[Chem. Pharm. Bull. 16(1) 92—100 (1968)]

UDC 547.94.02 : 582.38

The Plane Structure of Serratinine[1,2]

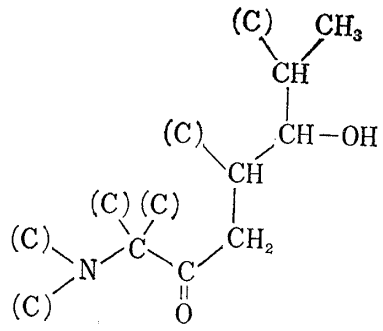
Yasuo Inubushi, Hisashi Ishii, Bompei Yasui, Masashi Hashimoto,[3a] and Takashi Harayama

Faculty of Pharmaceutical Sciences, Osaka University[3]

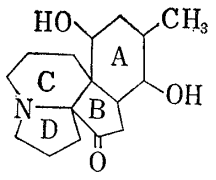
(Received March 31, 1967)

On the basis of chemical and spectroscopic evidence, the plane structure for serratinine was completly established as the formula (I).

In the preceding paper[1] of this series, the partial structure (A) for serratinine has been proposed. In the present paper, we wish to give a full account of experiments which permitted to assign the complete plane structure (I) to serratinine.



(A)

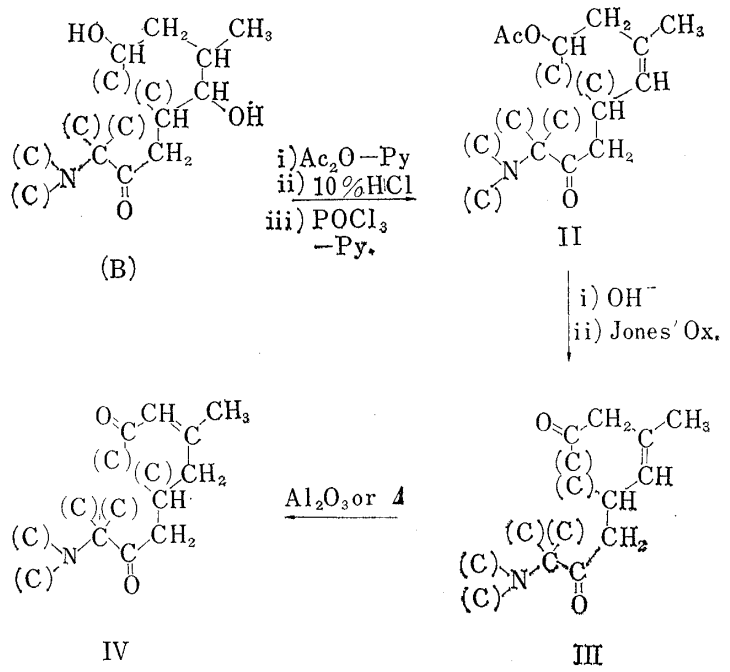


I

Chart 1

The presence of 1,4-glycol system in serratinine was demonstrated by the following reactions. Hydrolysis of anhydromonoacetylserratinine II*,[4] with alkali followed by oxidation with Jones' reagent gave dehydroanhydroserratinine II (III). This compound showed only end absorption in the ultraviolet (UV) spectrum and a 3H singlet due to a vinyl methyl group at 8.28τ in the NMR spectrum. Chromatography on basic alumina or distillation at 180° (3 mm Hg) of this ketone (III) caused a migration of the double bond to give an α, β-unsaturated ketone (IV) which showed a maximum at 238 mμ (log ε 4.04) in the UV spectrum and two carbonyl bands at 1728 and 1642 cm^{-1} in the infrared (IR) spectrum. These spectral data were consistent with the presence of the α,β-unsaturated ketonic group in its structure. Furthermore, the NMR spectrum of this compound still revealed a 3H singlet attributable to a vinyl methyl group at 8.02τ. Thus, the facile double bond isomerization is visualized in Chart 2. Consequently, we can extend the partial structure (A) for serratinine to the formula (B).

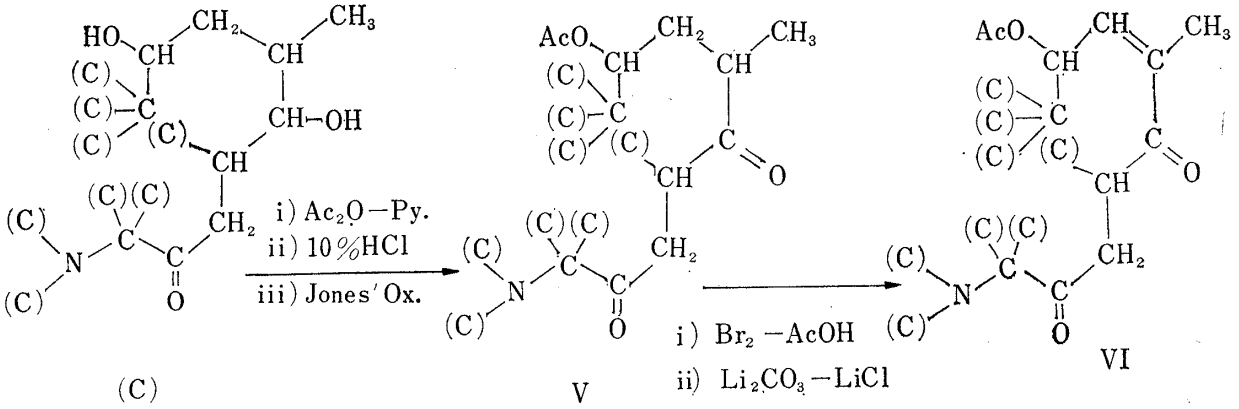
1) Studies on the Constituents of Domestic Lycopodium Genus Plants. Part V. Part IV: *Chem. Pharm. Bull.* (Tokyo), **16**, 82 (1968).
2) The preliminary report of this work appeared in *Tetrahedron Letters*, **1966**, 1537.
3) Location: *6–5, Toneyama, Toyonaka, Osaka-fu* ; *a*) Present address: *Research Laboratories, Fujisawa Pharmaceutical Co., Ltd., 1–52, Kashima-cho, Higashi-yodogawa-ku, Osaka.*
4) Physical constants and preparation of the compound marked with an asterisk in this paper appeared in the preceding paper.[1]



