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Structure of Gardneramine and 18-Demethylgardneramine*,[1]

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Gardneramine, the main alkaloid of *Gardneria* spp. (Loganiaceae), was found to have the structure I. At the same time a minor base, alkaloid G, was elucidated to be 18-demethylgardneramine (XVIII). Both alkaloids have an iminoether ring as the masked oxindole, some novel reactivities of which are described. Three trimethoxy oxindoles (XXXII a, b, c) were synthesized as the model compounds for locating the aromatic substituents of I.

Though the source plants of indole alkaloids mostly distribute in the tropical region of the world, some are also found in east asian countries. We have been studying indole alkaloid constituents of Japanese Loganiaceae, Apocynaceae, and Rubiaceae plants, and this paper deals with the structures of novel type indole alkaloids isolated from *Gardneria* species (Loganiaceae). We formerly published a comparative study on the alkaloidal constituents of four kinds of *Gardneria* plants.[3a] According to the result they can be divided into two groups. Alkaloidal constituents are very similar in *Gardneria nutans* Sieb. et Zucc. and *G. insularis* Nakai, but they are quite different from those of *G. multiflora* Makino[3b] and *G. Shimadai* Hayata, though all four species contain a common base, gardneramine (I), the structure of which is the subject of the present paper.[1] All the accompanying bases of the former two species possess the sarpagine type basic skeleton with a methoxyl group at $C_{(11)}$ on the aromatic ring as shown in Chart 1.[4,5] The latter group of the plants do not show the presence of any detectable amount of the above bases but contain a bisindole alkaloid and several accompanying minor bases, all of which have the novel skeleton of gardneramine type.[3a,6]

Gardneramine (I) crystallized from ether to colorless prisms, mp 134—135°, $[\alpha]_D$ −287°.[7] Elemental analysis and the molecular ion peak at *m/e* 412 on its mass spectrum revealed the molecular formula of $C_{23}H_{28}O_5N_2$. Absence of OH, NH or any sort of carbonyl group was evident from the infrared (IR) spectrum and in fact the starting material was recovered unchanged from attempted acetylation or sodium borohydride reduction of I. A strong IR absorption band was observed at 1590 cm^{-1} and thus the presence of an indolenine type –C=N– bond was suggested. In its nuclear magnetic resonance (NMR) spectrum shown in Fig. 1, signals due to three aromatic methoxyl groups (δ 3.85, 3.90, and 3.95) and one aliphatic methoxyl group (δ 3.36) were observed. Of the five oxygens of I, four thus formed methoxyl groups

* Dedicated to the memory of Prof. Eiji Ochiai.

1) For the preliminary communication see: S. Sakai, N. Aimi, A. Kubo, M. Kitagawa, M. Shiratori, and J. Haginiwa, *Tetrahedron Letters*, **1971**, 2057.

2) Location: *1-33, Yayoi-cho, Chiba 280, Japan.*

3) *a*) J. Haginiwa, S. Sakai, A. Kubo, K. Takahashi, and M. Taguchi, *Yakugaku Zasshi*, **90**, 219 (1970); *b*) In the literature of ref. 3a), the name of this plant was erroneously printed as *G. multifolia* Makino.

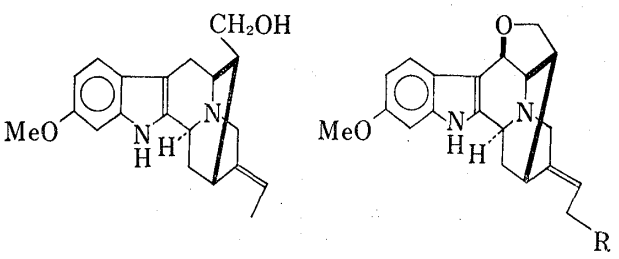
4) S. Sakai, A. Kubo, T. Hamamoto, M. Wakabayashi, K. Takahashi, H. Ohtani, and J. Haginiwa, *Chem. Pharm. Bull.* (Tokyo), **21**, 1783 (1973).

5) S. Sakai, N. Aimi, K. Katano, H. Ohhira, and J. Haginiwa, *Yakugaku Zasshi*, **94**, 225 (1974).

6) *a*) S. Sakai, N. Aimi, K. Yamaguchi, H. Ohhira, K. Hori, and J. Haginiwa, *Tetrahedron Letters*, **1975**, 715; *b*) S. Sakai, N. Aimi, K. Yamaguchi, E. Yamanaka, and J. Haginiwa, *ibid.*, **1975**, 719.

7) J. Haginiwa, S. Sakai, A. Kubo, and T. Hamamoto, *Yakugaku Zasshi*, **87**, 1484 (1967).

and the remaining one was ascribed to an etherial linkage. Taking account of the presence of the above three aromatic methoxyl groups and one aromatic proton (δ 6.40), this alkaloid was revealed to have an indole nucleus bearing three methoxyl groups on the benzene ring portion.



II: Gardnerine

III: R=H Gardnutine
IV: R=OH Hydroxy-gardnutine

Chart 1

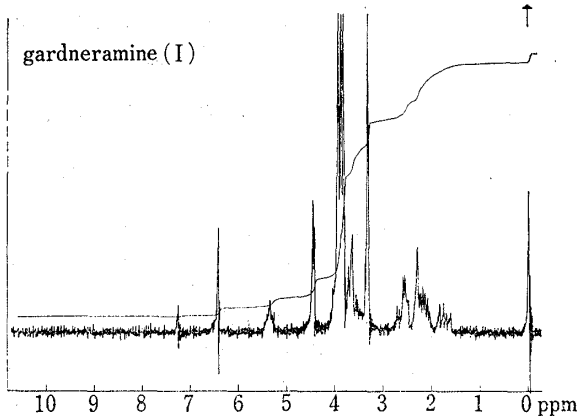
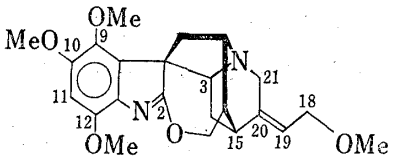


Fig. 1

As will be evident from the following text, the exact locations of the aromatic substituents remained unsettled till the last stage of the structural work. In the finally established structure (I) shown below, the positions of the aromatic methoxyl groups, the geometry of $-C_{(19)}=C_{(20)}-$ double bond and the absolute configuration were determined with the aid of X-ray crystallography, the result of which was published elsewhere.[8] In this paper the chemical part of our structural work will be described and therefore these features remained undetermined. But for the convenience of better understanding, structures of the derivatives are shown granting these features are already clarified.



Structure I

As mentioned above, one etherial linkage is present in the molecule of I. A clue to the elucidation of the structure around it came from the reaction of I in aqueous formic acid. Thus when I was heated in 85% formic acid a powder (V) was obtained. The molecular ion peak at *m/e* 430 in its mass spectrum indicated that the net change was addition of one mole of water to the mother base (I). The IR spectrum of the product, however, showed a considerable change in the molecular structure. Thus the presence of a newly formed carbonyl group was shown by a strong absorption band at 1705 cm^{-1}, and formation of NH and/or OH groups was indicated by the absorption bands at around 3425 cm^{-1}. The carbonyl group of V was not affected by attempted reduction with sodium borohydride suggesting an oxindole moiety. Acetylation of V afforded an acetate (VI), which was identical in every respects with a sample obtained from I on heating in glacial acetic acid. A tosylate (VII), mp 158—165°, was derived from V by the usual tosylation. When VII was treated with ethanolic potassium hydroxide at room temperature, the starting base (I) was regenerated in a good yield. From the above and other chemical and spectral evidences, it was concluded that I had a masked oxindole function as an oxindole iminoether, which was easily cleaved in an acidic medium to generate an oxindole moiety. The reverse reaction proceeded easily when the hydroxyl group of V was converted to a good leaving group as in the tosylate (VII).

It has been known that an iminoether of oxindole can be prepared in some special conditions such as in the presence of Meerwein's reagent under anhydrous condition. It is quite

8) N. Aimi, S. Sakai, Y. Iitaka, and A. Itai, *Tetrahedron Letters*, **1971**, 2061.

