

160. Yoshio Arata and Tsutomu Ohashi : Constituents of
Rhizoma nupharis. XXII.*¹ Structure of Nuphamine.

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We have already isolated sesquiterpene alkaloids, nupharidine,¹⁾ deoxynupharidine,¹⁾ dehydro-deoxynupharidine²⁾ and nupharamine³⁾ from the roots of *Nuphar japonicum* DC. and clarified their structures. The isolation of a new alkaloid from the roots and its structural determination will be described in this report.*³

The oily alkaloidal mixture obtained from the roots was subjected to repeated fractional distillation. An alkaloid, C₁₅H₂₃O₂N (I), b.p.₃ 150~153°, [α]_D²⁰ -60.48°, which was purified through a crystalline picrolonate, C₁₅H₂₃O₂N·C₁₀H₈O₅N₄, m.p. 159~160°, behaved differently on the thin-layer chromatography from the known alkaloids isolated from *Nuphar japonicum* DC., as shown in the experimental part, so that the alkaloid (I) has been named nuphamine.

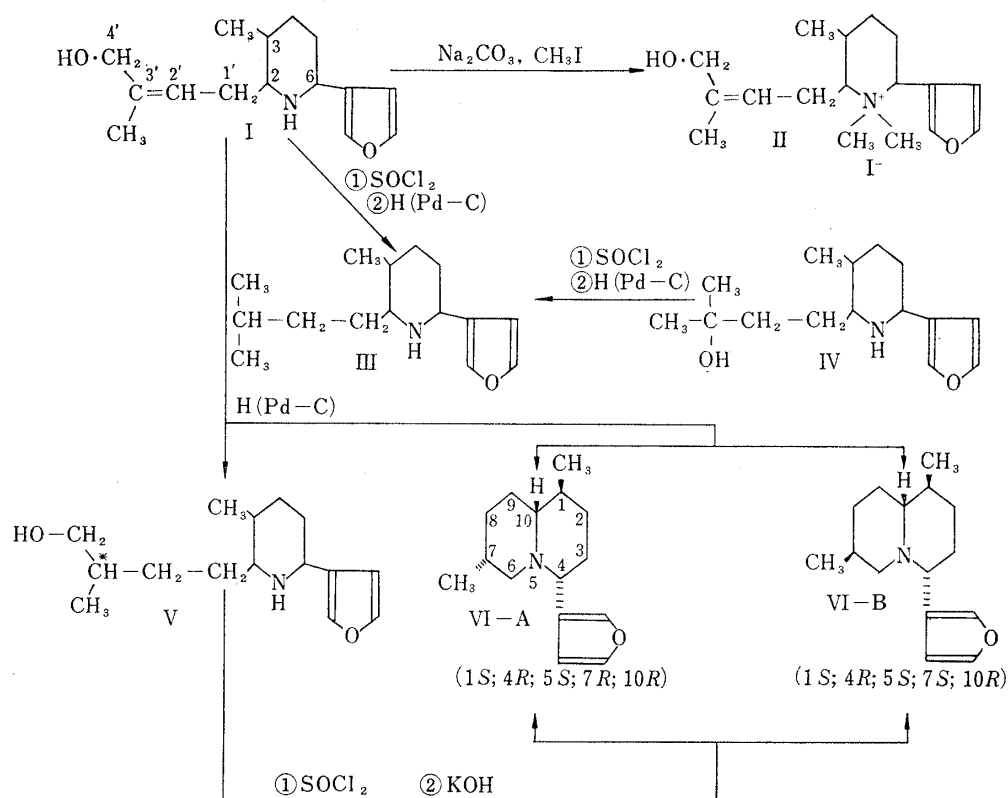


Chart 1.

*¹ Part XXI. Y. Arata : This Bulletin, 13, 907 (1965).

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*³ The preliminary paper has already appeared. Y. Arata, *et al.* : *Ibid.*, 13, 392 (1965).