(B) II IV III

Chart 2

Treatment of dehydromonoacelyserratinine II* (V) with bromine in acetic acid followed by dehydrobromination gave another α,β–unsaturated ketone (VI) wihch showed a maximum at 232 mμ (log ε 3.99) in the UV spectrum and a ketonic band at 1667 cm^{-1} coupled with an absorption at 1650 cm^{-1} due to a double bond in the IR spectrum. The NMR spectrum of this α,β–unsaturated ketone (VI) showed a 3H doublet (J=1.8 cps) attributable to a vinyl methyl group at 8.12τ, a doublet (J=6.2 cps) due to a proton geminal to an acetoxyl group at 5.02τ and a pair of quartets (J_1=1.8 cps: J_2=6.2 cps) centered at 3.18τ, attributable to an olefinic proton. The double resonacne technique showed that both doublets at 8.13 and 5.02τ collapse to a singlet, respectively, by irradiation of 3.18τ. This spectral evidence indicates that the proton geminal to an acetoxyl group shows no sign of spin coupling with any proton except only one olefinic proton. In other words, the carbon atom which is situated at the another side of the carbon atom bearing an acetoxyl group would be quaternary as shown in the formula (VI). Consequently, these findings allow us to extend the partial



(C) V VI

Chart 3

structure (B) for serratinine to the formula (C).

At the present stage of this work, it is appropriate to inspect each ring size in serratinine skeleton. Dehydration of deoxoserratinine* (VII) with $POCl_3$–pyridine gave bisanhydrodeoxoserratinine (VIII) which showed a characteristic absorption of homoannular diene chromophore at 266 mμ (log ε 3.89) in the UV spectrum. Treatment of this diene with diethyl acetylenedicar-

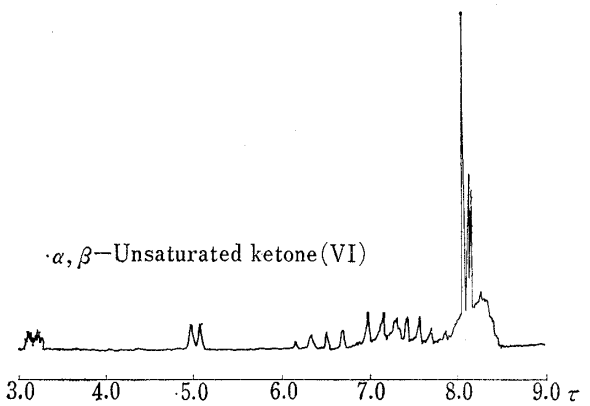


Fig. 1.

boxylate[5] followed by thermal decomposition afforded diethyl 4-methylphthalate[5] (IX) which was identified with an authentic sample by comparison of the IR spectra. On the basis of the Alder-Rickert rule,[5] we can readily infer that the ring A bearing a secondary methyl group is a six membered ring.

Our attention then turned to the B ring inluding a ketonic group. Since the IR spectra of the serratinine and its derivatives leaving the original ketonic group intact, showed an absorption between 1724 and 1748 cm^{-1}, it is assumed that the B ring is a five membered one.

By taking these two findings into consideration, we now can elaborate the partial structure (D) for serratinine.