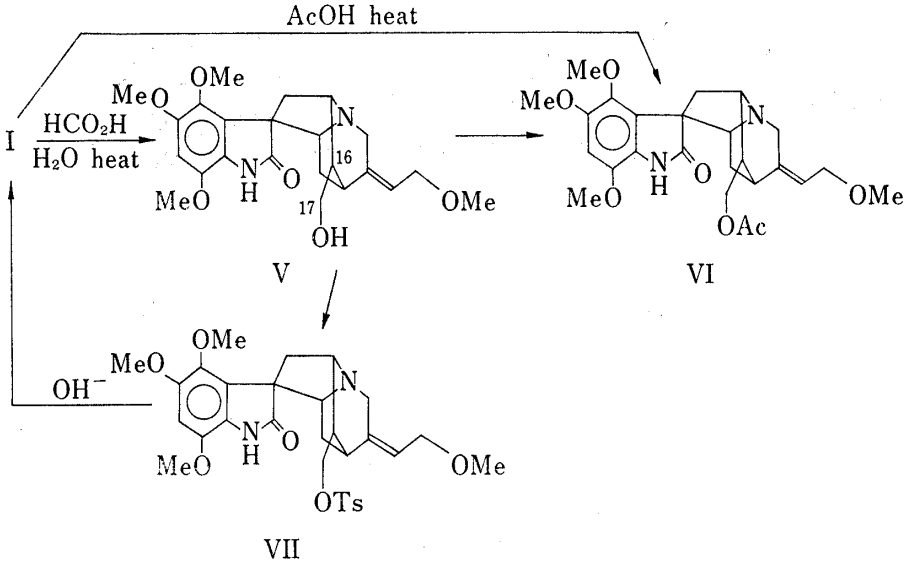


Chart 2

interesting to note that intramolecular version of this reaction easily proceeds in VII, in which $C_{(17)}$ carrying a leaving group lies spatially in the proximity of the carbonyl group of the oxindole moiety. In literature, a similar change was observed when gelsemine was treated with bromine.[9] Gardneramine (I) is the first natural indole alkaloid having an iminoether grouping. It should be noted that Scott suggested a possible role of this type of functionality for the skeletal change of corynanthé to strychnos type in the indole alkaloid biogenesis.[10]

Formation of acetate (VI) on treatment of I with hot acetic acid can be explained by acid catalyzed ring opening accompanied by concomitant nucleophilic attack of acetic acid molecule to $C_{(17)}$ carbon carrying the ether oxygen. This type of reactions is characteristic to the iminoether moiety of I and its derivatives, and several similar reactions were observed in the course of our structural work. (see below).

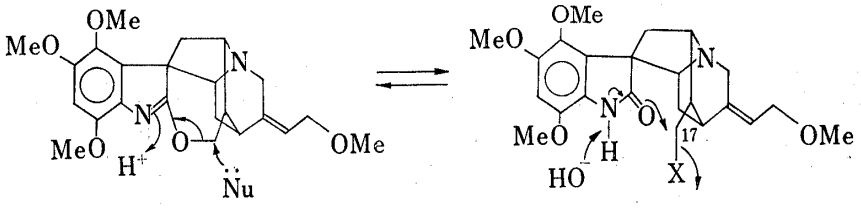
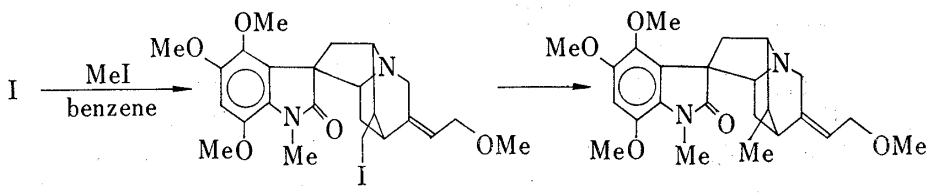


Chart 3

The NMR signal of $C_{(17)}$ methylene of V was observed as a doublet (J=7.5 Hz) at δ 3.98, which exhibited down field shift to δ 4.46 (d, J=7.8 Hz) in acetate (VI) and to δ 4.48 (d, J=8.7 Hz) in tosylate (VII), respectively, and thus suggested the adjacent carbon $C_{(16)}$ carried one hydrogen. This assumption was proved as follows.

When I was treated with excess of methyl iodide in dry benzene, a tertiary base (VIII), mp 143—145°, was obtained together with two quarternary salts, (IX) and (X). A signal due to a methyl group of N-methyl oxindole moiety was observed at δ 3.36 in the NMR spectrum of VIII. Reductive removal of the iodide residue of VIII by means of Zn in acetic acid afforded a halogen free oxindole (XI), in which the expected C-methyl signal was observed at δ 1.21 as a doublet with a coupling constant of 7.5 Hz. Structures of the two quarternary iodomethylates, (IX) and (X), are discussed in the later part of this paper.

9) R. Goutarel, M.-M. Janot, V. Prelog, R.P.A. Sneeden, and W.I. Taylor, *Helv. Chim. Acta*, **34**, 1139 (1951).
10) A.I. Scott, *Accounts Chem. Res.*, **3**, 151 (1970).



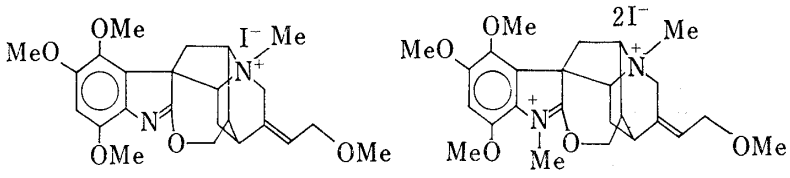


Chart 4

The above interesting addition reaction of methyl iodide to the oxindole iminoether system can be considered to occur in a similar way as the acetolysis reaction of I to give VI (Chart 2 and 3). To test generality of this reaction with methyl iodide, an iminoether (XIII) was synthesized through 3-ethyl-3-(3′-hydroxypropyl)-oxindole (XII). On the reaction of XIII with methyl iodide, the expected N-methyl oxindole (XIV) with iodide residue on the alkyl side chain was actually obtained. Details of this work will be published elsewhere.[11]

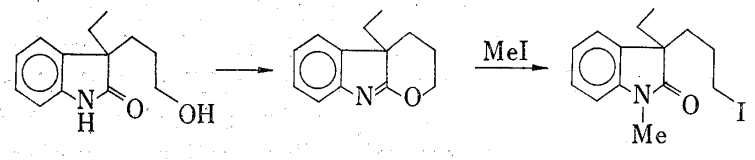


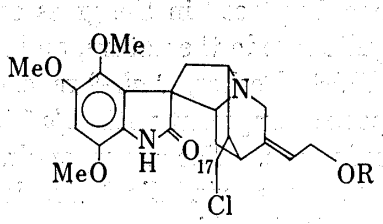
Chart 5

When treated in dilute hydrochloric acid under reflux, I yielded two chlorine-containing oxindoles (XV), $C_{23}H_{29}O_5N_2Cl$, and (XVI), $C_{22}H_{27}O_5N_2Cl$, both of which showed characteristic NMR signals due to $C_{(17)}$-methylene as respective doublets at δ 3.98 (J=8.0 Hz) (XV) and δ 3.97 (J=8.0 Hz) (XVI). Compound (XVI) was suggested to be a demethylation product of XV for no signal due to an aliphatic methoxyl group was observed in its NMR spectrum. In fact, treatment of the latter with dilute hydrochloric acid under reflux gave rise to XVI. The configuration of $C_{(19)}$=$C_{(20)}$ double bond of XV has been proved to be retained in the hydrolysis product (XVI). Full detail of this part has been described in our recent paper concerning the side chain geometry of several indole alkaloids.[5] Acetylation of XVI with acetic anhydride in pyridine gave an amorphous acetate (XVII).

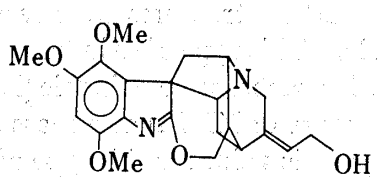
The above evidences and the NMR data summarized in Table I indicated the presence of the partial structure as shown.

Gardneramine (I) was regenerated from XV on treatment with ethanolic potassium hydroxide. When XVI was submitted to the same type base catalyzed dehydrochlorination reaction, the corresponding iminoether (XVIII) was obtained (Chart 3 and 6). This compound (XVIII) was found to be identical with an unknown base of *G. multiflora* Makino, which was previously reported under a tentative name of alkaloid G.[3a] Thus the structure of alkaloid G was revealed to be 18-demethyl gardneramine (XVIII), in which $C_{(19)}$=$C_{(20)}$ double bond had the same configuration as that of gardneramine (I) (see above).

11) S. Sakai, E. Yamanaka, T. Yokomizo, and M. Matsumoto, *Yakugaku Zasshi*, submitted.



XV : R=Me
XVI : R=H
XVII : R=Ac

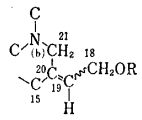


XVIII

Chart 6

TABLE I. NMR Data (δ)

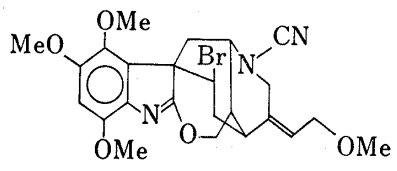
	$C_{18}H_2$	$C_{19}H$	$C_{21}H_2$	$C_{18}OR$
I (R=Me)	(3.90)[a]	5.39 t-t J=7.3, 2.5 Hz	(3.70)[a]	3.36 3H s
XV (R=Me)	(3.83)[a]	5.32 t-t J=6.7, 2.0 Hz	(3.75)[a]	3.31 3H s
XVI (R=H)	4.08 d J=6.7 Hz	5.36 t-t J=6.7, 2.0 Hz	(3.74)[a]	1.80 1H s
XVII (R=Ac)	4.50 d J=7.5 Hz	5.30 m	(3.76)[a]	2.04 3H s



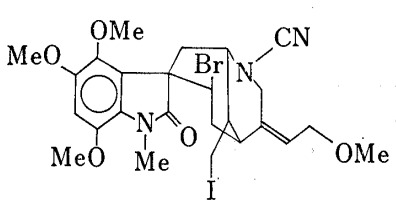
a) The corresponding signal is overlapped with other signal (s). The chemical shift values in the parenthesis were determined by the double resonance experiments.