1) Y. Arata, *et al.* : *Yakugaku Zasshi*, 66B, 138 (1946), 77, 236 (1957); This Bulletin, 10, 675 (1962); *Rep. Pharm. Kanazawa, Japan*, 12, 39 (1962); I. Kawasaki, *et al.* : *Chem. Bull. Japan*, 35, 697 (1962); O.F. Bohlmann : *Ber.*, 94, 3151 (1961).

2) Y. Arata : This Bulletin, 13, 907 (1965).

3) Y. Arata, *et al.* : *Yakugaku Zasshi*, 77, 792 (1957), 79, 729, 734 (1959), I. Kawasaki, *et al.* : *Bull. Chem. Soc. Japan*, 36, 1474 (1963).

The infrared absorption spectrum of nuphamine exhibited bands ascribable to hydroxyl and/or N-H group and the furan ring, the latter being also shown by the ultraviolet absorption maximum at $210.5\text{ m}\mu$ with the extinction coefficient of 7700. When I was treated with methyl iodide in the presence of sodium carbonate, the formation of N-methyl methiodide (II), $\text{C}_{17}\text{H}_{28}\text{O}_2\text{NI}$, m.p. 164° was observed and this fact suggested the secondary amine function in nuphamine.

Nuphamine (I) was chlorinated with thionyl chloride in anhydrous ether and the resulting chloro derivative was hydrogenated over palladium-carbon to form dihydrodeoxy compound (III), $\text{C}_{15}\text{H}_{25}\text{ON}$, $[\alpha]_D^{25} -63.56^\circ$. The infrared spectrum of III was completely identical with that of (–)-desoxynupharamine,^{3,4} derived from (–)-nupharamine (IV). III gave hydrochloride m.p. 233° , perchlorate, m.p. 128° and picrolonate, m.p. 176° , each of which did not show any melting point depression by the admixture with the corresponding derivatives of (–)-desoxynupharamine. It has been clarified that III is completely coincident with (–)-desoxynupharamine even in the steric configuration.

Therefore, I is considered to be composed of the similar carbon and nitrogen skeleton of desoxynupharamine and the positions of one hydroxyl group and one double bond are to be determined.

When reduced with palladium-carbon consuming one mole of hydrogen, I gave a crude mixture which was fractionally distilled to yield colorless liquid (VI), b.p. $110\sim 120^\circ$ and dihydronuphamine (V) $\text{C}_{15}\text{H}_{25}\text{O}_2\text{N}$, b.p. $155\sim 160^\circ$. The former fraction (VI) was separated into two compounds $\text{C}_{15}\text{H}_{23}\text{ON}$, (VI-A) $[\alpha]_D^{20} -113.17^\circ$ (perchlorate, m.p. 208° ; hydrochloride, m.p. 262°) and $\text{C}_{15}\text{H}_{23}\text{ON}$ (VI-B) $[\alpha]_D^{20} -97.20^\circ$ (perchlorate, m.p. $222.5\sim 226^\circ$) through the crystallization of the perchlorate. Furthermore, V was chlorinated with thionyl chloride followed by heating with potassium hydroxide solution, the expected VI-A and VI-B were derived in the same yield.