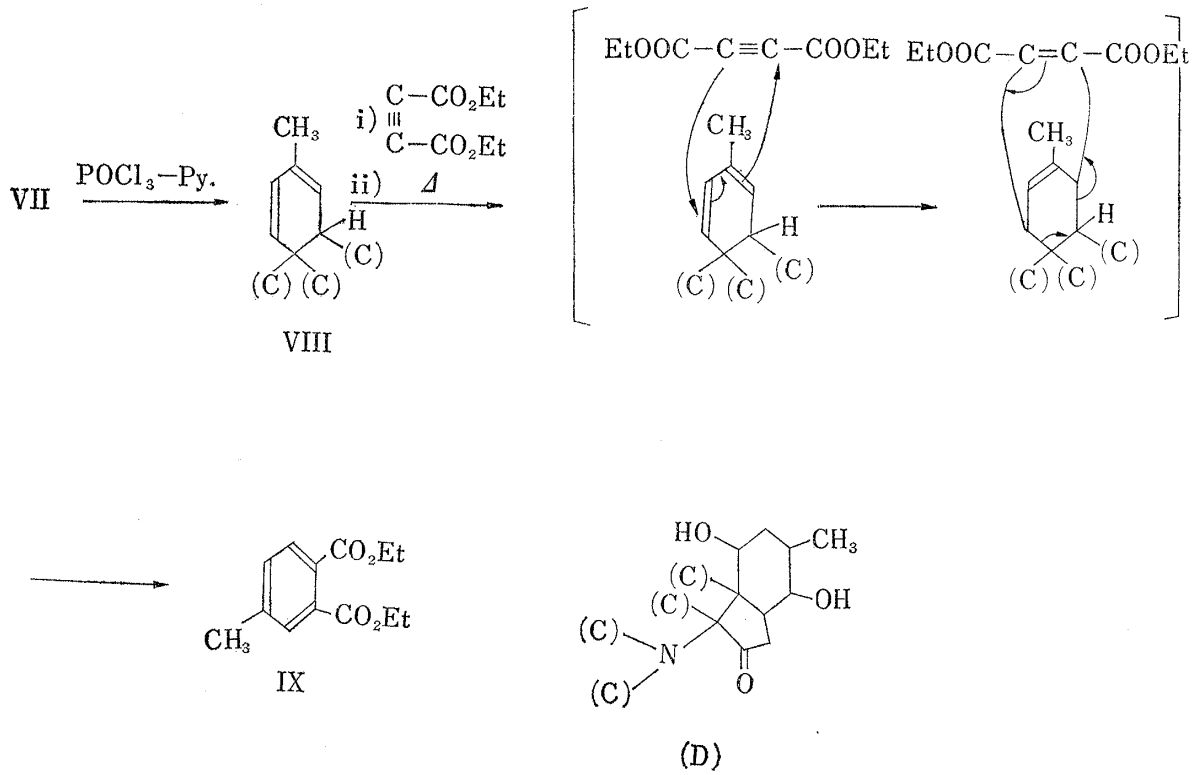


Chart 4

Chemical proof for establishing the partial structure close to the nitrogen atom was provided by successive Hofmann degradations of deoxoserratinine* (VII). Treatment of deoxoserratinine methiodide with potassium butoxide gave a crystalline methine (X) in good yield which showed a 1H multiplet due to an olefinic proton at 4.20τ in the NMR spectrum. This fact suggested that the ring cleavage had taken place between a quaternary carbon atom and nitrogen by this reaction as shown in Chart 5. Catalytic hydrogenation of the methine afforded a dihydro compound (XI) which showed no olefinic proton in the NMR spectrum. The elementary analyses of this base showed irregular figures, probably because of easy formation of carbonate, so characterization was achieved as its methiodide. The second stage Hofmann degradation of the dihydromethine (XI) afforded a mixture which was proved to consist of two components by thin-layer chromatography.[6] The formation of both components can be rationalized by assuming that the Hofmann degradation caused the ring fission in two different ways. This assumption was supported by the following obsrva-

5) K. Alder and H.F. Rickert, *Ann.*, **524**, 180 (1936).

6) Thin-layer chromatography was performed on Silica gel G and a solution of 1% $Ce(SO_4)_2$ in aq. 10% H_2SO_4 as a detection reagent was used. The reagent was sprayed on the plate and the plate was heated until the colored spot had been developed. The solvent system, $CHCl_3$–cyclohexane–diethylamine (2:7:1) was employed.

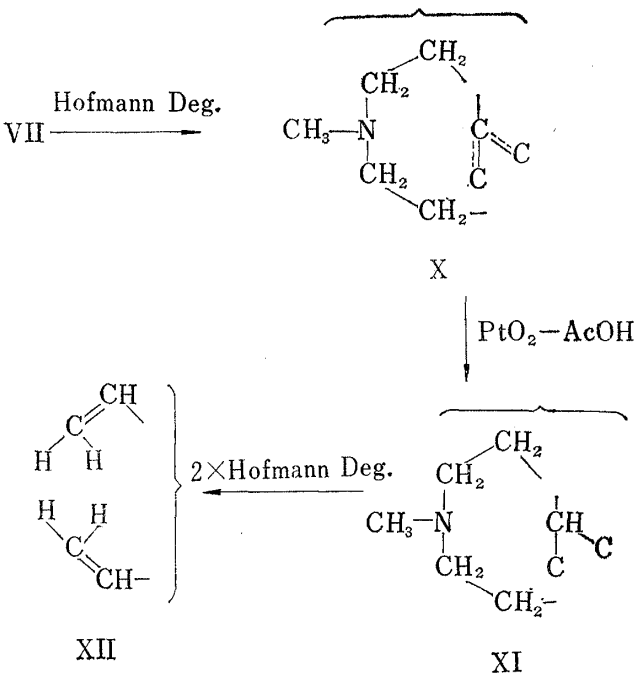


Chart 5

tion. Thus, methiodides from the mixture, without being separated, were subjected to further Hofmann degradation to give an oily neutral product, des-N-base (XII) which without any purification process was ascertained to be homogeneous by thin-layer chromatography.[6] NMR spectrum of this compound (XII) showed the complex singals corresponding to six protons between 3.7 and 5.2τ attributable to the olefinic protons. This finding led us to conclude that serratinine should have the $-CH_2-CH_2-N-CH_2-CH_2-$ system in the molecule.

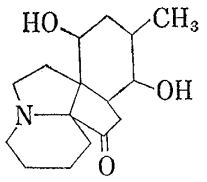
Oxidation of diacetylserratinine* (XIII) with potassium permanganate in aq. 80% acetone gave a neutral substance (XIV) whose IR spectrum showed a band at 1640 cm^{-1} due to a lactam carbonyl group on a six membered or larger ring. Since reduction of this lactam with lithium aluminum hydride gave α-dihydroserratinine (XV) which was also obtained by reduction of serratinine with the same reagent, the possibility of the skeletal rearrangement during the oxidation process would be excluded. This result suggests that one of two rings containing the nitrogen atom would be a six membered or larger ring.

Consideration of all experimental results mentioned so far, leaves only three possible formulae, (I), (E) and (F) for serratinine.

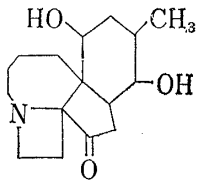
The conclusive evidence that the structure (I) is preferred to the rest, was obtained by the following experiments.

Treatment of diacetyldeoxoserratinine (XVI) with cyanogen bromide in chloroform gave a mixture, consisting of two components, which resisted to be separated into each component. This result shows that the ring fission caused by von Braun degradation presumably takes place in two ways. Treatment of this mixture with dimethylamine followed by careful chromatography provided two crystalline substances. One of them was a neutral substance (XVII), $C_{21}H_{30}O_4N_2$ which revealed a band due to an N-cyano group at 2240 cm^{-1} in the IR spectrum and a multiplet signal attributable to an olefinic proton between 4.76 and 5.28τ in the NMR spectrum. From this observation, the formation of the compound (XVII) may be rationalized by considering the ring cleavage between a quaternary carbon atom and nitrogen atom, followed by dehydrobromination. The other was a basic compound (XVIII), $C_{23}H_{37}O_4N_3$, which showed an N-cyano band at 2230 cm^{-1} in the IR spectrum. Analytical result revealed that this compound resulted from the substitution of a bromine atom with dimethylamine. Hofmann degradation of the methiodide of the compound (XVIII) furnished an olefinic compound (XIX) whose NMR spectrum showed signals attributable to olefinic protons corresponding to three protons.

