reaction mixture was cautiously poured into 100 ml of saturated sodium bicarbonate. The product was extracted with methylene chloride and the extract was washed with saturated sodium bicarbonate, dried ($MgSO_4$), and concentrated under reduced pressure. Crystallization from dichloromethane–ether gave 0.0369 g of the trichloro compound **17**, mp 198–200°, uv max (ethanol) 240 mμ (ε 5200). The melting point was undepressed upon admixture with the material prepared in A. In addition, the ir and nmr spectra were identical with those obtained from the preparation described in A.

4,6-Dichloro-17α,21-dihydroxypregna-4,6-diene-3,11,20-trione 21-Acetate (18). A. By Direct Chlorination in Propionic Acid.—To a cooled (0°) solution of 1.50 g of 6-chloro-17α,21-dihydroxypregna-4,6-diene-3,11,20-trione 21-acetate[5] in 16 ml of dry dimethylformamide and 10 ml of ether was added 4.0 ml (20% excess) of a 1.05 *M* solution of chlorine in propionic acid. The resulting solution was allowed to stand at 0° for 10 hr and then at room temperature for 5 hr. The reaction mixture was poured into 100 ml of water and extracted with methylene chloride. The organic layer was washed twice with 5% sodium bicarbonate and once with brine. The dried (Na_2SO_4) organic layer was evaporated and the residue was crystallized from methylene chloride–ether to give 600 mg (37%) of **18**, mp 255–257.5°. An additional crystallization from the same solvent system gave the analytical sample: mp 257.5–259.5°; $[\alpha]^{25}D$ +398.4° (*c* 0.5, $CHCl_3$); uv max (ethanol) 295 mμ (ε 15,750); ir ($CHCl_3$) 1750, 1730, 1720, and 1697 cm^{-1}; nmr ($CDCl_3$) δ 6.30 (s, 1, C-7 H).

Anal. Calcd for $C_{23}H_{26}Cl_2O_6$: C, 58.86; H, 5.58; Cl, 15.11. Found: C, 58.85; H, 5.79; Cl, 15.22.

B. By Dehydrochlorination of 17.—The trichloro compound **17** (0.044 g) was dissolved in 1 ml of pyridine and was allowed to stand at 25° for 75 min. The reaction mixture was then poured into 25 ml of 1 *N* hydrochloric acid and the product was extracted with ethyl acetate. The combined organic layers were dried ($MgSO_4$) and evaporated under reduced pressure to give 0.033 g of colorless solid, uv max (ethanol) 296 mμ (ε 19,700). Crystallization from methylene chloride–ether gave 0.0207 g (51%) of **18**, mp 258.5–261°, undepressed upon admixture with the sample described in A.

Crystallography.—Crystals of 4α,6,7α-trichloro-17α,21-dihydroxypregn-5-ene-3,11,20-trione 21-acetate (**17**, $C_{23}H_{27}Cl_3O_6$, mol wt 505.84) were grown from an ethyl acetate–hexane mixture. The crystal data are $a = 12.915 \pm 0.003$, $b = 15.310 \pm 0.005$, $c = 6.073 \pm 0.002$ Å, $\beta = 105.04 \pm 0.02°$ (at 21°, Cu $K_\alpha = 1.5418$ Å), $V = 1159.7$ Å, $D_m = 1.47$ g cm^{-3}, $D_c = 1.45$ g cm^{-3}, $Z = 2$, F(000) = 528; space group $P2_1$, (C_2^2, No. 4) (0*k*0 absent for *k* odd).

The intensities of 2266 independent X-ray diffraction maxima with $2\theta \leq 140°$ were measured on a Hilger and Watts Model Y290 four-circle diffractometer by a moving crystal-moving detector technique, using Ni-filtered Cu K_α radiation. Of these data, 410 were not significantly greater than background and were excluded from the structure analysis. The data were corrected for absorption (μ 39.1 cm^{-1}) by the method described by Coppens, *et al.*[22] as well as for the usual Lorentz and polarization effects. The crystal used was a rectangular prism with dimensions 0.14 × 0.18 × 0.24 mm.