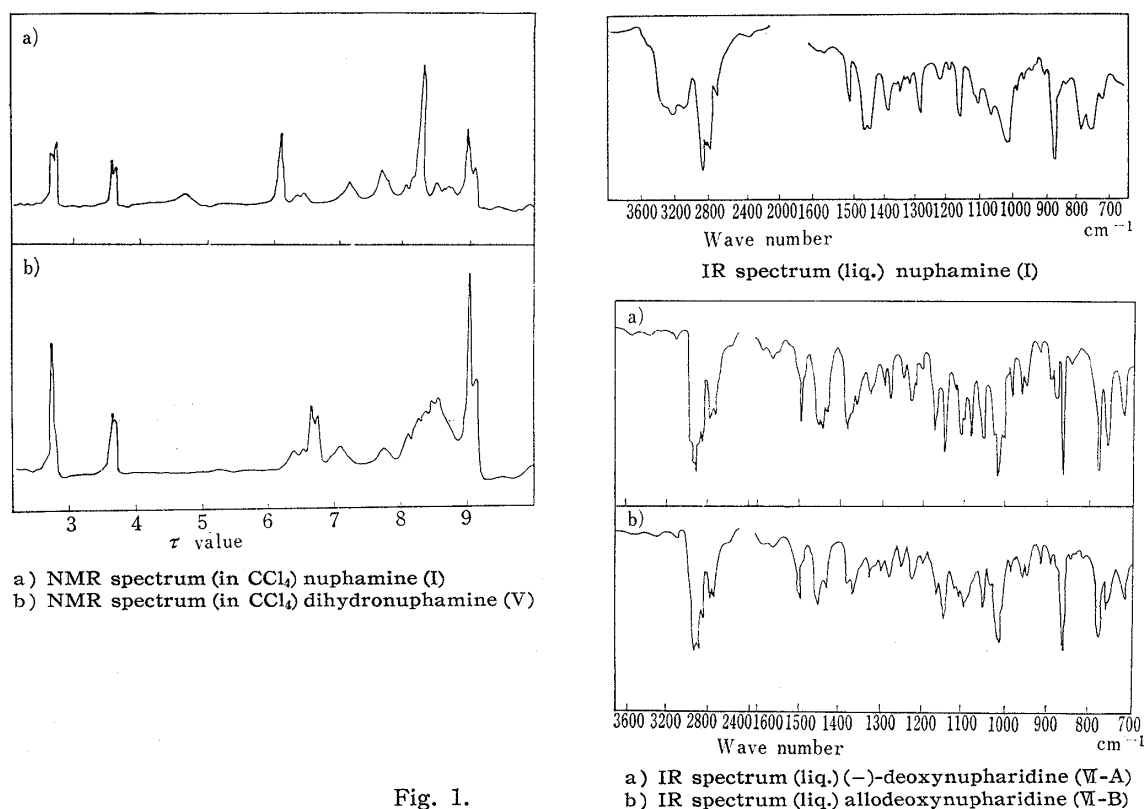


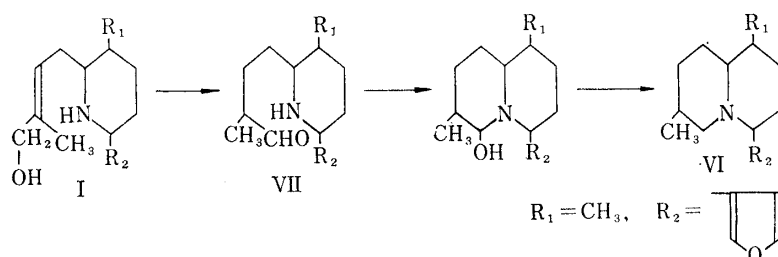
Fig. 1.

4) Y. Arata, *et al.*: *Yakugaku Zasshi*, 82, 782 (1962), 83, 79 (1963).

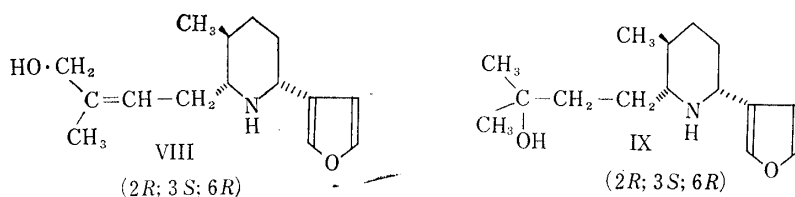
Now, the structure of I was studied by the measurements of nuclear magnetic resonance spectra (Fig. 1) of I and V. The spectrum of I was assigned as follows: $\text{CH}_3\text{-CH}$ (τ 9.09 doublet), $\text{CH}_3\text{-C}=\text{C}$ (τ 8.36 singlet), $\text{HO-CH}_2\text{-C}=\text{C}$ (τ 6.20 singlet), and a vinyl proton (τ 4.74 broad), while that of the dihydro compound (V) showed two methyl groups $\text{CH}_3\text{-CH}$ (6H, τ 9.10 doublet) and $\text{HO-CH}_2\text{-CH}$ (τ 6.70 doublet) but the vinyl proton's signal disappeared, concomitant with the disappearance of the signal of τ 8.36. High field shift of methylene signal bearing a hydroxyl group suggested the presence of allyl alcoholic moiety in the structure of I, and consequently, the structure of nuphamine is represented by the formula (I), considering the facts that I is composed of the skeleton of III.