Von Braun reaction of I yielded a cyano bromide (XIX), mp 214°, as the sole product. The ultraviolet (UV) and IR absorption curves of XIX indicated no change took place on the chromophore system of the starting material. When XIX was heated in glacial acetic acid, bromo and cyano groups were removed and $C_{(3)}$–$N_{(b)}$ bond was regenerated. At the same time the iminoether moiety was cleaved solvolytically and the product was oxindole (VI). This observation clearly demonstrated that no skeletal change occurred in the course of formation of XIX from I. This is an important fact, because X-ray structure determination was made on this cyano bromide (XIX) at the last stage of our structural work.[8]

Cleavage of the iminoether ring of the cyano bromide derivative (XIX) was successfully carried out by use of methyl iodide to give an $N_{(a)}$-methyl oxindole (XX), whose mass spectrum showed the molecular ion peaks at *m/e* 661 and 659 with equal intensities indicating the presence of a bromine atom and addition of one molecule of methyl iodide to XIX. The UV spectrum was oxindolic with the absorption maxima at 263 and 316 nm. The presence of an oxindolic carbonyl and an N-cyano groups was confirmed by the respective IR absorption bands at 1690 cm^{-1} and 2205 cm^{-1}. These observations clearly demonstrated that no participation of $N_{(b)}$ was required for the generation of the oxindole moiety from the parent compound, supporting the iminoether structure as the masked oxindole system.



XIX



XX

Chart 7

All the above partial structures were reasonably placed in the gross carbon skeleton of a sarpagine oxindole. This type of oxindole is quite rare in the nature, and the only example, as far as we know, is voacharotine oxindole (XXI).[12] Pecher, *et al.*[12] reported in their paper concerning the structure of XXI that a mass spectral fragment with *m/e* 172 (fragment a, in Chart 8) had a diagnostic value for this type of oxindoles. The corresponding fragment (fragment b, Chart 8) was observed in the mass spectra of our oxindolic derivatives at *m/e* 248 except for N-methylated oxindoles (for example VIII), in which the corresponding peak was observed at *m/e* 262 (248+CH_2). High resolution mass spectrometric determination revealed the elemental composition being $C_{13}H_{14}O_4N$ (*m/e* 248) in accordance with the fragment b. (Chart 8).

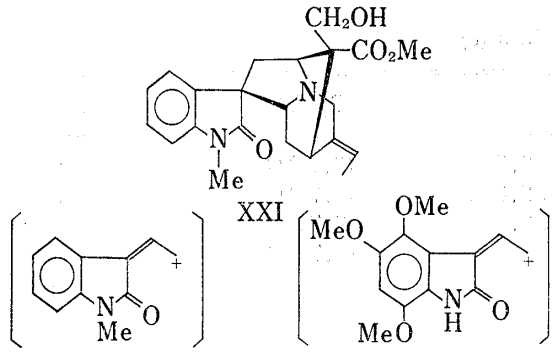


Chart 8

Chart 9

It is well known that 7-spiro-oxindoles epimerize at $C_{(7)}$ (and in some cases at $C_{(3)}$ also) to give equilibrated mixtures under various conditions.[13] All oxindolic derivarives of I, however, were obtained in only one forms as described above. No indication of epimerization at $C_{(7)}$ of the resulting oxindoles was observed. These facts strongly supported the proposed skeleton, in which participation of the lone pair electrons of $N_{(b)}$ was prohibited because of the position of $N_{(b)}$ at the bridge head of a quinuclidine ring. This stability of $C_{(7)}$ configuration toward equilibrium conditions was also noted in voacharotine oxindole (XXI).[12]

Posturation of the above fundamental skeleton to gardneramine (I) based partly on biogenetical consideration. As pointed out at the begining of this paper, all the accompanying bases of *G. nutans* Sieb. et Zucc. possess the sarpagine type basic skeleton, which would give rise to the proposed framework on biogenetic oxidation. Many *in vitro* examples of the similar type oxidative rearrangements of indole- to oxindole alkaloids are found in the literatures.[14] In those reactions bond migrations occur from α ($C_{(2)}$) to β ($_{(7)}$) positions on the indole nucleus and the resulting products are the corresponding 7-spiro-oxindoles.

As stated above, action of methyl iodide on I gave two quarternary iodomethylates, (IX) and (X), along with a tertiary addition product (VIII). (Chart 4). Repeated fractional crystallization afforded separated samples of a monoiodomethylate (IX), mp 207—209° (decomp.), and a diiodomethylate (X), mp 209° (decomp.). Monoiodomethylate (IX) showed a characteristic IR absorption band at 1597 cm^{-1}, which was ascribable to an indolenine type –C=N– bond. The UV spectrum showed an absorption curve which was similar to that of I. Therefore the chromophore system of I remained unchanged. Reduction of the C=N bond of IX was carried out by use of sodium borohydride. The resulting dihydro derivative (XXII), mp 225—228° (decomp.), was identified with a sample derived from 1,2-dihydrogardneramine (XXIII)[15] through iodomethylation. Thus the structure of IX was proved to be gardner-

12) J-C. Braekman, M. Tirions-Lampe, and J. Pecher, *Bull. Soc. Chim. Belges*, **78**, 523 (1969).
13) For example see: J.E. Saxton, in "The Alkaloids" ed. R.H.F. Manske, Academic Press, New York, London, 1965, Vol. 8, p. 67.
14) For example see: N. Finch, and W.I. Taylor, *J. Am. Chem. Soc.*, **84**, 3871 (1962); N. Finch, C.W. Gemenden, I.H-C. Hsu, and W.I. Taylor, *ibid.*, **85**, 1520 (1963); H. Zinnes, and J. Shavel, Jr., *J. Org. Chem.*, **31**, 1765 (1966).
15) N. Aimi, E. Yamanaka, J. Endo, S. Sakai, and J. Haginiwa, *Tetrahedron*, **29**, 2015 (1973).

amine-$N_{(b)}$-monoiodomethylate. The other iodomethylate (X) was analyzed as diiodomethylate. The most characteristic feature of this compound was a strong carbonyl-like IR absorption band at 1717 cm^{-1} (KBr), which caused much confusion in the early stage of our structural work. On reduction with sodium borohydride, this band disappeared and a reduction product (XXIV), mp 211° (decomp.), was given. The UV spectrum of XXIV having absorption maxima at 251 and 308 nm indicated the indolinic chromophore. The NMR signals due to $N_{(a)}$-methyl group at δ 2.82 and $C_{(2)}$-H at δ 4.17 gave additional support. These facts suggested that the above IR absorption band of X at 1717 cm^{-1} should be ascribed to an $-\overset{+}{N}=C-$ bond with an electronegative group at the α-position. An attempt to prepare compound (XXIV) from I through a different route was made. Thus 1,2-dihydrogardneramine (XXIII)[15] was methylated with HCHO–HCOOH to give an $N_{(a)}$-methyl derivative (XXV). An $N_{(a)}$-methyl oxindole (XXVI) was obtained as a by-product in this reaction, and the structure was confirmed by derivation from oxindole (V). (Chart 11). The formation mechanism of XXVI would be depicted as shown in Chart 11. The above compound (XXV) was then quarternalized by use of methyl iodide in dry benzene to give a methyliodide (XXVII), mp 239° (decomp.). The UV (λ_{max} 252 and 313.5 nm) and the NMR (δ 2.82 ($N_{(a)}$–Me) and δ 4.39 ($C_{(2)}$–H)) spectra agreed well with the structure XXVII. Its direct comparison (mp and IR) with XXIV, however, showed that they were not the same compound. Therefore, the two compounds must be stereoisomeric at $C_{(2)}$, which is the only centre of diastereoisomerism because the configurations of other asymmetric centers are fixed as a consequence of the rigid structure. This conclusion was confirmed by the observation that the mass spectra of the both compounds showed very similar fragmentation patterns and the circular dichroism (CD) spectra were nearly opposite. The Ph–N–C–O– systems of XXIV and XXVII were stable to acids or reduction conditions, and all the efforts to correlate each other have failed.