All calculations were performed on a GE-635 computer. The Fourier program used is a local revision of one originally written at the University of Wisconsin.[23] Local modifications of the Busing–Martin–Levy ORFLS[24] crystallographic least squares program were used for the refinement in which the function $\Sigma w (|F_o| - |F_c|)^2$ was minimized. In the final cycles of least squares refinement, the weights were taken as $w = 1/(6.25 + F_o + 0.02 F_o^2)$.

The scattering curves of Cromer and Waber[25] were used for Cl, O, and C and that of Stewart, Davidson, and Simpson[26] for H. The C scattering curve was corrected for both the real and imaginary parts of the anomalous dispersion.[27]

Registry No.—**2**, 26527-17-3; **3**, 26527-18-4; **6**, 26527-19-5; **7**, 19892-45-6; **17**, 26527-21-9; **18**, 26527-22-0.

Acknowledgments.—The authors are indebted to the following members of our physical chemistry department (Dr. P. Bommer, director): Mr. S. Traiman, Dr. V. Toome, Dr. T. Williams, and Dr. F. Scheidl for the ir spectra, uv spectra, nmr spectra, and microanalyses, respectively.

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(23) J. F. Blount, "A Three-Dimensional Crystallographic Summation Program for the CDC 1604 Computer," Ph.D. Thesis (Appendix), University of Wisconsin, 1965.

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Migration of the Double Bond in the Side Chain of Sterols with Iodine[1a]

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Received April 13, 1970

Several papers including our reports on fucosterol[2] and kauren[3] have already been published on the double bond migration with iodine wherein such behavior was presumed to be similar to acid migrations, but detailed investigations on this reaction have not been reported. When a solution of fucosterol acetate (**1**) or 24-methylene cholesterol acetate in benzene with iodine was refluxed, the $\Delta^{24(25)}$ isomer in a yield of *ca.* 50% and other unknown products were obtained.[2] Details of the double bond migration of sterol side chains are described in this paper.

Gas chromatographic analysis of the products of fucosterol acetate indicates that the product consists of at least three components.[4] By alumina chromatography and recrystallization, stigmasta-5,24-dien-3β-ol acetate (**3**) was obtained pure, but the other products could not be successfully separated by tlc or column chromatography. Catalytic reduction of the products with PtO_2 gave single compound which was identified as 24-ethylcholestanol acetate by mass spectrometry and glc. Thus, the by-products are assumed to be double bond isomers of fucosterol.

It was found by glc analysis of the ozonization products at periodic intervals that only the first component eluted from the glc is attacked by ozone very slowly.

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(1) (a) This paper constitutes part 1 of a series entitled "Studies on Steroids." (b) Institute of Applied Microbiology, University of Tokyo, Bunkyo-ku, Tokyo. (c) Central Laboratory, Sankyo Co. Ltd., Shinagawa-ku, Tokyo.