The degree of specific rotation and infrared spectrum of VI-A (Fig. 1) are completely coincident with that of (-)-deoxynupharidine and both the perchlorate and hydrochloride did not show any melting point depression on the admixture of the corresponding salts of (-)-deoxynupharidine.

Infrared spectrum of VI-B (Fig. 1) indicated the absorption band based on the *trans* quinolizidine (2795, 2770 cm^{-1}) and was different from that of VI-A in its fingerprint region. VI-B is considered to be a compound differing from the configuration of the methyl group at the 7-position. This compound has been named allodeoxynupharidine.



As it was shown in Chart 2, the ring closure reaction of I to VI, may have taken place by the shift of the double bond to give aldehyde (VII) which was further reduced by a ring closure during the catalytic reduction of I.



These facts clarified that I would be represented by formula VIII. Also, the fact that III was derived from both I and IV indicated that the absolute configuration of (-)-nupharamine (IV) is represented by formula IX. The steric structural correlation among (-)-deoxynupharidine (VI-A), (-)-nuphamine (VIII) and (-)-nupharamine (IX) could thus be achieved.

Experimental*4

Nuphamine (I)—Oily alkaloid, extracted³⁾ from the roots (100 kg.) of *Nuphar japonicum* DC. was

*4 IR spectra were measured with a Spectrophotometer, IRDS-402G of Japan Spectroscopic Co., Ltd., UV with a Hitachi Recording Spectrophotometer EPS-2U and NMR with a 3H-60 of Japan Electron Optics Laboratory Co., Ltd., and Varian A-60, 60 Mc (room temperature). Melting point were measured with a Yanagimoto, Micro-melting point Apparatus.

distilled fractionally and the fractionate of b.p.₃ 145~155° was developed over the thin-layer chromatography (TLC), 0.4 mm. of alumina : gypsum (20:3) (solvent : ether, 15°). Besides the reddish brown spot of nupharamine Rf 0.47, colored with the Wagner reagent, a black spot of Rf 0.70 (I) was detected. The crude alkaloid of b.p.₃ 150~160° was fractionally distilled and the latter portion of the distillate (1.5 g.) was recrystallized as a picrolonate.

Nuphamine (I) purified through picrolonate was obtained in colorless viscous liquid, b.p.₃ 150~153°, $[\alpha]_D^{20}$ -60.48° (in CHCl₃). IR cm⁻¹: $\nu_{OH,NH}$ 3620, 3305, ν_{CH} 3150 (furan) (CCl₄ dil. soln.), 3150, 1500, 875 (furan ring) (liq.) (Fig. 1). UV: λ_{max}^{EtOH} m μ (ϵ): 210.5 (7700), NMR: τ 9.09 (3H, doublet CH₃), 8.36 (3H, singlet CH₃), 7.24 (OH, NH disappeared by deuteration), 6.20 (2H, singlet HO-CH₂-C=), 4.74 (1H, broad >C=CH-CH₂-), 3.66 (1H, furan β -position), 2.73 (2H, furan α -position) (CCl₄ soln.) (Fig. 1). *Anal.* Calcd. for C₁₅H₂₃O₂N (I): C, 72.25; H, 9.30; N, 5.62. Found: C, 71.86; H, 9.50; N, 5.25.