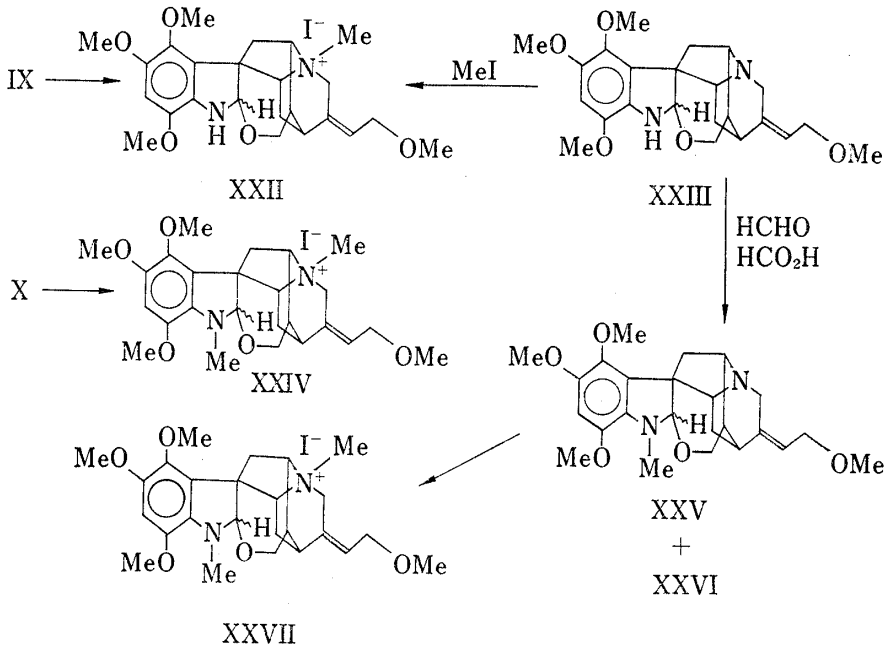


Chart 10

The observations described thus far allowed us to give gardneramine the structure I'. There still remained three problems to be solved, *eg.* 1) location of methoxyl groups on the aromatic ring, 2) geometry of the $C_{(19)}=C_{(20)}$ double bond and 3) the absolute configuration of the molecule. Next we tried to decide the positions of aromatic methoxyl groups by com-

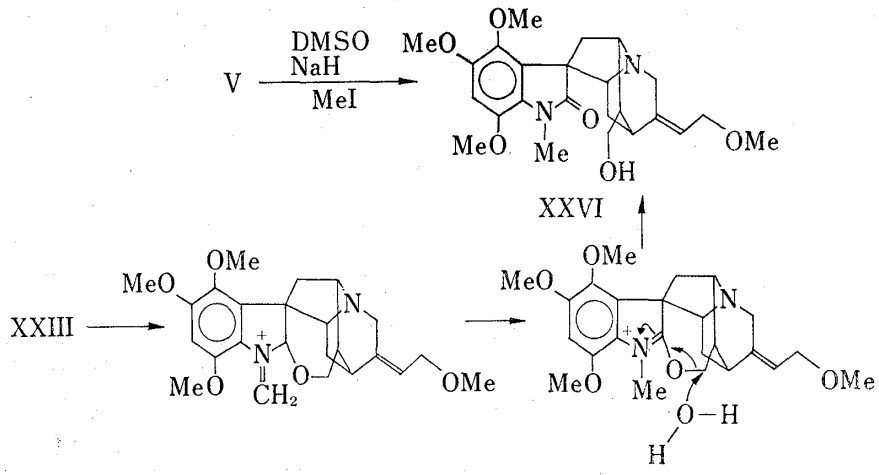
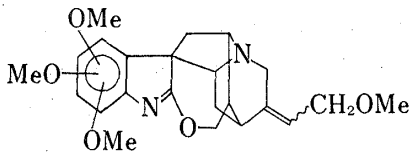


Chart 11

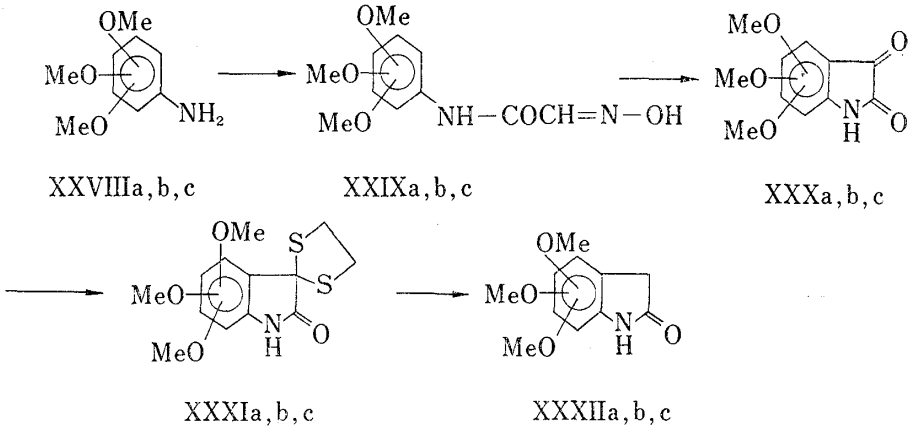
paring UV spectra of suitable derivatives of gardneramine with those of model compounds. To this aim syntheses of several trimethoxylated oxindoles were made.



Structure I′

Syntheses were carried out according to the general procedure to isatin derivatives.[16] As shown in Chart 12, respective trimethoxy anilines (XXVIII a—c) were converted to the corresponding isatins (XXX a—c) and their thioketal derivatives (XXXI a—c) were then desulfurized by Raney nickel to the desired oxindoles (XXXII a—c). It had been reported[17] that when ethanol was used for the solvent on desulfurization of isatin dithioketal the main product was the corresponding 3-ethyl oxindole. In the present experiments, however, the products of desulfurization of XXXI a and b in ethanol were almost exclusively normal oxindoles. This difference of reactivity would be due to steric effect of $C_{(4)}$ methoxyl group.

In this way, three out of four possible trimethoxy oxindoles were synthesized, but none of their UV spectra (see experimental) agreed with that of oxindolic derivatives of I. Un-



XXXIIa : 4, 6, 7-trimethoxyoxindole
XXXIIb : 4, 5, 6-trimethoxyoxindole
XXXIIc : 5, 6, 7-trimethoxyoxindole

Chart 12

16) C.S. Mervel and G.S. Hiers, "Organic Synthese," Coll. Vol. I. ed. by A.H. Blatt, John Wiley and Sons, Inc., New York, 1941, p. 327.
17) E. Wenkert and N.V. Bringi, *J. Am. Chem. Soc.*, **80**, 5575 (1958).

fortunately, 4,5,7-trimethoxy oxindole, having the substituents at the correct positions, could not be synthesized by this procedure, and therefore we failed to decide exact positions of the methoxyl groups on the aromatic ring of gardneramine (I).

The above mentioned three ambiguous features of the structure of gardneramine were consequently clarified with the aid of X-ray crystallography. As reported[8] structure analysis of XIX revealed that 1) the aromatic methoxyl groups were located at $C_{(9)}$, $C_{(10)}$, and $C_{(12)}$, 2) the configuration of $C_{(19)}$=$C_{(20)}$ was Z and 3) the absolute configuration was as shown by the structure I. The absolute configuration was in accord with empirical uniformity of the absolute configuration of the natural terpenoid indole alkaloids. It should be noted that $C_{(19)}$=$C_{(20)}$ double bond has Z type configuration. As far as we know, all the other natural indole alkaloids have the corresponding double bond of E type, and gardnermine (I) seems to be the first example of indole alkaloid with Z type side chain.

Experimental

All melting points were determined in glass capillary tubes using a H_2SO_4 bath, and are uncorrected. IR spectra were measured by a Model EPI-G3 Spectrophotometer (Hitachi Co.) and UV spectra by a Model EPS-3T Spectrophotometer (Hitachi Co.). NMR spectra were run using a JNM4H-100 or MH-100 NMR Instrument (Japan Electron Optics Co.) with tetramethylsilane (TMS) as an internal reference in $CDCl_3$ unless otherwise specified. Mass spectra were taken with a Model RMU-6E Mass Spectrometer (Hitachi Co.) using a direct inlet system. CD spectra were measured by a Model J-20 Recording Spectropolarimeter (Japan Spectroscopic Co.). A DIP-SL Automatic Polarimeter (Japan Spectroscopic Co.) was used for measurement of optical rotations.

Hydrolysis of I with aq. Formic Acid. Formation of Oxindole (V)——A solution of I (1.07 g) in 85% formic acid was refluxed on an oil bath for 6 hr. Evaporation of the solvent *in vacuo* yielded a syrupy residue which was dissolved in $CHCl_3$. The organic layer was washed with dil. $NaHCO_3$ and water. The crude amorphous product was chromatographed over Al_2O_3 (Woelm, grade II, 50 g). Elution with $CHCl_3$–MeOH (1%) afforded an amorphous foam (V) (0.98 g). Mass Spectrum *m/e*: 430 (M^+). NMR δ: 3.30 (3H, s, $C_{(18)}$–OMe); 3.70 (3H, s), 3.82 (6H, s) (Arom.-OMe×3); 3.98 (2H, d, J=7.5 Hz, $C_{(17)}H_2$); 5.29 (1H, t, J=7 Hz, $C_{(19)}H$); 6.44 (1H, s, Arom.-H); 7.88 (1H, broad s, NH). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3425 (NH, OH); 1705 (C=O). UV λ_{max}^{MeOH} nm: 253 (plateau) and 315. $\lambda_{max}^{MeOH+HCl}$ nm: 260 and 320.