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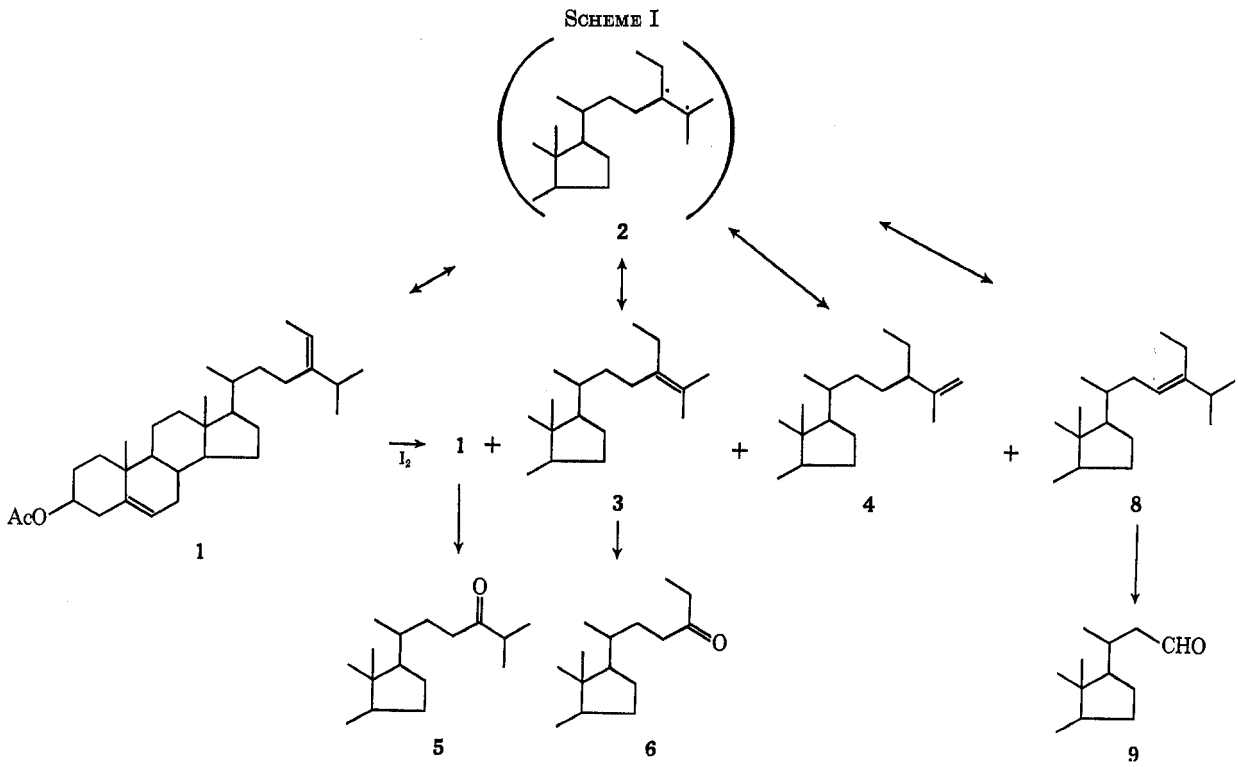
(3) K. Mori, M. Matsui, N. Ikekawa, and Y. Sumiki, *Tetrahedron Lett.*, **29**, 3395 (1966).

(4) Recently, a similar gas chromatographic result was reported by L. J. Goad, and T. W. Goodwin, *Eur. J. Biochem.*, **7**, 502 (1969).

SCHEME I



Thus after partial ozonization, followed by zinc–acetic acid reduction and alumina chromatography, four crystalline substances were obtained: nonaffected substance, mp 126–128° (**4**), two ketonic substances, mp 126–129° (**5**) and mp 165–168° (**6**), and an alcohol, mp 154–156° (**7**).

The assignment of compound **4** as stigmasta-5,25-dien-3β-ol acetate was based on the ozonization which afforded formaldehyde and on the nmr and ir spectra. Recently, stigmasta-5,25-dien-3β-ol was isolated by Sucrow[5] from *Momordia charantia* and also by Manzoor-i-Khuda[6] from *cleroden infortunatum* named as clerosterol. Direct comparison with Sucrow's sample shows no melting point depression and the same ir and nmr spectra, except for the difference of a minor point of nmr. Configuration at C_{24} of these samples might be different and our synthetic Δ^{25}-sterol was presumed to be a 24*R* and 24*S* mixture.

Compounds **5**, **6**, and **7** were identified as 3β-hydroxy-5-cholesten-24-one acetate, 3β-hydroxy-26-nor-5-cholesten-24-one acetate, and 24-nor-5-cholene-3β,23-diol 3-acetate, respectively, by direct comparison with authentic samples. 24-Nor-5-cholene-3β,23-diol may be derived from corresponding aldehyde (**9**) during zinc–acetic acid reduction of the ozonide. The C_{25} ketone derived from compound **4** was not detected in crude ozonization product by glc analysis.