Picrolonate: Recrystallization from a mixture of EtOH and ether gave yellow needles, m.p. 126~130°, which contains one mole of crystalline EtOH. *Anal.* Calcd. for C₂₅H₃₁O₇N₅·C₂H₅OH: C, 57.75; H, 6.36. Found: C, 57.95; H, 6.66. Drying at 100° for 7 days, EtOH was splitted to give the amorphous compound, m.p. 159~160°. *Anal.* Calcd. for C₂₅H₃₁O₇N₅: C, 58.47; H, 6.08; N, 13.64. Found: C, 58.63; H, 6.23; N, 13.81.

N-Methylnuphamine Methiodide (II)—0.1 g. of I was dissolved in 6 ml. of 80% MeOH, and it was warmed for 3 hr. with 0.1 g. of Na₂CO₃ and 0.3 ml. of CH₃I. After the reaction, solvent was removed and the residue was treated with CHCl₃. The residue from CHCl₃ was recrystallized from a mixture of AcOEt and Me₂CO to give colorless needles, m.p. 164°. *Anal.* Calcd. for C₁₇H₂₈O₂NI (II): C, 50.32; H, 6.91. Found: C, 50.39; H, 7.14.

Dihydrodeoxynuphamine(III) ((-)-Desoxynupharamine)—0.5 g. of I was dissolved in 20 ml. of absolute ether, and to this a solution of 0.5 ml. of SOCl₂ and 20 ml. of absolute ether was added dropwise. After it was kept for 1.5 hr. at room temperature, K₂CO₃ solution was added to make alkaline and extracted with ether. From the ether soluble extract, colorless liquid of b.p.₃ 150~160° (bath temperature) was obtained. Yield, 0.43 g. Beilstein's test was positive (IR cm⁻¹: ν_{C-O} 680).

0.428 g. of chloro compound (chlorodeoxynuphamine) was dissolved in EtOH and it was reduced catalytically with Pd-C, absorbing about 2 moles of H₂. After the separation of Pd-C, the filtrate was evaporated to dryness resulting a HCl salt as colorless needles. The base (III) purified through HCl salt afforded colorless liquid b.p.₃ 130~137° (bath temperature). $[\alpha]_D^{18}$ -63.56°. IR spectrum was coincident with that of (-)-desoxynupharamine. IR cm⁻¹: 1500, 1160, 870, 722 (furan) (CCl₄ soln.). *Anal.* Calcd. for C₁₅H₂₅ON (III): C, 76.54; H, 10.71; N, 5.95. Found: C, 76.13; H, 11.12; N, 6.01.

Hydrochloride: Recrystallized from acetone to give colorless needles, m.p. 233°. No melting point depression was observed on the admixture of HCl salt of (-)-desoxynupharamine derived from (-)-nupharamine (IV). *Anal.* Calcd. for C₁₅H₂₆ONCl: C, 66.29; H, 9.57. Found: C, 66.02; H, 9.75.

Perchlorate: Recrystallized from water to give colorless needles, m.p. 128°. No melting point depression was observed on the admixture with (-)-desoxynupharamine perchlorate.

Picrolonate: Recrystallized from a mixture of EtOH and ether to give yellow needles, m.p. 176°. No melting point depression was observed by the admixture of (-)-desoxynupharamine picrolonate.

Reduction of Nuphamine (Preparation of Dihydronuphamine (V), (-)-Deoxynupharidine (VI-A) and Allodeoxynupharidine (VI-B))—Catalytic reduction of 3.0 g. of nuphamine (I) in EtOH was carried out with Pd-C. The reaction was stopped when 2900 ml. of H₂ was absorbed and then the catalyst was filtered. Evaporation of the filtrate gave the residue which was fractionally distilled to yield colorless liquid (VI) (yield 0.8 g.), b.p.₃ 110~120° (bath temperature) and colorless viscous liquid (V), b.p.₃ 155~160° (yield 1.4 g.). The latter fraction was solidified when kept standing.

The base (VI) was heated with acetic anhydride in the presence of NaOAc once, in order to separate the mixed secondary amine and then the basic portion (VII) was obtained in colorless liquid, b.p.₃ 113~115°, yielding 0.6 g. Perchlorate was treated with AcOEt to separate into two portions, AcOEt hardly soluble and soluble portions.

