

Until the tautomeric structures are established, we prefer to use the trivial names guanine 7-N-oxide and xanthine 7-N-oxide (nominal formulae IIa and IIIa). These may well exist in part as the 7-N-hydroxy derivatives, 2-amino-7-hydroxy-6(1H)-purinone (7-hydroxyguanine) (IIb) and 7-hydroxy-2,6(1H,3H)-purinedione (7-hydroxyxanthine) (IIIb), or as other tautomers, and a final designation must await further details of the structure. The hydrolysis to N-hydroxyglycine does demonstrate the presence of the oxygen on the 7-nitrogen, but does not prove the presence of an N-hydroxy group in the neutral molecule, since it was accomplished in 6 *N* HCl, in which each of these compounds adds a fourth proton.

The dimethyl derivative from xanthine 7-N-oxide shows no strong absorption near 220 mμ at any pH and it may be argued that the oxygen on the nitrogen is methylated and thus masks any characteristic absorption of the N-oxide function. Such a structure is supported by the failure of the methylated derivative to give a color with ferric chloride, in contrast to the guanine and xanthine oxides. The spectrum does not resemble the single maximum spectra of 1,3-, 3,7-, or 1,7-dimethylxanthines[15] but is most analogous to one of 7,9-dimethyl-1,2,3,6-tetrahydropurine-2,6-dione-betaine (V).[16] Although only one proton should remain to be removed, there are two p*K* values of 5.1 and >13, also in analogy to V which has p*K* values of 3.18 and 12.05. The dimethyl derivative may thus be tentatively assigned the structure IV, with the greater basicity relative to V comparable with the greater basicity of xanthine oxide relative to xanthine.

Experimental Section

Analyses were performed by Galbraith Laboratories, Inc., Knoxville, Tenn., and Spang Microanalytical Laboratory, Ann Arbor, Mich. Melting points were determined on either a heated block or a capillary apparatus, each calibrated. Chromatographic analyses were performed, ascending, on Whatman No. 1 paper at 25° with the development solvents: (A) 5% disodium hydrogen phosphate–isoamyl alcohol[17]; (B) 3% ammonium chloride[16]; (C) *t*-butyl alcohol–methyl ethyl ketone–formic acid–water (40:30:15:15 by volume).[18]

Guanine 7-N-Oxide Hemihydrochloride.—A mixture of 144 ml. of trifluoroacetic acid and 100 ml. of 30% H_2O_2 was magnetically stirred and 50 g. of finely powdered guanine was added slowly enough to prevent caking. It dissolved slowly and the temperature rose to about 40° because of the exothermic character of the reaction. Stirring was continued overnight when, to decompose the excess H_2O_2, about 50 mg. of 10% Pd–C was added; additional Pd–C was added twice during the day, during which time solids separated. The mixture was cooled overnight and the solids were collected and dissolved in 400 ml. of hot 2 *N* HCl. The Pd–C was removed by filtration of the hot solution. When cooled, 38 g. (62%) of matted needles was obtained. For recrystallization 4 g. was dissolved in 60 ml. of *N* HCl and cooled. *Anal.* Calcd. for $C_5H_5N_5O_2 \cdot 0.5HCl$: C, 32.39; H, 2.99; Cl, 9.52; N, 37.73. Found: C, 32.41; H, 3.20; Cl, 9.19; N, 37.48.

(10) J. Stenhouse, *Ann.*, **45**, 372 (1843); **46**, 227 (1843).
(11) P. Rochleder, *ibid.*, **73**, 57 (1850).
(12) R. Maly and F. Hinteregger, *Monatsh. Chem.*, **2**, 87 (1881).
(13) E. Fischer, *Ann.*, **215**, 275 (1882).
(14) L. F. Cavalieri, J. F. Tinker, and G. B. Brown, *J. Am. Chem. Soc.*, **71**, 3973 (1949).
(15) L. F. Cavalieri, J. J. Fox, A. Stone, and N. Chang, *ibid.*, **76**, 1119 (1954).
(16) W. Pfleiderer, *Ann.*, **647**, 161 (1961).
(17) C. E. Carter, *J. Am. Chem. Soc.*, **72**, 1466 (1952).
(18) K. Fink, R. E. Cline, and R. M. Fink, *Anal. Chem.*, **35**, 389 (1963).