By exhaustive ozonization of iodine rearrangement products and subsequent separation of the dinitrophyenylhydrazones, formaldehyde, acetaldehyde, acetone, and ethyl isopropyl ketone were obtained. Thus, it is clear from these results that double bond of fucosterol migrated in the presence of iodine to Δ^{23} (**8**), Δ^{24} (**3**), Δ^{25} (**4**), and $\Delta^{24(28)}$ (starting material). The yield of these isomers was calculated roughly from glc analysis of the crude product from partial ozonization: Δ^{23} 22%; Δ^{24}, 37%; Δ^{25}, 27%; and $\Delta^{24(28)}$, 14%.

Another compound having a same double bond system was synthesized and subjected to a same reaction conditions to further confirm this rearrangement. Citronellol acetate was converted to 3,7-dimethyloctanol-6-one acetate (**10**) by hydroboration followed by chromic acid oxidation. 3,7-Dimethyl-6-methyleneoctanol acetate (**11**), which has the same double bond system as fucosterol, was obtained by a Wittig reaction of **10**. Treatment of **11** with iodine furnished a reaction mixture which exhibited three peaks by glc as similar as fucosterol. The Δ^6 isomer (**14**), which exhibited a longer retention time on glc than other isomers, was obtained by preparative glc. By ozonization of the fraction which was removed from **14**, 3,6-dimethyloctanol-7-one acetate (**15**), 3,7-dimethyloctanol-6-one acetate (**17**), and 5-acetoxy-3-methylpentanal (**16**) were obtained from the nonvolatile fraction, and formaldehyde and methyl isopropyl ketone from the volatile fraction as their dinitrophenylhydrazones. Evidently in this case, a similar reaction as with fucosterol occurred.

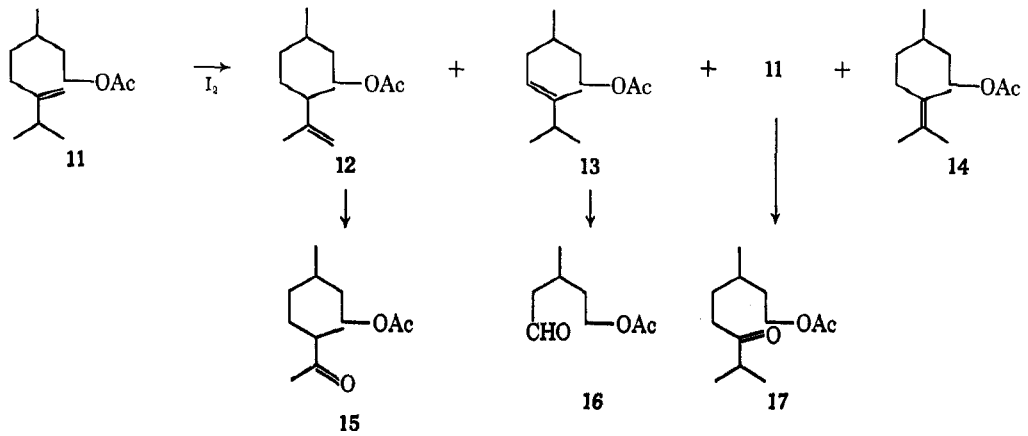
Double bond migration in the steroid skeleton with iodine was also investigated: 5α-cholest-7-en-3β-ol acetate gave a mixture of 5α-cholest-8(14)-en-3β-ol and 5α-cholest-14-en-3β-ol (2:1); $\Delta^{8(14)}$ steroid gave the Δ^{14} isomer in theoretical yield; and a Δ^{22} double bond was unaffected.

With respect to the mechanism, the following result may be important. Glc analyses of the reaction products of stigmasta-5,24-dienol acetate (**3**) and stigmasta-5,25-dienol acetate (**4**) with iodine provided that both products were the same mixture as from fucosterol acetate. Hence the reaction of double bond isomers may take place *via* the same intermediate which might have a diradical structure as **2**. The key point of the reaction may be that both C-24 and C-25 are tertiary carbons.