Acetylation of V——The oxindole (V) 221 mg was acetylated in pyridine (2 ml) and Ac_2O (4 ml) under reflux for 1 hr. Ice water was added and the product was extracted with $CHCl_3$. The organic layer was washed with dilute Na_2CO_3 and subsequently with water. Evaporation of the solvent *in vacuo* gave a brownish syrup which was subjected to column chromatography on Al_2O_3 (Woelm, grade II, 20 g). Elution with benzene–$CHCl_3$ (1: 1) afforded acetate (VI) (88 mg) as an amorphous powder. *Anal.* Calcd. for $C_{25}H_{32}O_7N_2$: C, 63.56; H, 6.83; N, 5.93. Found: C, 63.46; H, 7.03; N, 5.93. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3425 (NH), 1715 (C=O). UV λ_{max}^{EtOH} nm (log ε): 220 (sh. 4.30), 252 (inf. 3.83) and 313 (3.60). NMR δ: 2.02 (3H, s, O·CO·CH_3); 3.31 (3H, s, $C_{(18)}$–OMe); 3.78 (3H, s), 3.82 (6H, s) (Arom.–OMe×3; 4.46 (2H, d, J=7.8 Hz, $C_{(17)}H_2$); 5.31 (1H, m, $C_{(19)}H$); 6.45 (1H, s, Arom. H); 7.58 (1H, broad s, NH).

Acetolysis of I——Gardneramine (I) 210 mg was heated in glacial acetic acid (10 ml) under reflux for 5 hr. The solvent was removed *in vacuo* to give a reddish syrup which was chromatographed over Al_2O_3 (Woelm, grade II, 20 g). Elution with benzene–$CHCl_3$ (1: 1) afforded an amorphous powder (197 mg) (VI). *Anal.* Calcd. for $C_{25}H_{32}O_7N_2$: C, 63.54; H, 6.83; N, 5.93. Found: C, 63.46; H, 6.95; N, 5.95. Mass Spectrum *m/e*: 472 (M^+, 37%), 457 (24), 413 (100), 248 (28), 233 (14). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3425 (NH), 1715 (C=O). UV λ_{max}^{MeOH} nm: 255 (inflection), 315. CD Spectrum $\Delta\varepsilon^{320}$: +1.53, $\Delta\varepsilon^{288}$: −3.58, $\Delta\varepsilon^{258}$: +13.3, $\Delta\varepsilon^{230}$: −56.4, $\Delta\varepsilon^{209}$: +56.4.

Rf values on thin-layer chromatography (TLC) (silica gel G, solvent system, benzene–EtOH–Et_2NH, (8: 1.5: 0.5)) and comparison of IR spectra proved that the acetolysis product of I was identical with the acetate of V (VI).

Tosylate (VII)——Compound V (80 mg) was dissolved in 2 ml of pyridine, and 350 mg of tosyl chloride was added to it. The solution was left at room temperature over night, and was poured over ice-water. Collection of the precipitate on a glass filter gave the crude product as an amorphous powder (57 mg), which showed a single spot on TLC. Recrystallization from ether gave colorless prisms, mp 158—165°. *Anal.* Calcd. for $C_{30}H_{36}O_8N_2S$: C, 61.64; H, 6.20; N, 4.79. Found: C, 61.64; H, 6.23; N, 4.94. IR ν_{max}^{KBr} cm^{-1}: 3320 (NH); 1720 (C=O); 1365, 1174 (OTs). UV λ_{max}^{MeOH} nm (log ε): 253 (inf., 3.81), 313 (3.58). NMR δ: 2.42 (3H, s, Me on Ts group); 3.29 (3H, s, $C_{(18)}$–OMe); 3.78—3.82 (9H, s, Arom.-OMe×3); 4.48 (2H, d, J=8.7 Hz, $C_{(17)}H_2$); 5.30 (1H, m, $C_{(19)}H$); 6.45 (1H, s, Arom.-H); 7.35, 7.87 (4H, A_2B_2, J=8 Hz, Arom.-4H on Ts group.)

Regeneration of I from Tosylate (VII)——To a solution of VII (36 mg) in EtOH (15 ml), 5 ml of aqueous KOH (12%) was added. The solution was stirred at room temperature for 4 hr. The usual work up afforded

39 mg of a crude product which was crystallized from ether to give prisms (9 mg). No depression of mp was observed on admixture with an authentic specimen of I. Comparison of behavior on TLC and IR spectrum with those of I also proved their identity.

Gardneramine Iodomethylates (VIII), (IX) and (X)——To a solution of I (1 g) in dry benzene, CH_3I (4 ml) was added gradually. After the solution was left overnight at room temperature, the resulting precipitates were collected and washed with benzene. The filtrate and washings were combined. Removal of the solvent and crystallization from MeOH afforded colorless needles of $N_{(a)}$-methyl oxindole (VIII) (349 mg), which showed mp 143—145°. *Anal.* Calcd. for $C_{24}H_{31}O_5N_2I$: C, 51.99; H, 5.63; N, 5.05. Found: C, 51.77; H, 5.58; N, 4.80. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1688 (C=O). UV λ_{max}^{MeOH} nm: 262 (sh.), 273 (sh.) and 316. NMR δ: 3.29 (3H, s, $C_{(18)}$–OMe or $N_{(a)}$–Me); 3.36 (3H, s, $N_{(a)}$–Me or $C_{(18)}$–Me); 3.74, 3.79, 3.81 (each 3H, s, Arom.-OMe×3); 5.30 (1H, m, $C_{(19)}$H), 6.46 (1H, s, Arom.-H). Mass Spectrum *m/e*: 554 (M^+, 1%), 539 (M^+—Me, 5), 427 (M^+—I, 100), 262 (fragment b+CH_2 (Chart 8), 28), 237 (9).

Fractional crystallization of the crude quarternary salts obtained above afforded two salts.

Monoiodomethylate (IX) 528 mg was obtained after recrystallization from MeOH–ether and subsequently from MeOH. Faintly yellow plates of mp 207—209° (decomp.) (IX) showed the following physical properties: *Anal.* Calcd. for $C_{24}H_{31}O_5N_2I$: C, 51.99; H, 5.63; N, 5.05; I, 22.89. Found: C, 51.76; H, 5.63; N, 4.87; I, 23.05. IR ν_{max}^{KBr} cm^{-1}: 1597 (C=N), UV $\lambda_{max}^{EtOH-H_2O(1:1)}$ nm (log ε): 222 (4.49), 265.5 (3.79) and 335 (3.78); λ_{max}^{2N-HC} nm: 224.5 (4.38), 256.5 (3.87) and 355 (3.78).

From the mother liquor of monoiodomethylate (IX), diiodomethylate (X) (206 mg) was obtained after repeated recrystallization from MeOH–ether or MeOH. Colorless prisms, mp 209° (decomp.). *Anal.* Calcd. for $C_{25}H_{34}O_5N_2I_2$: C, 43.12; H, 4.92; N, 4.02; I, 36.45. Found: C, 43.09; H, 5.05; N, 4.10; I, 35.97. IR ν_{max}^{KBr} cm^{-1}: 1717 $\left(-\overset{+}{N}=C-\right)$ (Me). UV λ_{max}^{EtOH} nm (log ε): 266 (3.75), 332 (3.56). NMR δ: 3.30, 3.33 (each 3H, s, $N^+_{(a)}$–Me and $C_{(18)}$–OMe); 3.85—3.90 (12H, Arom.-OMe×3 and $N^+_{(b)}$–Me).

$NaBH_4$ Reduction of Monoiodomethylate (IX)——To a solution of IX (100 mg) in MeOH (10 ml) and H_2O (2 ml), 150 mg of $NaBH_4$ was added portionwise. After the solution was refluxed for 1.5 hr, the solvent was removed *in vacuo* and the residue was dissolved in water. Extraction with $CHCl_3$ followed by the usual work up afforded dihydro derivative (XXII) (45 mg) as colorless needles. mp 225—228° (decomp.). *Anal.* Calcd. for $C_{24}H_{33}O_5N_2I \cdot 1/2H_2O$: C, 50.98; H, 5.97; N, 4.95; I, 22.44. Found: C, 51.11; H, 6.03; N, 4.80; I, 22.61. IR ν_{max}^{KBr} cm^{-1}: 3350 (NH). UV λ_{max}^{EtOH} nm (log ε): 245 (3.91), 310 (3.59). $\lambda_{max}^{2N\ HCl}$ nm (log ε): 226 (4.33), 289.5 (3.53). NMR δ: 3.33 (3H, s, $C_{(18)}$–OMe); 3.74—3.87 (12H, Arom.-OMe×3, and $N^+_{(b)}$-Me), 4.25 (1H, broad s, NH), 5.20 (1H, s, $C_{(2)}$H).