It gives a blue color with ferric chloride. The solution requires heating to enhance solubility and give a deep color.

Guanine 7-N-Oxide.—A 10-g. sample of the hemihydrochloride was dissolved in 200 ml. of water and 150 ml. of 1 *N* sodium hydroxide, and was reprecipitated by slow addition of 50 ml. of 20% acetic acid. It was washed at the centrifuge three times by suspension in 500 ml. of water and the sediment was dried under vacuum in the centrifuge tubes to yield 8.3 g. of analytically pure anhydrous material. *Anal.* Calcd. for $C_5H_5N_5O_2$: C, 35.93; H, 3.01; N, 41.90. Found: C, 35.71; H, 3.01; N, 41.75.

Behavior of Guanine in Trifluoroacetic Acid.—The solids which separated from a solution of 25 g. of guanine in 72 ml. of hot trifluoroacetic acid were collected and air dried for several weeks. *Anal.* Calcd. for $C_5H_5N_5O \cdot 4C_2HO_2F_3$: N, 11.3. Found: N, 12.2.

A sample which was washed with ethanol on the filter was also analyzed. *Anal.* Calcd. for $C_5H_5N_5O \cdot 2C_2HO_2F_3$: N, 18.0. Found: N, 20.3.

These samples liquefied in a capillary tube at 110–120°, but on a heated block they dried promptly and decomposed above 260°. When dissolved in HCl or in pH 6.9 buffer, the spectra were identical with that of guanine. They migrated as does guanine on paper chromatograms.

The semisolid cake from this quantity of guanine and trifluoroacetic acid was overlaid with 50 ml. of 30% H_2O_2. A stirring bar was placed on top of the cake which dissolved slowly, and the next day a yield of guanine 7-N-oxide similar to the foregoing was obtained.

Xanthine 7-N-Oxide Hydrochloride.—A solution of 20 g. of guanine 7-N-oxide·0.5HCl in 300 ml. of 18% HCl was heated at 97° for 17 hr., and then cooled in the refrigerator. The 17 g. (71%) of yellow prisms was collected and dried under vacuum over KOH. *Anal.* Calcd. for $C_5H_4N_4O_3 \cdot HCl \cdot H_2O$: N, 25.18; Cl, 15.92. Found: N, 24.80; Cl, 15.98.

Xanthine 7-N-Oxide Hemihydrochloride.—A sample recrystallized from 1 *N* HCl yielded white prisms, darkening at 220–350° without further decomposition. It was dried at 110° before analysis. *Anal.* Calcd. for $C_5H_4N_4O_3 \cdot 0.5HCl$: C, 32.22; H, 2.44; Cl, 9.52; N, 30.06. Found: C, 31.55; H, 2.93; Cl, 9.62; N, 29.97.

Xanthine 7-N-Oxide Mono- and Dihydrates.—The crude hydrochloride (17 g.) was dissolved in 400 ml. of 1 *N* NaOH, charcoal was added and the solution was filtered. About 18 ml. of acetic acid was added slowly with stirring, and the mass of fine white needles was collected and dried under vacuum to yield 13 g. (93%). *Ancl.* Calcd. for $C_5H_4N_4O_3 \cdot H_2O$: N, 30.1. Found: N, 30.0.

Recrystallization of 0.5 to 1 g. of the base or its hydrochloride from 500 ml. of boiling water yielded a mixture of prisms and needles. The fine needles could be obtained in large clusters (0.01 to 0.5 mm. × several cm.) by slow cooling on a hot plate. When solutions were cooled further in the refrigerator, or when cooled rapidly, prisms were also obtained, most of which adhered to the walls of the flask. Homogeneous samples of each type of crystals were obtained: prisms (*Anal.* Calcd. for $C_5H_4N_4O_3 \cdot 2H_2O$: C, 29.42; H, 3.95; N, 27.44. Found: C, 29.62; H, 3.33; N, 27.89); needles (*Anal.* Calcd. for $C_5H_4N_4O_3 \cdot H_2O$: C, 32.27; H, 3.25; N, 30.11. Found: C, 32.79; H, 3.11; N, 30.11).