(5) W. Sucrow, *Tetrahedron Lett.*, **26**, 2217 (1965), *Chem. Ber.*, **99**, 2765 (1966).

(6) M. Manzoor-i-Khuda, and S. Sarela, *Tetrahedron*, **21**, 797 (1965), M. Manzoor-i-Khuda, *Tetrahedron Lett.*, **22**, 2377 (1966).

SCHEME II



Experimental Section[7]

Reaction of Fucosterol Acetate (1) with Iodine.—A solution of 3.0 g of fucosterol acetate and 200 mg of iodine in 150 ml of benzene was refluxed for 15 hr. After washing the solution with 1% $Na_2S_2O_3$ and water, the benzene layer was dried over anhydrous Na_2SO_4. Evaporation of the solvent gave 2.9 g of crude product.

Glc analysis of the product showed three peaks: peak 1, retention time 27.6 min (compound **4**); peak 2 (shoulder), retention time 28.8 min (fucosterol acetate and compound **8**); and peak 3, retention time 33.2 min (compound **3**). Column packing was 1.5% OV-17 on Gas-Chrom P (80–100 mesh), 200 cm × 4 mm i.d., column temperature 232°, carrier gas flow rate 40 ml/min.

Catalytic Hydrogenation of the Crude Product.—Crude iodine reaction product, 180 mg, was dissolved in 8 ml of glacial acetic acid and reduced with 60 mg of platinum oxide. About 2 molar equiv of hydrogen was absorbed over a period of 2 hr. The reduction product showed the same gas chromatographic behavior and ir and mass spectra as β-sitostanol acetate.

Ozonization of Crude Product.—Crude product of iodine reaction, 300 mg, in 5 ml of methylene chloride was ozonized with 6.8 mol/min flow rate of O_3 for 90 sec under Dry Ice–acetone cooling. Treatment with 50 mg of Zn and 1 ml of acetic acid gave 220 mg of oily substance, which was placed on an alumina column chromatograph.

Fraction eluted with 20% hexane–benzene gave 12 mg of stigmasta-5,25-dien-3β-ol acetate: mp 126–128° from methanol; $[\alpha]^{25}D$ −43.2° (*c* 1.35, $CHCl_3$); nmr ($CDCl_3$) δ 0.65 (s, 3, C_{18}-CH_3), 1.00 (s, 3, C_{19}-CH_3), 1.57 (s, 3, H_3C—C(R)=), 2.02 (s, 3, $-OCOCH_3$), 4.69 (m, 2, CH_2=C), 5.38 (m, H olefin); ir (KBr) 3099, 1647, 886 cm^{-1} (C=CH_2). *Anal.* Calcd for $C_{31}H_{50}O_2$: C, 81.83; H, 11.08. Found: C, 81.92; H, 10.75. By saponification with 5% KOH–MeOH by a usual manner, free sterol, mp 129–132°, $[\alpha]^{25}D$ −40.5° (*c* 0.33, $CHCl_3$), was obtained.

Physical constants of our sample and Sucrow's one differ negligibly, but slightly higher melting points were reported by Khuda. Sucrow's sample shows two peaks at 4.60–4.70 ppm instead of 4.69 ppm of our sample.

From benzene fraction two ketonic substances, mp 126–129° and mp 165–166°, were obtained by recrystallization from methanol. These compounds were identified as **5** and **6**, respectively, by the direct comparison with authentic samples.

Fraction eluted with 10% ether–benzene gave 30 mg of 24-nor-5-cholene-3β,23-diol 3-acetate (**7**), mp 154–156° from methanol, which was also identified with an authentic specimen by direct comparison.

Glc analysis of the ozonization crude product on 1.5% OV-17 (200 cm × 4 mm i.d., column temperature 280°) showed that the product contained 22% of **9** (retention time 7.5 min), 27% of **4** (retention time 11.2 min), 7% of **5** (retention time 14.3 min), and 14% of **6** (retention time 15.3 min).