Osmolation of its acetate (XX), followed by periodic acid oxidation furnished an aldehyde (XXI). In the NMR specrum, this aldehyde showed the spectrum to be expected for the ABX spin system. Thus, the aldehydic proton designated as an X component, appeared as a quartet with the spin coupling constants of J_{AX}=3.1 cps and J_{BX}=2.1 cps, respectively, at 0.21τ. The two protons of the α methylene group to an aldehyde group were designated as H_A and H_B, and these components were observed as two quartets with the coupling constants J_{AB}=15 cps, J_{AX}=3.1 cps, J_{BX}=2.1 cps, respectively, at 7.05 and 7.92τ. The double resonance



(E)



(F)

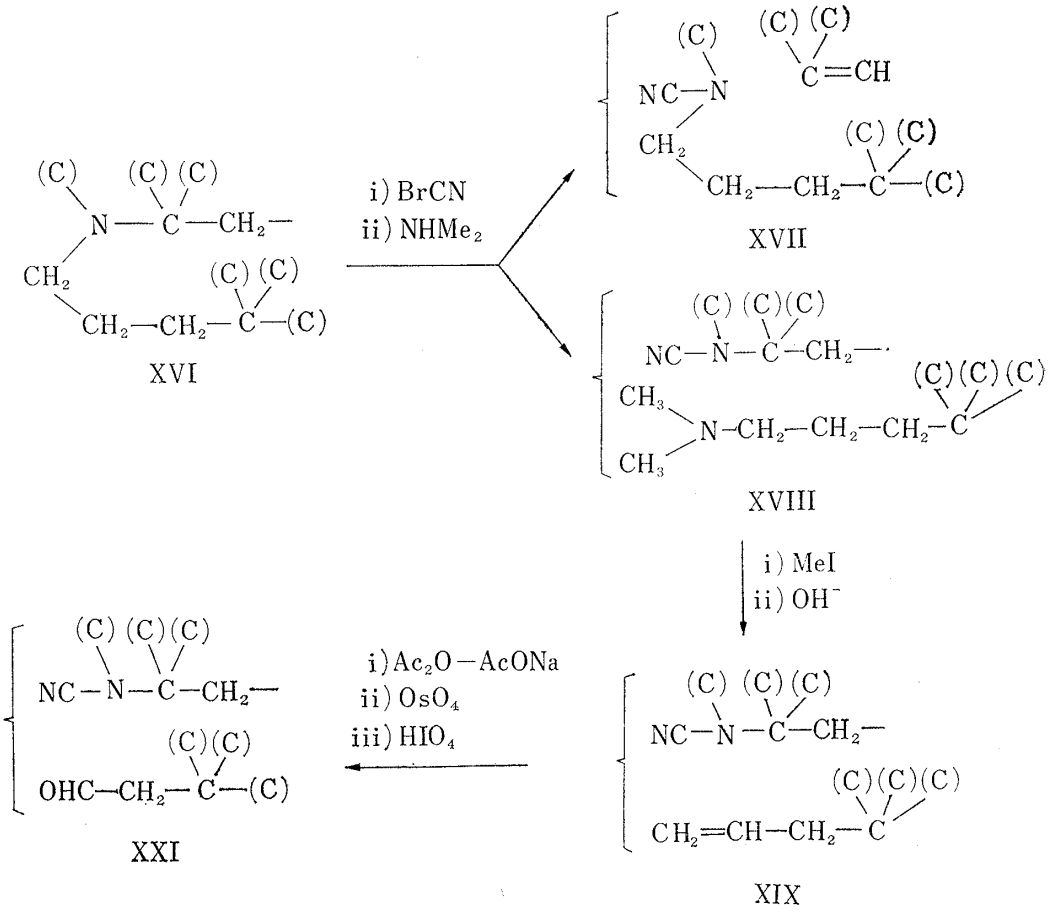


Chart 6

technique provided the strong support for the assignment toward the ABX system mentioned above. This spectroscopic evidence indicates that the aldehyde (XXI) should possess the  -CH_2-CHO grouping in the molecule. It is still ambiguous that von Braun degradation caused the fission of either (C) or (D) ring in the serratinine molecule. However, that the aldehyde (XXI) was derived from serratinine by the sequence of reactions mentioned above, indicates that serratinine should possess the -N-CH_2-CH_2-CH_2-  group-

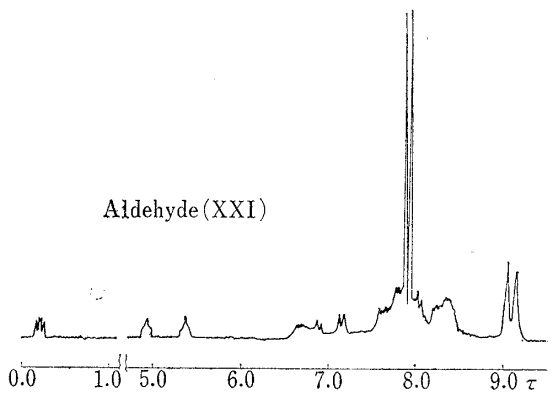


Fig. 2.

ing in the molecule.[7] This structure requirement can only be fulfilled by supposing the structure (I), not others, for serratinine.

Consequently, the structure of serratinine can be depicted by the formula (I).

Experimental[8]

Dehydroanhydroserratinine II (III)——A solution of 70 mg of anhydromonoacetylserratinine II* (II) in 5% NaOH–MeOH was heated for 1 hr under reflux. The cooled solution was made acidic with AcOH and evaporated to dryness *in vacuo*. The residue was taken up in water, made alkaline with NH_4OH and extracted with $CHCl_3$. The extract was dried over anhydr. K_2CO_3 and the solvent was evaporated. Recrystallization from benzene gave 55 mg of colorless needles, mp 196—198° (Anhydroserratinine II). *Anal.* Calcd. for $C_{16}H_{23}O_2N$: C, 73.53; H, 8.87. Found: C, 73.37; H 8.88. IR cm^{-1}: ν_{O-H} 3175; $\nu_{C=O}$ 1730. NMR τ: 4.43 (1H, m., vinyl proton); 6.32 (1H, m., >CHOH); 8.28 (3H, s., vinyl methyl). To a stirred solution of 300 mg of anhydroserratinine II in 20 ml of acetone was added 0.75 ml of Jones' reagent under ice cooling. The mixture was stirred at room temperature for 25 minutes. After decomposition of the excess reagent with MeOH, the reaction mixture was diluted with water, made alkaline with NH_4OH, and extracted with $CHCl_3$. The extract was dried over anhydr. K_2CO_3 and evaporated to dryness *in vacuo* to afford 210 mg of the residue. The residue in *n*–hexane was chromatographed on 300 mg of active charcoal. Elution with *n*–hexane gave 87 mg of crystals, which were recrystallized from *n*–hexane to give colorless prisms (III), mp 105—106°. *Anal.* Calcd. for $C_{16}H_{21}O_2N$: C, 74.10; H, 8.16. Found: C, 74.34; H, 7.95. IR cm^{-1}: $\nu_{C=O}$ 1740, 1700. NMR τ: 4.54 (1H, m., olefinic proton); 8.28 (3H, s., vinyl methyl). UV spectrum of this compound (III) exhibited no characteristic absorption over 220 mμ.