1,2-Dihydrogardneramine $N_{(b)}$-Monoiodomethylate (XXII)——To a solution of 1,2-dihydrogardneramine (XXIII)[15] (100 mg) in 20 ml of dry benzene, 2.5 ml of CH_3I was added. The solution was stirred at room temperature for 16.5 hr. The resulting crystalline precipitate was collected and was recrystallized from MeOH–ether to give colorless needles of mp 235—235.5° (decomp.) which weighed 120 mg. Comparison of mixed mp and IR spectra proved its identity with the reduction product of monoiodomethylate (IX).

1-Methyl-1,2-dihydrogardneramine (XXV)——1,2-Dihydrogardneramine (XXIII) 400 mg was heated in 4 ml of HCHO (>37%) under reflux for 2 hr. After addition of 8 ml of HCOOH (99%) the solution was refluxed for 1.75 hr under N_2. The solvent was removed *in vacuo* and the residue was treated in the usual way to give a syrup (530 mg), which was then subjected to Al_2O_3 column chromatography. From the fraction eluted with benzene–$CHCl_3$ (4: 1), 226 mg of N-methylated derivative (XXV) was obtained after recrystallization from ether–hexane. From the fraction eluted with $CHCl_3$ 224 mg of N-methyl oxindole (XXVI) was obtained, which was further purified with silica gel column chromatography to give 83 mg of the pure sample. Recrystallization from acetone gave colorless needles (XXVI) (27 mg). XXV: mp 107—109°. UV λ_{max}^{EtOH} nm (log ε): 250.5 (3.90), 307 (3.54). λ_{max}^{2N-HCl} nm (log ε): 228 (3.93), 236 (sh., 3.90), 287 (3.57). NMR δ: 2.85 (3H, s, $N_{(a)}$–Me); 3.30 (3H, s, $C_{(18)}$–OMe); 3.78—3.82 (9H, Arom.–OMe×3); 4.33 (1H, s, $C_{(2)}$H). Mass Spectrum *m/e*: 428 (M^+, 100%), 234 (67), 204 (25). XXVI: mp 151—152.5°. *Anal.* Calcd. for $C_{24}H_{32}O_6N_2$: C, 64.84; H, 7.26; N, 6.30. Found: C, 64.94; H, 7.33; N, 6.24. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1693 (C=O). UV λ_{max}^{EtOH} nm (log ε): 256 (3.88), 313.5 (3.62). NMR δ: 3.28, 3.36 (each 3H, s, $C_{(18)}$–OMe and $N_{(a)}$–Me). Mass Spectrum *m/e*: 444 (M^+, 100%), 429 (89), 413 (75), 399 (58), 262 (50).

$N_{(a)}$-Methyl Oxindole (XXVI)——Oxindole (V) (200 mg) was added to a preheated solution of NaH (50%) (26 mg) in 5 ml of dimethyl sulfoxide (DMSO). After the solution was stirred for 15 min at room temperature, 0.04 ml of CH_3I was added. The solution was then stirred at room temperature for 3.5 hr under N_2. The usual work up afforded a crude product which was chromatographed over Al_2O_3 (15 g). Amorphous powder (185 mg) obtained from $CHCl_3$ eluate was identified with the sample obtained from the reaction of XXIII and CH_2O–HCOOH.

1-Methyl-1,2-dihydrogardneramine Iodomethylate (XXVII)——To a solution of XXV (35 mg) in dry benzene (2 ml), 0.5 ml of CH_3I was added and the solution was left for 30 min at room temperature. Crystals separated from the solution were collected and recrystallized from MeOH–ether to give colorless needles (38 mg) (XXVII). mp 239° (decomp.). *Anal.* Calcd. for $C_{25}H_{35}O_5N_2I \cdot 1/2H_2O$: C, 51.82; H, 6.09; N, 4.83. Found: C, 52.04; H, 6.11; N, 4.83. UV λ_{max}^{EtOH} nm (log ε): 252 (3.92), 313.5 (3.53). $\lambda_{max}^{2N\ HCl}$ nm (log ε): 226

(4.31), 289 (3.52). NMR δ: 2.82 (3H, s, $N_{(a)}$-Me); 3.35 (3H, s, $C_{(18)}$-OMe); 3.78—3.88 (each 3H, s, $\times 4$, Arom.-OMe$\times 3$ and $N^+_{(b)}$-Me); 4.39 (1H, s, $C_{(2)}$-H). Mass Spectrum *m/e*: 442 (M^+-HI, 3%), 428 (M^+-CH_3I, 100), 414 (13), 413 (9), 399 (14), 397 (12), 383 (6), 370 (10), 369 (9). CD Spectrum (c=0.08, MeOH): $\Delta\varepsilon^{300}$: −0.25, $\Delta\varepsilon^{274}$: −0.2, $\Delta\varepsilon^{248}$: +5.0.

$NaBH_4$ Reduction of Diiodomethylate (X)——$NaBH_4$ 350 mg was added to a solution of diiodomethylate (X) (230 mg) in 30 ml of EtOH and the solution was stirred at room temperature for 3.5 hr. After addition of a few drops of AcOH, the solvent was removed *in vacuo* to about a half volume. Water was added and the solution was extracted with $CHCl_3$. The organic layer was washed with water, dried and the solvent was removed under reduced pressure. Recrystallization from MeOH–ether gave 65 mg of XXIV, which showed mp 211° (decomp.). *Anal.* Calcd. for $C_{25}H_{35}O_5N_2I \cdot 1/2H_2O$: C, 51.82; H, 6.09; N, 4.83; I, 21.90. Found: C, 51.99; H, 6.18; N, 4.83; I, 22.37. UV λ_{max}^{EtOH} nm (log ε): 251 (3.95), 308.5 (3.59). $\lambda_{max}^{2N\ HCl}$ nm (log ε): 226 (4.31), 287.5 (3.56). NMR δ: 2.82 (3H, s, $N_{(a)}$-Me); 3.35 (3H, s, $C_{(18)}$-OMe); 3.59 (3H, s, $N^+_{(b)}$-Me), 3.8—4.0 (Arom.-OMe$\times 3$); 4.17 (1H, s, $C_{(2)}$H). Mass Spectrum *m/e*: 442 (M^+-HI, 4%), 428 (M^+—MeI, 100), 414 (10), 413 (16), 399 (8), 397 (34), 383 (12), 370 (7), 369 (21). CD Spectrum (c=0.08, MeOH): $\Delta\varepsilon^{308}$: +1.1, $\Delta\varepsilon^{272}$: +0.5, $\Delta\varepsilon^{244}$: −4.3.

Treatment of I with Dilute Hydrochloric Acid—Gardneramine (I) 1.194 g in 2N HCl (20 ml) was refluxed on an oil bath for 3 hr. The solution was basified with aq. Na_2CO_3 (20%) and the resulting amorphous precipitate was extracted with $CHCl_3$. The organic layer was washed with water and dried over anhydrous Na_2SO_4. Removal of the solvent afforded a colorless powder, which was submitted to column chromatographic separation using Al_2O_3 (Brockmann, activity II—III, 80 g). The first eluate with benzene–$CHCl_3$ (2: 1) and (1: 1) was chromatographed over another column of Al_2O_3 (Woelm, activity II, 80 g) to give XV (461 mg) as an amorphous powder. Compound XVI (234 mg) was obtained from the $CHCl_3$ eluate of the first column and was similarly purified through another column of Al_2O_3 (Woelm, activity II, 50 g). XV was amorphous powder. *Anal.* Calcd. for $C_{23}H_{29}O_5N_2Cl$: C, 61.53; H, 6.51; N, 6.24. Found: C, 61.50; H, 6.67; N, 6.27. IR Spectrum $\nu_{max}^{CHCl_3}$ cm^{-1}: 3430 (NH), 1705 (C=O). UV λ_{max}^{MeOH} nm: 254 (sh.), 312. Mass Spectrum *m/e*: 450 (M^++2, 0.4%), 448 (M^+, 1.3), 413 (M^+—Cl, 100), 412 (M^+—HCl, 28), 248 (23), 233 (10). NMR δ: 3.31 (3H, s, $C_{(18)}$-OMe); 3.75 (3H, s), 3.81 (6H, s) (Arom.-OMe$\times 3$); 3.98 (2H, d, J=8.0 Hz, $C_{(17)}H_2$); 5.32 (1H, t-t, J=6.7 and 2.0 Hz, $C_{(19)}$H); 6.45 (1H, s, Arom.-H); 7.80 (1H, broad s, NH). XVI: mp 106—111° (from ether–benzene). *Anal.* Calcd. for $C_{22}H_{27}O_5N_2Cl \cdot 1/2\ H_2O$: C, 59.53; H, 6.35; N, 6.31. Found: C, 59.40; H, 6.44; N, 6.57. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3430 (NH, OH), 1710 (C=O). UV λ_{max}^{MeOH} nm (log ε): 255 (sh, 3.93), 313 (3.71). Mass Spectrum *m/e*: 436 (M^++2, 1%), 434 (M^+, 4), 399 (M^+—Cl, 100), 248 (24), 233 (11). High resolution measurement of the fragment peak of *m/e* 248: Calcd. for $C_{13}H_{14}O_4N$; 248.0923. Observed; 248.0950. NMR δ: 3.75 (3H, s), 3.84 (6H, s) (Arom.-OMe$\times 3$); 3.97 (2H, d, J=8.0 Hz, $C_{(17)}H_2$); 4.08 (2H, d, J=6.7 Hz, $C_{(18)}H_2$); 5.36 (1H, t-t, J=6.7 and 2.0 Hz, $C_{(19)}$H); 6.44 (1H, s, Arom.-H); 8.20 (1H, broad s, NH).