Exhaustive Ozonization of the Product.—A solution of 100 mg of the crude product in methylene chloride solution was ozonized for 20 min. After treatment with Zn and acetic acid, a saturated solution of 2,4-dinitrophenylhydrazine in 2 *N* HCl was added to the solution and stirred for 10 min, then extracted with methylene chloride. The products, 143 mg, were purified by preparative tlc using benzene as a solvent. Extraction of the band of R_f 0.7 gave 5 mg of dinitrophenylhydrazones. It was confirmed by tlc that this material was a mixture of hydrazones of formaldehyde (retention time 3.9 min), acetaldehyde (retention time 5.9 min), acetone (retention time 7.7 min), and ethyl isopropyl ketone (retention time 12.5 min) using a column of 1.5% OV-1 on Shimalite W (80–100 mesh), 220°, 300 cm× 4 mm i.d. These retention times were also identical with those of standard samples using a column of 1.5% OV-17.

3,7-Dimethyloctanol-6-one Acetate (10).—A solution of $NaBH_4$, 450 mg, in 3 ml of diglyme was added to a mixture of 3 ml of $BF_3 \cdot Et_2O$ and 3 ml of diglyme at 24° for 30 min under stirring and the generator was heated at 70–80° for 1 hr. The diborane gas generated during this period was introduced to a solution of 5.1 g of citronellol acetate in 10 ml of tetrahydrofuran. Water (3 ml), 4.5 ml of 3 *N* NaOH, and 4.5 ml of 30% H_2O_2 were added under ice cooling and the mixture was stirred for 1 hr at 34° and then extracted with ether. Drying over anhydrous Na_2SO_4 and evaporation of the ether gave 4.65 g of a mixture of hydroxy compounds.

To a solution of the hydroxy compounds in 450 ml of pyridine 6 g of chromic oxide in 60 ml of pyridine was added and the mixture was allowed to stand overnight. The reaction mixture was extracted with ether and the ether layer was washed with 10% HCl, 10% $NaHCO_3$, and then with water, After drying over anhydrous Na_2SO_4 and evaporation of the ether, 3.42 g of oily product was obtained, which was placed on an alumina column. From the ether–petroleum ether (1:9) elutates 1.2 g of **10** was obtained: bp 120–124° (18 mm); ir (KBr) 1700 cm^{-1}.

3,7-Dimethyl-6-methyleneoctanol Acetate (11).—A solution of 1.0 g of **10** in 2 ml of tetrahydrofuran was added to the ylide solution prepared in the following manner: 8 ml of dimethyl sulfoxide was added to 720 mg of NaH and the mixture was heated at 75–80° for 45 min until the evolution of hydrogen ceased. A solution of 5.36 g of triphenylphosphine methylbromide in 15 ml of dimethyl sulfoxide was added under cooling on ice and this ylide solution was used after stirring for 10 min at room temperature. After stirring for 16 hr at 55°, the reaction mixture was poured on ice–water and extracted with *n*-hexane affording a desacetyl compound from the extract. Then the product was acetylated with pyridine and acetic anhydride and was purified by silica gel chromatography. From the petroleum ether fraction 560 mg of **11** was obtained: nmr ($CDCl_3$) δ 0.95 (d, 3, *J* = 3.6 Hz, CH_3-CH), 1.02 [d, 6, *J* = 4.2 Hz, $(CH_3)_2$CH-], 1.94 (s, 3, $OCOCH_3$), 3.97 (t, 2, *J* = 4.3 Hz, CH-O), 4.60 (d, 2, *J* = 4.5 Hz, C=CH_2); ir ($CHCl_3$) 1740 (C=O), 889 cm^{-1} (C=CH_2).