Further elution with benzene followed by ether gave an α,β–unsaturated ketone (IV), (*vide infra*).

α,β-Unsaturated Ketone (IV)——i) By chromatography: A solution of 35 mg of the pure ketone (III) in *n*–hexane was chromatographed on alumina (Merck, basic, Grade I). Elution with *n*–hexane gave 25 mg of solid mass which was recrystallized from *n*–hexane to afford colorless needles (IV), mp 117—118.5°.

ii) By distillation: Distillation of 10 mg of the pure ketone (III) at 180° (3 mm Hg) was carefully repeated until thin-layer chromatography of the distillate had shown no spot due to the ketone (III). Recrystallization of the distillate from *n*–hexane gave 5 mg of colorless needles (IV), mp 117—118.5°, which were identified with the sample obtained by the i) method by comparison of IR spectra and mixed melting point determination. *Anal.* Calcd. for $C_{16}H_{21}O_2N$: C, 74.10; H, 8.16. Found: C, 74.40; H, 8.34. IR cm^{-1}: $\nu_{C=O}$ 1728, 1642. UV λ_{max} mμ (log ε): 238 (4.04). NMR τ: 4.04 (1H, m., olefinic proton); 8.02 (3H, s., vinyl methyl).

α,β-Unsaturated Ketone (VI)——To a stirred solution of 300 mg of dehydromonoacetylserratinine II* (V) in 75 ml of AcOH was added dropwise a solution of bormine in AcOH (1.1 w/v%) until coloration owing to bromine had persisted for 1 hr and evaporated to dryness *in vacuo*. The residue was dissolved in 20 ml of dimethylformamide and 400 mg of LiCl and 300 mg of Li_2CO_3 were then added. The mixture was heated at 110° for 3 hr, cooled, and evaporated to dryness *in vacuo*. The ethereal solution of the residue was extracted with aq. 5% HCl solution. The aqueous solution was made alkaline with NH_4OH and extracted with $CHCl_3$. The extract was dried over anhydr. K_2CO_3 and evaporated. The residue in benzene was chromatographed on alumina. Elution with benzene followed by ether gave 120 mg of crystalline mass, which was recrystallized from *n*–hexane to give colorless pillars (VI), mp 112—114°. *Anal.* Calcd. for $C_{18}H_{23}O_4N$: C, 68.12; H, 7.31. Found: C, 67.95; H, 7.05. IR cm^{-1}: $\nu_{C=O}$ 1739, 1667; $\nu_{C=C}$ 1650; ν_{C-O} 1230, 1221: UV λ_{max} mμ (log ε); 232 (3.99). NMR τ: 3.18 (1H, a pair of q., J_1=1.8 cps, J_2=6.2 cps, olefinic proton); 5.02 (1H, d., J=6.2 cps, >CH–OAc); 8.08 (3H, s., –OCOCH$_3$); 8.12 (3H, d., J=1.8 cps, vinyl methyl).

Bisanhydrodeoxoserratinine (VIII)——To a solution of 150 mg of deoxoserratinine* (VII) in 2 ml of pyridine was added 25 drops of $POCl_3$ under ice cooling. The mixture was allowed to stand overnight and evaporated to dryness *in vacuo*. The residue was taken up in water, made alkaline with NH_4OH, and

7) In a preliminary communication,[2] the same deduction was deduced from the mass spectral evidence which mainly depended on the appearance of *m/e* 336 (M^+–27) peak in the spectrum of diacetylserratinine which had been measured at Prof. Ayer's laboratory. However, the sufficient data accumulated by measuring mass spectra of a number of serratinine derivatives leaving the original ketone function intact, on our machine (Hitachi mass spectrometer Model RMU 60 equipped with a direct inlet system, S.H. 300°, target current 70 μA, chamber voltage 80 V), indicated that the earlier spectrum is not reproducible. In our spectra, the appearance of M^+–28 instead of M^+–27 was observed as the common characteristic peak to the serratinine type alkaloids. This descrepancy might be ascribed to the difference of the inlet system or to that of the electron impact condition. The detail of mass spectroscopic studies of this type alkaloids will be discussed in other paper.

8) Refer to the footnote 19) in the preceding paper (Part IV).

extracted with ether. The extract was dried over anhydr. K_2CO_3 and evaporated to dryness *in vacuo*. Distillation of the residue at 90° (2 mm Hg) gave 116 mg of colorless oil (VIII) which showed only one spot on thin-layer chromatography. UV λ_{max} mμ (log ε): 266 (3.89).

Picrate——Prepared as usual in ether and recrystallized from anhydr. ether; the picrate formed yellow prisms, mp 145—147°. *Anal.* Calcd. for $C_{16}H_{23}N \cdot C_6H_3O_7N_3$: C, 57.63; H, 5.72. Found: C, 57.47; H, 5.98.