Samples of each, dried at 118°, were again analyzed. *Anal.* Calcd. for $C_5H_4N_4O_3$: C, 35.72; H, 2.40; N, 33.33. Found: C, 35.78; H, 2.46; N, 33.44. Found: C, 35.87; H, 2.53; N, 33.40.

When dried at 118°, the prisms lost 17% and the needles 9.7% of their weight, corresponding to 2- and 1 H_2O, respectively. Each maintained its shape but became opaque. After a year at room temperature, the prisms retained their shape. They were opaque and anhydrous based upon nitrogen analysis and ϵ determination. After this time the needles still retained their water.

Xanthine 7-N-Oxide Hemisulfate.—A sample of the hydrate was recrystallized from 4 *N* H_2SO_4, and prisms were obtained. *Anal.* Calcd. for $C_5H_4N_4O_3 \cdot 0.5H_2SO_4$: N, 25.80; S, 7.38. Found: N, 26.08; S, 7.64.

A sample recrystallized from 0.5 *N* H_2SO_4 yielded typical needles of the monohydrate. *Anal.* Calcd.: N, 30.1. Found: N, 30.1.

The spectra of all the hydrates and salts and their R_f values on paper chromatography were identical. Dilute solutions gave a pale pink color with ferric chloride, but when heated to increase solubility a deep purple is obtained.

Dimethyl Derivative of Xanthine 7-N-Oxide.—A suspension of 3.7 g. of xanthine 7-N-oxide in 100 ml. of dimethylformamide was stirred magnetically, and 20 ml. of dimethyl sulfate was added. The mixture was heated to 40–44° and became water clear in 2 hr. After 17 hr. it was concentrated under vacuum, at a bath temperature of 85°, to about one-third volume. It was diluted with 200 ml. of water, stirred until homogeneous, and cooled at 4°, and 2.5 g. of white crystals was collected. A sample was recrystallized from water to yield needles, m.p. above 350°, which gave no color with ferric chloride. It is not identical with the 7-hydroxytheophylline reported[19] to melt at 210 to 213°. *Anal.* Calcd. for $C_7H_8N_4O_3$: C, 42.85; H, 4.11; N, 28.56. Found: C, 42.75; H, 4.12; N, 28.95.

In another run, when the initial reaction temperature was 50° and after concentration at a bath temperature of 70°, a small crop identical with the foregoing was collected after the diluted mixture was cooled to room temperature. When cooled at 4°, another product separated. In a capillary melting point tube, it shrinks from 185 to 200° and decomposes to 240°, varying with the temperature on insertion. *Anal.* Calcd. for $C_7H_8N_4O_6S$: C, 30.44; H, 2.92; N, 20.28; S, 11.61. Found: C, 30.32; H, 3.23; N, 19.86; S, 11.54.

The residues, from six melting points to 240°, were combined and recrystallized from 0.1 *N* NaOH by addition of dilute HCl. The product corresponded to the first dimethyl derivative of xanthine 7-N-oxide in R_f values and spectra.

Xanthine Hydrochloride.—Xanthine was recrystallized from concentrated HCl according to Strecker,[20] to yield prisms stable in air. *Anal.* Calcd. for $C_5H_4N_4O_2 \cdot HCl$: N, 18.9. Found: N, 18.7.

Recrystallization from 2 *N* HCl yielded a chloride-free sample of xanthine.