Reaction of 3,7-Dimethyl-6-methyleneoctanol Acetate with Iodine.—A solution of **11**, 1.0 g, in 50 ml of benzene was refluxed with 50 mg of iodine for 15 hr. The reaction mixture was treated by the same method as for **1**. Glc analysis of the product in-

(7) Melting points were determined microscopically on a hot stage and are uncorrected. The nmr spectra were determined on a Japan Electric Optical Lab. Model C-60 and mass spectra were determined on a Hitachi Model RMU-6 (single focusing). The gas chromatograph used in this study was Shimiadzu Model 4APF with hydrogen flame ionization detector.

dicated that the product consisted with three components having the retention times of 22.2, 23.0, and 32.0 min on a 5% OV-17 column, 3 m × 6 mm i.d. The product was fractionated into two portions, fraction A including the 22.2 and 23.0 min peaks and fraction B including the 32.0 min peak.

The structure of fraction B was determined as **14** by the spectra: nmr ($CDCl_3$) δ 0.94 (d, 3, J = 4.8 Hz, CH_3-CH), 1.63 (s, 9, CH_3 on double bond), 2.04 (s, 3, O-Ac), 4.10 (t, 2, J = 6.5 Hz, H_2C-O); ir (liquid film) 1740 cm^{-1} (C=O).

Fraction A in methylene chloride was ozonized for 30 min under cooling with Dry Ice–acetone. After the reaction mixture was treated with Zn and acetic acid, the mixture was filtered. Then, to the filtrate, a saturated solution of 2,4-dinitrophenylhydrazine in 2 *N* HCl was added and the mixture was stirred for 10 min at room temperature. The product was extracted with methylene chloride. Thus, 113 mg of a hydrazone mixture was obtained, which was separated by preparative tlc on silica gel G.

From the fraction, R_f 0.8, 13 mg of methyl isopropyl ketone DNP, mp 121°, was obtained. This substance agreed in melting point and ir with an authentic specimen. The DNP showing R_f 0.6, 9 mg, was identical with formaldehyde DNP in all respects.

From the fraction of R_f 0.2, 71 mg of yellow oil was obtained which was purified by alumina chromatography. The *n*-hexane–benzene (7:3) fraction gave 5 mg of oil, the structure of which was determined as **15** DNP by the nmr and mass spectra: nmr ($CDCl_3$) δ 1.05 (d, 3, J = 6.0 Hz, CH_3-CH), 1.25 (s, H_3CCH-C=N), 1.58 (s, 3, H_3CC=N), 2.05 (s, 3, O-Ac); mass spectrum *m/e* 394. By glc analysis, this substance was shown to contain a small amount of **17** DNP by the comparison with authentic sample. The benzene fraction gave 20 mg of 5-acetoxy-3-methylpentanal (**16**) DNP. The structure was confirmed by the nmr and mass spectra: nmr ($CDCl_3$) δ 1.00 (d, 2, J = 4.5 Hz, CH_3-CH), 2.05 (s, 3, O-Ac), 4.15 (t, 2, J = 6.0 Hz, H_2C-OAc); mass spectrum *m/e* 338.

The retention times of DNP derivatives of **16**, **17**, and **15** were 4.4, 7.35, and 8.1 min on a 2% OV-1 at 270°, and 6.4, 9.0, and 11.0 min on a 1.5% OV-17 at 270°.

Registry No.—**5**, 20981-59-3; **6**, 26308-99-6; **7**, 10184-81-3; **10**, 26314-72-7; **11**, 26358-50-9; **14**, 26358-51-0; **15** DNP, 26358-52-1; **16** DNP, 26314-73-8; stigmasta-5,25-dien-3β-ol, 2364-23-0; stigmasta-5,25-dien-3β-ol acetate, 2456-00-0.

Acknowledgment.—The authors are grateful to Dr. W. Sucrow, Organic Chemistry Institute of Technischen Universitat Berlin, for the samples of stigmasta-5,25-dien-3β-ol acetate and 24-nor-5-cholene-3β,23-diol 3-acetate.