Reaction of Bisanhydrodeoxoserratinine (VIII) with Diethyl Acetylenedicarboxylate (IX)——A mixture of 66 mg of bisanhydrodeoxoserratinine (VIII) in 55 mg of diethyl acetylenedicarboxylate was heated successively at 100° for 2 hr, at 170° for further 2 hr and at 200° for 10 minutes. The cooled solution was diluted with ether and washed with aq. 2% HCl solution. The ethereal solution was dried over anhydr. $MgSO_4$ and evaporated. After distillation of the oily residue at 120° (2 mm Hg), the distillate in *n*-hexane was chromatographed on alumina. Elution with *n*-hexane gave 25 mg of colorless oil (IX) which was purified again by distillation at 120° (2 mm Hg). IR cm^{-1}: $\nu_{C=O}$ 1730; $\nu_{aromatic}$ 1610, 1572; ν_{C-O} 1280 (film). NMR τ (CCl_4): 2.34 (1H, broad d., $J=13$ cps, aromatic proton); 2.56 (1H, broad s., aromatic proton); 2.74 (1H, broad d., $J=13$ cps, aromatic proton); 5.67 (4H, q., $J=7$ cps, $2\times OC\underline{H}_2CH_3$); 7.54 (3H, s., aryl methyl); 8.61 (6H,, t., $J=7$ cps, $2\times OCH_2C\underline{H}_3$). IR spectrum of this substance was identical with that of authentic diethyl 4-methylphthalate which had been synthesized by an unequivocally established method given in the literature.[5] Hydrolysis of the substance (IX) with 5% NaOH in MeOH and sublimation of the product afforded 4-methyl phthalic anhydride, mp 90—92°, which was also identified with an authentic sample by comparison of IR spectra and mixed melting point determination.

Deoxoserratinine Methine (X)——To a solution of 30 g of *tert*-butanol dissolving 240 mg of potassium was added 450 mg of deoxoserratinine* (VII) methiodide and 2 ml of benzene. The mixture was heated for 7 hr under reflux, cooled, and evaporated to dryness *in vacuo*. To the residue was added water and extracted with $CHCl_3$. The extract was dried over anhydr. K_2CO_3 and evaporated. The residue in benzene was chromatographed on alumina. Elution with benzene left 280 mg of crystaline mass which was recrystallized from *n*-hexane to give colorless pillars (X), mp 105—106°. *Anal.* Calcd. for $C_{17}H_{29}O_2N$: C, 73.07; H, 10.46. Found: C, 72.94; H, 10.70. IR cm^{-1}: ν_{O-H} 3472, 3448; $\nu_{C=C}$ 1626. NMR τ: 4.20 (1H, m., olefinic proton); 6.22 (1H, m., $>C\underline{H}$–OH); 6.52 (1H, m., $>C\underline{H}$–OH); 7.74 (3H, s., $>NC\underline{H}_3$); 9.01 (3H, d., $J=6$ cps, $>CH$–CH_3).

Dihydromethine (XI)——A solution of 200 mg of deoxoserratinine methine (X) in 40 ml of AcOH was hydrogenated over 105 mg of PtO_2 at atmospheric pressure and room temperature until absorption of hydrogen had ceased. The filtrated solution was evaporated to dryness *in vacuo*. The residue was dissolved in water, made alkaline with NH_4OH, and extracted with $CHCl_3$. The extract was dried over anhydr. K_2CO_3 and evaporated. The residue in benzene was chromatographed on alumina and elution with benzene followed by ether left 170 mg of crystals which were recrystallized from *n*-pentane to give colorless pillars (XI), mp 86—88°. Facile carbonate formation on exposure to air was observed. IR cm^{-1}: ν_{O-H} 3559, 3436. NMR τ: 6.05 (1H, m., $>C\underline{H}OH$); 6.27 (1H, m., $>C\underline{H}OH$); 7.72 (3H, s., $>N$–$C\underline{H}_3$); 9.03 (3H, d., $J=7$ cps, $>CH$–$C\underline{H}_3$).

Methiodide——Treatment of 60 mg of the free base (XI) with methyl iodide in acetone afford 70 mg of the methiodide, which was recrystallized from MeOH–acetone to give crystals, mp 241—244°. *Anal.* Calcd. for $C_{17}H_{31}O_2N \cdot MeI$; C, 51.06; H, 8.09. Found: C, 51.27; H, 7.91. IR cm^{-1}: ν_{O-H} 3311.

Des-N-base (XII)——A solution of 280 mg of dihydromethine (XI) methiodide in MeOH was treated with freshly precipitated silver hydroxide, filtered, and evaporated to dryness *in vacuo*. The residue was pyrolyzed at 140° *in vacuo* (2 mm Hg). The distilled oil from a reaction vessel showed two spots on thin-layer chromatography and all attempts to separate this mixture were unsuccessful. Hofmann degradation of methiodides derived from this mixture, without being separated, was performed by the same condition mentioned above, and a crude oily product was shown to be virtually homogeneous by thin-layer chromatography. The product was dissolved in ether, washed successively with aq. 1% HCl solution saturated with NaCl and aq. 1% $NaHCO_3$ solution saturated with NaCl. The ethereal layer was dried over anhydr. K_2CO_3 and evaporated. Distillation of the residue at 130° under reduced pressure (2 mm Hg) gave 110 mg of colorless oil (XII). *Anal.* Calcd. for $C_{16}H_{26}O_2$: C, 76.75; H, 10.47. Found: C, 76.85; H, 10.48. IR cm^{-1}: ν_{O-H} 3571, 3460; $\nu_{C=C}$ 1634; $\delta_{C=C}$ 997, 917 ($CHCl_3$). NMR τ: 3.7—5.2 (6H, olefinic proton); 6.22 (2H, m., $2\times>C\underline{H}$–OH); 9.01 (3H, d., $J=7$ cps, $>CH$–$C\underline{H}_3$).

Lactam (XIV)——A solution of 350 mg of $KMnO_4$ in aq. 80% acetone was added to a stirred solution of 250 mg of diacetylserratinine* (XIII) in 2.5 ml of aq. 80% acetone in which 500 mg of $MgSO_4$ was suspended. The mixutre was heated at 45° for 1.5 hr and the precipitates were dissolved by bubbling sulfur dioxide gas. The solution was then acidified with sulfuric acid and diluted with water. After removal of acetone under reduced pressure, the acidic solution was extracted with ether. The extract was dried over anhydr. $MgSO_4$ and evaporated. The residue in benzene was chromatographed on alumina. Elution with benzene gave 50 mg of crystals which were recrystallized from benzene–ether to afford colorless needles (XIV), mp 215—216°. *Anal.* Calcd. for $C_{26}H_{27}O_6N$: C, 63.64; H, 7.21; N, 3.71. Found: C, 63.36; H, 7.29; N, 3.64. IR cm^{-1}: $\nu_{C=O}$ 1740, 1640; ν_{C-O} 1225. NMR τ: 4.97 (1H, m., $>C\underline{H}OAc$); 5.14 (1H, m., $>C\underline{H}$–OAc); 7.94 (3H, s., $-OCOC\underline{H}_3$); 8.04 (3H, s., $-OCOC\underline{H}_3$); 9.05 (3H, d., $J=6$ cps, $>CH$–$C\underline{H}_3$).