Oxidation of Caffeine.—A solution of 1.0 g. of caffeine in 6 ml. of trifluoroacetic acid and 3 ml. of 30% hydrogen peroxide was stirred at room temperature for 3 days. At this time an additional 3 ml. of 30% hydrogen peroxide was added and stirring was continued for another 5 days. A new product began to form and 4 ml. of 30% hydrogen peroxide was added. After 14 days the excess hydrogen peroxide was decomposed with 10% Pd–C. The catalyst was removed by filtration, and the filtrate was treated with ethanol–ether and cooled below 0°. The resulting white solid was collected (160 mg.), m.p. 133–140°. Recrystallization from ethanol gave white plates, m.p. 148–149°. Several investigators report melting points of 145 to 155.5°[12,21,22] for N,N′-dimethylparabanic acid. *Anal.* Calcd. for $C_5H_6N_2O_3$: C, 42.26; H, 4.26; N, 19.72. Found: C, 42.42; H, 4.37; N, 19.89.

Under similar conditions oxidation of theophylline gave the same N,N′-dimethylparabinic acid as determined by melting point, ultraviolet, and infrared spectra. Theobromine, which would be expected to give N-methylparabanic acid, was partially oxidized in 2 days to a complex mixture. In all cases the parabanic acid derivatives were discernible on paper chromatograms because the spots became pink to red in air. In no case did any spots on the chromatograms give a color with ferric chloride.

Oxidation of 1,3,7,9-Tetramethyluric Acid.—A mixture of 2.0 g. of tetramethyluric acid, 5 ml. of trifluoroacetic acid, and 2.5 ml. of 30% hydrogen peroxide was stirred at room temperature for 17.5 hr. The excess peroxide was decomposed with 10% Pd–C. The mixture was then filtered, treated with 50 ml. of methanol, and cooled below 0°. White needles were obtained, yielded 0.40 g. The filtrate was evaporated and the residual oil was triturated with methanol. An additional 0.66 g. of white material was obtained. Both fractions were combined and recrystallized from ethyl acetate to yield white prisms, m.p. 203–215°. This material was recrystallized from ethanol as white crystals, m.p. 200–202°. Fischer reports[13] m.p. 203° for allocaffeine. *Anal.* Calcd. for $C_8H_9N_3O_5$: C, 42.29; H, 3.92; N, 18.5. Found: C, 42.30; H, 4.25; N, 17.7.

It was hydrolyzed to allocaffeic acid, m.p. 158°.[13]

Hydrolysis of Guanine N-Oxide and Xanthine N-Oxide.—Samples of guanine, xanthine, guanine N-oxide (II), and xanthine N-oxide (III) (500 mg. each) were heated with 5 ml. of con-

(19) H. Goldner, G. Dietz, and E. Carstens, *Z. Chem.*, **4**, 454 (1964).
(20) A. Strecker, *Ann.*, **108**, 146 (1858).
(21) R. Behrend and L. Fricke, *ibid.*, **327**, 253 (1903).
(22) M. Menschutkin, *ibid.*, **178**, 202 (1875).

centrated HCl at 180° overnight in sealed tubes, and treated for the isolation of *p*-toluenesulfonylglycine as described.[14] From guanine and xanthine *p*-toluenesulfonylglycine, m.p. 147°, was obtained. From guanine N-oxide and xanthine N-oxide, about 50 mg. of a crystalline compound was obtained which had m.p. 135°. A mixture melting point with *p*-toluenesulfonamide, m.p. 137°, was depressed to 105°. A mixture melting point with an authentic sample of hydroxyglycine, m.p. 135°,[23] showed no depression. Comparison of the infrared spectra of the products obtained with that of hydroxyglycine proved the two to be identical.

(23) W. Traube, *Ber.*, **28**, 2300 (1895).

Acknowledgment.—We are indebted to Dr. Edward A. Kaczka, Merck & Co., Inc., for a sample of N-hydroxyglycine, and to Marvin J. Olsen for very capable assistance.

Studies toward the Synthesis of the Proposed Structure for the Cockroach Sex Attractant[1]

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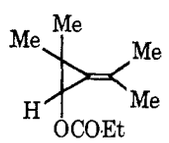
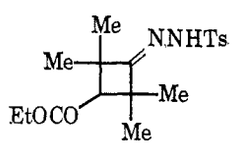
Thermal decomposition of the sodium salt of II has been examined with a view to synthesize I, the proposed structure for the cockroach sex attractant. The decomposition reaction affords a highly complex mixture. Besides the azine VI, two liquid components have been separated by preparative vapor phase chromatography. One of these compounds has been identified as 2,5-dimethylhexatriene (VII). Although the second compound appeared to represent structure I from its spectroscopic properties, it has been shown to possess structure XIII on the basis of chemical evidence. Mechanisms for the formation of VII are amply discussed.