Formation of XVI from XV——A solution of 95 mg of XV in 15 ml of 2N HCl was heated under reflux for 24 hr. The reaction solution was treated in a similar manner as described above to give a crude product (80 mg). After purification with Al_2O_3 column chromatography, demethylation product XVI (48 mg) was obtained together with the unchanged XV (10 mg).

Acetylation of XVI——Compound XVI (111 mg) was acetylated with pyridine (1 ml) and Ac_2O (2 ml) at room temperature for 2 days. Water was added to the reaction mixture and the solution was basified with aq. Na_2CO_3. Extraction with $CHCl_3$ afforded a colorless syrup which was chromatographed over Al_2O_3 (Woelm, activity II, 20 g). From the fraction eluted with benzene–$CHCl_3$ (1: 1) 95 mg of XVII was obtained as an amorphous powder. Mass Spectrum *m/e*: 440 (M^+, 100%), 381 (17), 248 (22), 233 (11). NMR δ: 2.04 (3H, s, OAc); 3.77 (3H, s), 3.83 (6H, s) (Arom.-OMe$\times 3$); 3.95 (2H, d, J=8 Hz, $C_{(17)}H_2$); 6.48 (2H, d, J=6 Hz, $C_{(18)}H_2$); 5.30 (1H, m, $C_{(19)}$H); 6.45 (1H, s, Arom.-H); 7.55 (1H, broad s, NH).

Alkali Treatment of XV——A solution of XV (76 mg) in EtOH (5 ml) containing 3% KOH was refluxed for 3 hr. The usual work up afforded a syrupy residue, from which 43 mg of crystals was obtained by treating with ether. Recrystallization from ether gave 24 mg of prisms which melted at 131—132°. Mixed mp and comparison of the IR spectra proved the identity with authentic gardneramine (I).

Alkali Treatment of XVI. Formation of 18-Demethylgardneramine (Alkaloid G) (XVIII)——Alcohol XVI (94 mg) was refluxed in 3% KOH–EtOH under N_2 for 5 hr. Purification of the crude product on Al_2O_3 chromatography afforded 72 mg of an amorphous powder (XVIII). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3350 (OH), 1582 (C=N). UV λ_{max}^{MeOH} nm: 264, 279 (sh.), 325. Mass Spectrum *m/e*: 398 (M^+, 100%). NMR δ: 3.85, 3.90, 3.94 (each 3H, s, Arom–OMe$\times 3$); 4.12 (2H, d, J=7 Hz, $C_{(18)}H_2$); 4.46 (2H, d, J=2 Hz, $C_{(17)}H_2$); 5.42 (1H, broad t, J=7 Hz, $C_{(19)}$H), 6.47 (1H, s, Arom.-H). Comparison of the IR spectra proved the identity of XVIII with an amorphous base, alkaloid G, isolated from *G. multiflora* Makino.[3]

Gardneramine Cyanobromide (XIX)——To a solution of I (575 mg) in dry benzene (15 ml), 200 mg of cyanogen bromide was added and the solution was refluxed for 2 hr. As a small amount of the unchanged I remained in the reaction solution as evidenced by TLC, 200 mg of cyanogen bromide was added and the solution was further refluxed for 1.5 hr. After the volatile materials were removed *in vacuo*, the residue was dissolved in CH_2Cl_2. The organic layer was then washed with water, dried over anhydrous Na_2SO_4 and the solvent was removed *in vacuo* to give 701 mg of a yellowish powder. Crystallization from EtOH afforded

colorless needles (XIX). The yield was 260 mg. mp 214°. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 2240 (CN), 1590 (C=N). UV λ_{max}^{MeOH} nm: 263, 283, 323. Mass Spectrum *m/e*: 519 (M^++2, 100%), 517 (M^+, 100). NMR δ: 3.39 (3H, s, $C_{(18)}$-OMe); 3.87, 3.93, 4.01 (each 3H, s, Arom.-OMe×3); 4.25 (1H, d, J=12 Hz, $C_{(17)}$H); 4.57 (1H, d-d, J=12 and 5 Hz, $C_{(17)}$H); 5.03 (1H, m, $C_{(3)}$H); 5.69 (1H, t, J=6 Hz, $C_{(19)}$H); 6.48 (1H, s, Arom.-H). *Anal.* Calcd. for $C_{24}H_{28}O_5N_3Br$: C, 55.60; H, 5.44; N, 8.10. Found: C, 55.63; H, 5.40; N, 8.25.

Reaction of XIX with Acetic Acid. Formation of VI——A solution of 50 mg of XIX in 4 ml of AcOH was heated under reflux for 4 hr. The solvent was removed *in vacuo* and the residue was extracted with $CHCl_3$. The usual work up afforded a syrup which was then subjected to preparative TLC (Silica gel G, the solvent system: benzene–EtOH–Et_2NH 8: 1.5: 0.5). The main product was identified as oxindole (VI) by comparison of their IR spectra and *Rf* values on TLC.

Reaction of XIX with CH_3I——A solution of XIX (100 mg) in dry benzene (40 ml) containing 1.5 ml of CH_3I was refluxed for 20 hr. Removal of the volatile materials afforded a glassy residue, which was chromatographed on Al_2O_3 (Woelm, activity I, 30 g). Elution with benzene–$CHCl_3$ (1: 9) afforded 114 mg of XX as an amorphous powder. IR ν_{max}^{CHCl} cm^{-1}: 2205 (CN), 1690 (C=O). UV λ_{max}^{MeOH} nm: 263, 316. Mass Spectrum *m/e*: 661 (M^++2, 100%), 659 (M^+, 100). NMR δ: 3.33 (3H, s, $C_{(18)}$-OMe or $N_{(a)}$-Me), 3.39 (3H, s, $N_{(a)}$-Me or $C_{(18)}$-OMe); 3.80, 3.83, 4.01 (each, 3H, s, Arom.-OMe×3); 5.10 (1H, d-d. J=11 and 2 Hz, $C_{(3)}$H); 5.68 (1H, t, J=7 Hz, $C_{(19)}$H), 6.51 (1H, s, Arom.-H).

2,3,5-Trimethoxyaniline Isonitrosoacetate (XXIXa)——A solution of 2,3,5-trimethoxyaniline (XXVIIIa)[18] (1.54 g) in 12% HCl (3 ml) was added to a solution of chloral hydrate (1.54 g) and ammonium acetate (12.7 g) in water (20 ml). After made homogeneous by adding 30 ml of 70% EtOH–H_2O, the solution was left at room temperature for 1 hr. Hydroxyl amine hydrochloride (2.1 g) in water (8.5 ml) was added and the mixture was heated first gently at 80° and then under reflux for 3 hr. A white precipitate separated from the reaction mixture on ice-cooling was collected.

The yield was 520 mg (24%). An analytical sample recrystallized from MeOH, colorless needles, mp 190—192°. *Anal.* Calcd. for $C_{11}H_{14}O_5N_2$: C, 51.96; H, 5.55; N, 11.02. Found: C, 51.64; H, 5.21; N, 11.07. UV λ_{max}^{MeOH} nm: 283. λ_{min}^{MeOH} nm: 265.

From the mother solution of the above precipitate (XXIXa), unchanged aniline (XXVIIIa) was recovered.

4,6,7-Trimethoxyisatin (XXXa)——Under dry condition, finely powdered XXIX a (732 mg) was gradually added to warm polyphosphoric acid (8.9 g) (bath temperature: 60°). The dark red colored solution was further stirred at 75—80° for 1 hr. From the ice-cooled reaction mixture orange yellow crystals separated. The crude material weighed 563 mg. An additional amount (103 mg) of the isatin (XXXa) was obtained by extraction with $CHCl_3$. Recrystallization from BuOH afforded orange yellow needles of mp 248—250°. IR ν_{max}^{KBr} cm^{-1}: 3170 (NH); 1740, 1715 (C=O). UV λ_{max}^{MeOH} nm: 364. Mass Spectrum *m/e*: 237 (M^+).