Reduction of 10 mg of the lactam (XIV) with 30 mg of $LiAlH_4$ in 20 ml of dry ether gave 4 mg of α-dihydroserratinine (XV) which was obtained from serratinine by the same reduction method (*vide infra*).

α-Dihydroserratinine (XV)——A solution of 12 mg of serratinine* (I) and 50 mg of lithium aluminum hydride in 30 ml of dry ether were refluxed. After 4 hr, wet ether was added dropwise to decompose the excess reagent. The organic layer was separated, dried over anhydr. K_2CO_3 and evaporated. Recrystallization of the residue from acetone gave 12 mg of colorless plated (XV), mp 277—279°. *Anal.* Calcd. for $C_{16}H_{27}O_3N$: C, 68.29; H, 9.67. Found: C, 68.55, H, 9.77. IR cm^{-1}: ν_{O-H} 3430, 3350, 3180.

Picrate——Prepared as usual in acetone and recrystalilzed from acetone–ether; yellow prisms, mp 215—217°. *Anal.* Calcd. for $C_{16}H_{27}O_3N \cdot C_6H_3O_7N_3$: C, 51.76; H, 5.92. Found: C, 51.76; H, 6.06.

Methiodide——Prepared as usual in acetone and recrystallized from methanol–acetone; colorless plates, mp 248—250°. *Anal.* Calcd. for $C_{16}H_{27}O_3N \cdot CH_3I$: C, 48.23; H, 7.14. Found: C, 47.96; H, 7.39.

Diacetyldeoxoserratinine (XVI)——To a solution of 300 mg of deoxoserratinine* (VII) in 4 ml of pyridine was added 4 ml of acetic anhydride. The solution was heated at 100° for 4 hr and evaporated to dryness *in vacuo*. The residue was dissolved in water, made alkaline with NH_4OH, and extracted with $CHCl_3$. The extract was dried over anhydr. K_2CO_3 and evaporated. The residue in benzene was chromatographed on alumina. Elution with benzene followed by ether gave 300 mg of solid mass which was recrystallized from *n*-hexane to give colorless plates (XVI), mp 140—140.5°, $[\alpha]_D^{22}$ +18.0 (c=1.00, EtOH). *Anal.* Calcd. for $C_{20}H_{31}O_4N$: C, 68.74; H, 8.94. Found: C, 68.57; H, 8.85. IR cm^{-1}: $\nu_{C=O}$ 1724; ν_{C-O} 1244, 1230; M^+: 349;[9] pK_a' 9.8.[10]

Diacetyldeoxoserratinine Cyanobromide——To a solution of 2 g of cyanogen bromide in 5 ml of dry $CHCl_3$ was added dropwise a solution of 200 mg of diacetyldeoxoserratinine (XVI) in 2 ml of $CHCl_3$. The mixture was allowed to stand at room temperature for 45 hr. The solvent and the excess cyanogen bromide were removed *in vacuo*. The residue was taken up in $CHCl_3$, and washed with aq. 5% HCl. A quantity of starting material was recovered from the washing aq. 5% HCl solution. The $CHCl_3$ solution was dried over anhydr. K_2CO_3. Removal of the solvent *in vacuo* left 141 mg of crude product which was available for the following experiments without purification. For analysis recrystallization of the crude product from acetone–ether was repeated until its melting point had raised to 186—188°, 8 mg of pale yellow pillars was obtained. *Anal.* Calcd. for $C_{21}H_{31}O_4N_2Br$: C, 55.38; H, 6.86. Found: C, 55.57; H, 6.90. IR cm^{-1}: $\nu_{C\equiv N}$ 2174; $\nu_{C=O}$ 1739, 1727; ν_{C-O} 1235.

Treatment of Diacetyldeoxoserratinine Cyanobromide with Dimethylamine——i) Ene–N–cyano Compound (XVII): Crude diacetyldeoxoserratinine cyanobromide (250 mg) was dissolved in a solution of 20 g of dimethylamine in 15 ml of methanol. The mixture was allowed to stand at room temperature for 48 hr, and evaporated to dryness *in vacuo*. The residue was taken up in water, made alkaline with NH_4OH and extracted with $CHCl_3$. The extract was dried over anhydr. K_2CO_3 and evaporated to leave the residue which was then chromatographed on alumina (Woelm, acidic, Grade III) in $CHCl_3$ solution. Elution with $CHCl_3$ gave 74 mg of a neutral compound. Recrystallization from ether–*n*-hexane gave 65 mg of colorless needles (XVII), mp 137—140°, which were insoluble in aq. 5% HCl solution, and showed a negative Beilstein's test. *Anal.* Calcd. for $C_{21}H_{30}O_4N_2$; C, 67.35; H, 8.08. Found: C, 67.48; H, 7.96. IR cm^{-1}: $\nu_{C\equiv N}$ 2240; $\nu_{C=O}$ 1720; ν_{C-O} 1245. NMR τ: 4.76—5.28 (3H, 1×olefinic proton and 2×>C<u>H</u>–OAc); 7.98 (3H, s., –OCOC<u>H</u>$_3$); 8.03 (3H, s., –OCOC<u>H</u>$_3$); 9.08 (3H, d., J=7 cps, >CH–C<u>H</u>$_3$).

ii) N–Cyano–Dimethylamino Compound (XVIII): Successive elution of chromatographic column above with MeOH gave a compound, which was dissolved in water, made alkaline with NH_4OH, and extracted with $CHCl_3$. The extract was dried over anhydr. K_2CO_3 and evaporated. Recrystallization of the residue from ether–*n*-hexane gave 120 mg of colorless needles (XVIII), mp 139—141°. *Anal.* Calcd. for $C_{23}H_{37}O_4N_3$: C, 65.84; H, 8.89. Found: C, 65.60; H, 9.03. IR cm^{-1}: $\nu_{C\equiv N}$ 2230; $\nu_{C=O}$ 1730; ν_{C-O} 1230.