Sulfur-Containing Polypeptides. XIII. Bis Cystine Peptide Derivatives[1-3]

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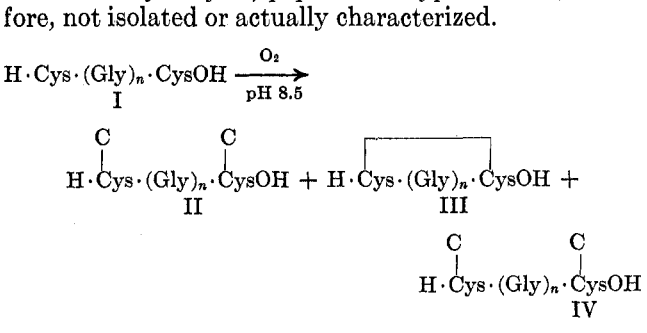
Received December 16, 1970

A method of synthesis of parallel cyclic bis cystine peptides has been devised. The cyclic dimers have been found to undergo base-catalyzed rearrangement to the corresponding cyclic monomers.

As the role of the disulfide bridges in protein molecules becomes better established,[5] the need for model peptides containing suitably placed sulfur–sulfur bonds has become more critical. The necessity for evaluating the chemical and conformational properties of cystine-containing peptides was recognized some years ago by Rydon, *et al.*[6] In a classic series of papers which to this date represent the only serious effort to evaluate the effect of the disulfide bond on the chemical and physical properties of a homologous series of peptides, Rydon, *et al.*, obtained the following data. Air oxidation of the L-cysteinyl-polyglycyl-L-cysteines (I), generated by the action of sodium in liquid ammonia on the fully blocked peptides, provided a varying series of products depending on the number of glycine residues separating the two sulhydryls. In no case was the parallel bis cystine peptide, IV, obtained; rather the antiparallel isomer, II, and the cyclic monomer, III, were the major products. Furthermore, Rydon, *et al.*, found that, as the number of glycine residues separating the two cysteine residues was increased to four or greater, the cyclic monomer was essentially the only product of the oxidation. In these investigations, the presence of the antiparallel bis cystine peptides II (n = 1, 2, 3) was established indirectly by careful dinitrophenylation experiments and subsequent controlled hydrolysis; peptides of type II were, therefore, not isolated or actually characterized.



Other investigators have noted, however, that a 20-membered disulfide "loop" (n = 4) may not be the most stable form under all conditions. For example, treatment of oxytocin[7] or lysine vasopressin[8,9] (both n = 4 systems) with weak bases resulted in loss of biological activity and formation of a dimer; oxidation of oxytocein in concentrated solution was observed to provide

* To whom correspondence should be addressed.

(1) The preceding paper of this series: R. G. Hiskey and B. F. Ward, Jr., *J. Org. Chem.*, **35**, 1118 (1970).

(2) Supported by Grants A-3416 and GM-07966 from the Institute of Arthritis and Metabolic Diseases and the Institute of General Medical Science, National Institutes of Health, U. S. Public Health Service.

(3) The following abbreviations have been employed in the text: Bz = benzoyl; Tr = trityl; Z = carbobenzoxy; WRK = 2-ethyl-5-phenylisoxazolium 3′-sulfonate; Phth = phthaloyl; BOC = *tert*-butyloxycarbonyl; Bzh = benzhydryl; DCC = *N,N*-dicyclohexylcarbodiimide; WSC = ethyl-*N*′-(3-dimethylaminopropyl)carbodiimide hydrochloride.

(4) Abstracted in part from the dissertations of G. W. Davis, M. E. Safdy, R. A. Upham, and W. C. Jones, Jr. Submitted to the University of North Carolina in partial fulfillment of the requirements for the Ph.D. degree, 1966–1969.

(5) See, for example, H. Neuman, I. Z. Steinberg, J. R. Brown, R. F. Goldberger, and M. Sela, *Eur. J. Biochem.*, **3**, 171 (1967).

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(9) A. V. Schally and J. F. Barrett, *J. Amer. Chem. Soc.*, **87**, 2497 (1965).