4,6,7-Trimethoxyisatin Ethylene Dithio Ketal (XXXIa)——To a solution of ethanedithiol (0.65 ml) and BF_3–ether (1.08 ml) in AcOH (10.8 ml), isatin (XXXa) (726 mg) was added, and the solution was left at room temperature. On addition of ice cracks to the reaction mixture, the crude material (885 mg) (92%) separated, which was crystallized from MeOH to form colorless cubes, mp 230—232°. *Anal.* Calcd. for $C_{13}H_{15}O_4NS_2$: C, 49.84; H, 4.82; N, 4.47. Found: C, 49.95; H, 4.78; N, 4.60. IR ν_{max}^{KBr} cm^{-1}: 3170 (NH), 1700 (C=O). UV λ_{max}^{MeOH} nm (log ε): 303 (3.73). Mass Spectrum *m/e*: 313 (M^+).

4,6,7-Trimethoxyoxindole (XXXIIa)——To a solution of the dithio ketal (XXXIa) (676 mg) in EtOH (50 ml), Raney Ni W-7 (prepared from 14.6 g of the alloy) suspended in EtOH (70 ml) was added. After the reaction mixture was refluxed for 3.5 hr at 95°, EtOH was removed from the filtrate, and the residue was passed through an Al_2O_3 (2 g) column using $CHCl_3$–MeOH (1: 1) as the elution solvents. The crude yield was 279 mg (58%). Recrystallization from MeOH gave XXXIIa as colorless pillars of mp 215—218°. *Anal.* Calcd. for $C_{11}H_{13}O_4N$: C, 59.18; H, 5.87; N, 6.28. Found: C, 59.06; H, 5.67; N, 6.12. UV λ_{max}^{MeOH} nm (log ε): 220 (4.40), 281 (3.36, sh), 289 (3.37). Mass Spectrum *m/e*: 223 (M^+). NMR δ (DMSO-d_6): 3.28 (2H, s, $C_{(3)}H_2$); 3.64, 3.78, 3.80 (each 3H, s, Arom.-OMe×3); 6.27 (1H, s, $C_{(5)}$H); 10.42 (1H, s, NH).

4,5,6-Trimethoxyisatin (XXXb)——Hydroxylamine hydrochloride (1 g) in water (4 ml) was added to a solution of 3,4,5-trimethoxyaniline[19] (XXVIIIb) (752 mg), 10% HCl (3 ml) and chloral hydrate (750 mg) in 10 ml of water. After addition of 6.1 g of ammonium acetate and EtOH (10 ml) the solution was stirred under heating (80°) for 1.5 hr. Removal of the solvents followed by drying in a dessicator afforded a brownish black residue. The above reaction product was then dissolved in conc. H_2SO_4 and the solution was stirred at 50° for 1 hr. After the solution was poured over ice-water, the reaction product was extracted with a mixture of benzene–$CHCl_3$ (1: 1). Crystallization of the resulting product from EtOH gave 4,5,6-trimethoxyisatin (XXXb) (417 mg, 43%) as orange needles, mp 213—217° (decomp.). IR ν_{max}^{KBr} cm^{-1}: 3300 (NH); 1750, 1700 (C=O). The same compound has been synthesized from XXVIIIb *via* a different route,[20] (Lit. mp 194—195°).

18) H. Richtzenhain and P. Nippus, *Chem. Ber.*, **82**, 408 (1949).
19) C. Graebe and M. Suter, *Ann. Chem.*, **340**, 222 (1905).
20) F. Benington, R.D. Morin, and L.C. Clark, Jr., *J. Org. Chem.*, **20**, 1454 (1955).

4,5,6-Trimethoxyisatin Ethylene Dithio Ketal (XXXIb)——4,5,6-Trimethoxyisatin (XXXb) was converted to the corresponding dithio ketal (XXXIb) in a similar manner as above with a yield of 85%. The mp was 231—233° (decomp). *Anal.* Calcd. for $C_{13}H_{15}O_4NS_2$: C, 49.84; H, 4.82; N, 4.47. Found: C, 49.60; H, 4.84; N, 4.31.

4,5,6-Trimethoxyoxindole (XXXIIb)——Desulfurization was carried out using Raney Ni in EtOH in a similar way as described for XXXIIa. Purification of the reaction product through a column of Al_2O_3 afforded XXXIIb in 27% yield. Colorless pillars from MeOH, mp 156—156.5°. *Anal.* Calcd. for $C_{11}H_{13}O_4N$: C, 59.18; H, 5.87; N, 6.28. Found: C, 58.96; H, 5.85; N, 6.19. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3440, 3200 (NH); 1700 (C=O). UV λ_{max}^{MeOH} nm (log ε): 216 (4.38), 261 (3.80). NMR δ: 3.54 (2H, s, $C_{(3)}H_2$); 3.78, 3.82, 3.96 (each 3H, s, Arom.-OMe×3); 6.29 (1H, s, $C_{(7)}H$); 9.98 (1H, s, NH). Mass Spectrum *m/e*: 223 (M^+).

5,6,7-Trimethoxyisatin (XXXc)——Using 2, 3,4-trimethoxyaniline (XXVIIIc)[21] as the starting material, 5,6,7-trimethoxyisatin (XXXc) was synthesized according to the literature.[22] mp 196—198° (from isoamyl alcohol) (Lit. 175—177°). NMR δ: 3.82, 3.92, 4.00 (each 3H, s, Arom.-OMe×3); 6.92 (1H, s, $C_{(4)}H$), 8.00 (1H, s, NH). Mass Spectrum *m/e*: 238 (M^+).

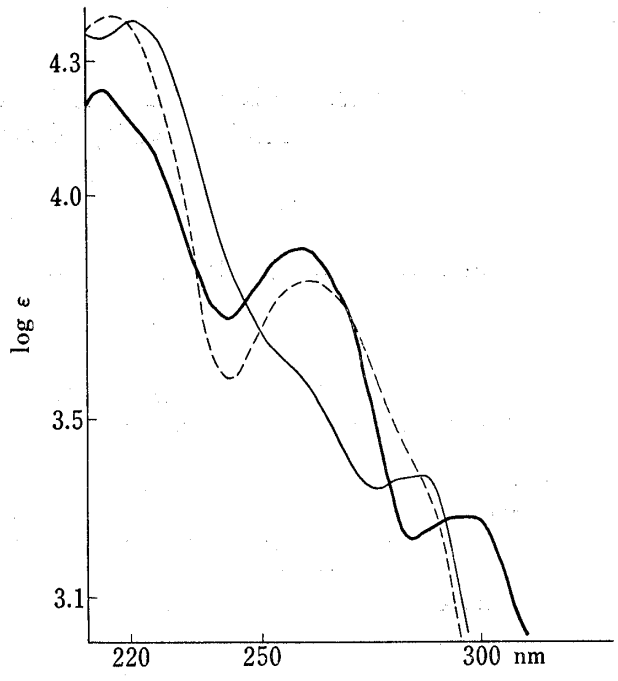


Fig. 2

———: XXXIIa
- - - - - - - -: XXXIIb
——————: XXXIIc

5,6,7-Trimethoxyisatin Ethylene Dithio Ketal (XXXIc)——In a similar manner as described above, the isatin (XXXc) was converted to the corresponding dithio ketal (XXXIc) with a yield of 48%. mp 166—167° (from MeOH). *Anal.* Calcd. for $C_{13}H_{15}O_4NS_2$: C, 49.84; H, 4.79; N, 4.47. Found: C, 49.89; H, 4.83; N, 4.30. Mass Spectrum *m/e*: 313 (M^+).

5,6,7-Trimethoxyoxindole (XXXIIc)——To a solution of XXXIc (400 mg) in dioxane (20 ml), Raney Ni W-7 prepared from 4 g of the alloy was added and the reaction mixture was refluxed for 3 hr under stirring. Crystallization of the product from benzene afforded 5,6,7-trimethoxyoxindole (XXXIIc) which showed the mp of 153—155°. The yield was 135 mg (47%). mp 153—155°. *Anal.* Calcd. for $C_{11}H_{13}O_4N$: C, 59.18; H, 5.87; N, 6.28. Found: C, 59.06; H, 5.67; N, 6.12. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3430 (NH); 1730, 1703 (C=O). Mass Spectrum *m/e* 223 (M^+). UV λ_{max}^{MeOH} nm (log ε): 259 (3.97), 295 (3.45). NMR δ: 3.49 (2H, s, $C_{(3)}H_2$); 3.80, 3.85, 3.92 (each 3H, s, Arom.-OMe×3); 6.60 (1H, s, $C_{(4)}H$); 8.60 (1H, s, NH).

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