Methiodide——Treatment of 80 mg of free base (XVIII) in 2 ml of acetone with 1 ml of methyl iodide afforded crystals which were recrystallized from acetone–ether to give 80 mg. of colorless prisms, mp 246—249°. *Anal.* Calcd. for $C_{23}H_{37}O_4N_3 \cdot CH_3I \cdot H_2O$: C, 49.74; H, 7.31. Found: C, 50.19; H, 7.50. IR cm^{-1}: $\nu_{C\equiv N}$ 2220; $\nu_{C=O}$ 1735, 1725; ν_{C-O} 1240, 1228.

Hofmann Product (XIX) from (XVIII)——To a solution 70 mg of N–cyanodimethylamino compound (XVIII) methiodide was added 7.5 g of KOH pellets. The mixture was heated on a water–bath for 3 hr cooled, and extracted with $CHCl_3$. The extract was dried over anhydr. K_2CO_3 and evaporated. The residue in benzene was chromatographed on alumina. Elution with benzene gave 25 mg of crystals, which were recrystallized from ether to provide colorless pillars (XIX), mp 186—188°. *Anal.* Calcd. for $C_{17}H_{26}O_2N_2$: C, 70.31; H, 9.02. Found: C, 70.42; H, 9.03. IR cm^{-1}: ν_{O-H} 3376; $\nu_{C\equiv N}$ 2191; $\nu_{C=C}$ 1637; δ_{C-H} 988, 911. NMR τ: 4.0—5.0 (3H, –C<u>H</u>=C<u>H</u>$_2$); 6.22 (1H, m., >C<u>H</u>–OH); 6.42 (1H, m., >C<u>H</u>OH); 9.03 (3H, d., J=6 cps, >CH–C<u>H</u>$_3$).

---

9) Mass spectrum was obtained with a Hitachi mass spectrometer modle RMU 6C equipped with a heated inlet system.

10) A pK_a' value was measured in 1/10 N H_2SO_4 (1 ml)–EtOH (5 ml)–H_2O (4 ml) solvent system by titration with 1/10 N NaOH solution.

Acetylation of Compound (XIX)——A solution of 0.65 g of Hofmann product (XIX) given above in 45 ml of acetic anhydride containing with 1g of anhydr. sodium acetate was heated at 120° for 7 hr, cooled, and poured into ice water. After addition of K_2CO_3 for decomposition of excess acetic anhydride, the aqueous solution was extracted with $CHCl_3$. The extract was dried over anhydr. K_2CO_3 and evaporated. The residue in benzene was chromatographed on alumina. Elution with benzene followed by ether gave 550 mg of prisms, which were recrystallized from ether to afford colorless prisms (XX), mp 229—231°. *Anal.* Calcd. for $C_{21}H_{30}O_4N_2$: C, 67.35; H, 8.08. Found: C, 67.28; H, 8.17. IR cm^{-1}: $\nu_{C\equiv N}$ 2220; $\nu_{C=O}$ 1730; $\nu_{C=C}$ 1640; ν_{C-O} 1230; δ_{C-H} 985, 910. NMR τ: 3.9—5.4 (5H, m., one $-C\underline{H}=C\underline{H}_2$ group and $2\times>C\underline{H}OAc$); 7.89 (3H, s., $-OCOC\underline{H}_3$); 7.96 (3H, s., $-OCOC\underline{H}_3$); 9.11 (3H, d., $J=6$ cps, $>CH-C\underline{H}_3$).

Aldehyde (XXI)——A solution of 500 mg of osmium tetroxide in 16 ml of dioxane was added to a solution of 550 mg of (XX) in 20 ml of dioxane and 1 ml of pyridine. The mixture was allowed to stand at room temperature for 6 days. After decomposition of excess reagent with hydrogen sulfide, the solution was filtrated and the precipitate was washed with ethanol. The combined solution was evaporated to dryness *in vacuo*. The residue in $CHCl_3$ was chromatographed on alumina. Elution with $CHCl_3$ followed by ethyl acetate gave 280 mg of crystals which were recrystallized from ethyl acetate or acetone to give colorless prisms, diol, mp 209—212°. *Anal.* Calcd. for $C_{21}H_{32}O_6N_2$: C, 61.74; H, 7.90. Found: C, 61.84; H, 8.13. IR cm^{-1}: ν_{O-H} 3410; $\nu_{C\equiv N}$ 2240; $\nu_{C=O}$ 1735; ν_{C-O} 1235, 1216.

A solution of 200 mg of periodic acid in 2 ml of water was added to a solution of 250 mg of the diol in 10 ml of dioxane. The mixture was allowed to stand at room temperature overnight, diluted with water and extracted with $CHCl_3$. The extract was washed with aq. 1% K_2CO_3, followed by water, and dried over anhydr. $MgSO_4$. Removal of the solvent left 190 mg of solid mass, which was recrystallized from MeOH–ether to give colorless prisms (XXI), mp 177—180°. *Anal.* Calcd. for $C_{20}H_{28}O_5N_2\cdot\frac{1}{2}H_2O$: C, 62.33; H, 7.58. Found: C, 62.16; H, 7.99. IR cm^{-1}: $\nu_{C\equiv N}$ 2230; $\nu_{C=O}$ 1730; ν_{C-O} 1235. NMR τ: 0.21 (1H, q., $J=3.1$ cps, $J=2.1$ cps, $-C\underline{H}O$); 4.91 (1H, m., $>C\underline{H}-OAc$); 5.36 (1H, m., $>C\underline{H}OAc$); 7.05 (1H, q., $J_1=3.1$ cps, $J_2=15$ cps, $-CH_AH_B-CHO$); 7.88 (3H, s., $-OCOC\underline{H}_3$); 7.92 (1H, q., $J_1=2.1$ cps, $J_2=15$ cps, $-CH_AH_B-CHO$); 7.93 (3H, s., $-OCOC\underline{H}_3$); 9.09 (3H, d., $J=6$ cps, $>CH-C\underline{H}_3$).

Acknowledgement We are grateful to Miss Y. Mano, Kyoto University, for elementary analyses and to Dr. T. Shingu, Kyoto University, for NMR spectral measurements.