The highly potent sex attractant of the female American cockroach, *Periplaneta americana* L., has aroused considerable interest among several organic chemists. In connection with an earlier structural proposal I for the attractant,[2,3] the thermal decomposition of the sodium salt of the tosylhydrazone II was examined in detail.[4] The method[5] seemed particularly

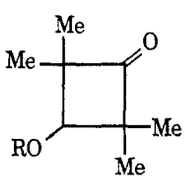


I

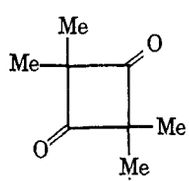


II

interesting because the desired intermediates III and IV could be readily obtained from the commercially available dimethylketene dimer V. Decomposition



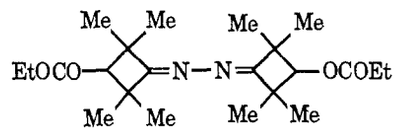
III, R = H
IV, R = CO·Et



V

of the salt under a mild vacuum gave a sweet-smelling liquid whose vapor phase chromatogram indicated the presence of at least 13 components. Decomposition of the same salt in molten acetamide at 120° under a nitrogen atmosphere yielded a liquid which was found to contain 23 components. A complete separation and identification of this mixture appeared an arduous task. The complexity of this formidable mixture was, however, substantially reduced by partial separation by column chromatography over silica gel. The resulting mixture A was shown to contain 11 components. The residue on the column (mixture B) was discarded.

Occasionally, a solid (m.p. 100–101°) could be separated by careful column chromatography of the crude mixture. The infrared spectrum of the solid in CCl_4 showed, besides other bands, a strong band at 5.75 μ and a medium intensity band at 5.93 μ. The n.m.r. spectrum in $CDCl_3$ showed a three-proton triplet (J = 7 c.p.s.) at τ 8.85, four methyl resonances at τ 8.84, 8.80, 8.69, and 8.59, a two-proton quartet (J = 7 c.p.s.) at τ 7.64, and a sharp one-proton singlet at τ 5.39. The elemental analysis corresponded to the formulation $C_{22}H_{36}N_2O_4$. The solid was soon identified as the azine VI by comparison with an authentic sample.[6] For preparative purposes, the liquid mixture



VI

A was chromatographed at 140° (Carbowax 20 M). Only two components (C and D) were well separated and were obtained in a reasonably pure form for further characterization.

Compound C.—The n.m.r. spectrum of this compound was comprised of only three resonance lines: a three-proton signal at τ 8.17 slightly coupled (J = 1 c.p.s.) to a two-proton signal at τ 5.08, and a sharp

(1) Abstracted from the Ph.D. Thesis (part two) submitted by the author to the Department of Chemistry, Harvard University, April 1965.

(2) M. Jacobson, M. Beroza, and R. T. Yamamoto, *Science*, **137**, 48 (1963).

(3) After the completion of this work, M. Jacobson, M. Beroza, and R. T. Yamamoto [*ibid.*, **147**, 748 (1965)] withdrew their earlier structure[2] for the attractant.

(4) Recently, J. Meinwald, J. W. Wheeler, A. A. Nimetz, and J. S. Liu, [*J. Org. Chem.*, **30**, 1038 (1965)] have reported several interesting attempts to synthesize the attractant. Their study included the approach described in the present article. The results obtained in this laboratory differ sufficiently from those reported by these authors to warrant a separate publication.

(5) Base-catalyzed thermal decomposition of cyclobutanone tosylhydrazone has been reported to yield methylenecyclopropane in an excellent yield: L. Friedman and H. Shechter, *J. Am. Chem. Soc.*, **83**, 1002 (1960).

(6) Readily obtained by stirring the keto ester IV with a stoichiometric amount of hydrazine in methanol.