Dihydronuphamine (V): Colorless needles, m.p. 42.5~43°, IR cm⁻¹: 3610 (free OH), 3330 br. (bond OH), 1500, 1160, 1030, 870 (furan) (CCl₄ soln.). NMR: τ 9.10 (6H, doublet, CH₃-CH<), 7.12 (OH, NH singlet), 6.70 (2H, doublet, HO-CH₂-CH-), 3.66 (1H, furan β -position), 2.73 (2H, furan α -position). (CCl₄ soln.) (Fig. 1). *Anal.* Calcd. for C₁₅H₂₅O₂N (V): C, 71.67; H, 10.03; N, 5.57. Found: C, 71.59; H, 9.94; N, 5.63.

(-)-Deoxynupharidine (VI-A): AcOEt hardly soluble. Recrystallization of perchlorate from EtOH gave colorless needles, m.p. 208°. No melting point depression was observed by the admixture with that of (-)-deoxynupharidine. *Anal.* Calcd. for C₁₅H₂₄O₅NCl: C, 53.91; H, 7.19; N, 4.19. Found: C, 53.84; H, 7.26; N, 4.38. The base purified through perchlorate is colorless liquid, b.p.₃ 118~120° (bath temperature), $[\alpha]_D^{20}$ -113.17° and the IR spectrum was completely in accordance with that of (-)-deoxynupharidine. IR cm⁻¹: 2795, 2770 (*trans* quinolizidine),*⁵ 3130, 1505, 1157, 1030, 873, 785 (furan) (liq.) (Fig. 1). The

*⁵ These values are a little different from the ones previously reported and obtained by the measurement with a grating spectrophotometer.

hydrochloride was recrystallized from EtOH, gave colorless plates, m.p. 262°. No melting point depression was observed by the admixture with that of (–)-deoxynupharidine.

Allodeoxynupharidine (VI-B): AcOEt soluble perchlorate was recrystallized from a mixture of AcOEt and ether to give colorless prisms, m.p. 225.5~226°. *Anal.* Calcd. for $C_{15}H_{24}O_5NCl$: C, 53.91; H, 7.19; N, 4.19. Found: C, 53.63; H, 7.34; N, 4.05. The base purified through the perchlorate gave colorless liquid, b.p._{3,6} 120~125° (bath temperature), $[\alpha]_D^{25}$ –97.20° and its IR spectra indicated a clear difference from VI-A in the finger print region. IR cm^{-1} : 2795, 2770 (*trans* quinolizidine), 3130, 1505, 1157, 1030, 873, 785 (furan).

Ring Closure of Dihydronupharine (V) (Preparation of (–)-Deoxynupharidine (VI-A) and Allodeoxynupharidine (VI-B))—1.4 g. of dihydronupharine (V) was dissolved in 30 ml. of absolute ether, to this a solution of 1 ml. of thionyl chloride and 15 ml. of absolute ether was added dropwise, kept for 2 hr. at room temperature. Water and K_2CO_3 were added to separate ether layer. Ether residue showed positive Beilstein's test. It was heated for 1.5 hr. in 10 ml. of MeOH, 2 ml. of H_2O and 0.8 g. of KOH. After the evaporation to dryness *in vacuo*, it was extracted with ether which portion gave 0.5 g. of colorless liquid, b.p.₃ 115~120° (bath temperature). It was converted to perchlorate which was separated into two, AcOEt hardly soluble and soluble portions. The former VI-A was recrystallized from EtOH, m.p. 208°, colorless needles. Yield: 0.2 g. No melting point depression was observed by the admixture with (–)-deoxynupharidine perchlorate. The latter (VI-B) was recrystallized from a mixture of AcOEt and ether to give colorless plate, m.p. 225.5~226°. Yield: 0.2 g. No melting point depression was observed by the admixture of allodeoxynupharidine perchlorate.

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Summary

A new alkaloid (I), $C_{15}H_{23}O_2N$ has been isolated from *Nuphar japonicum* DC. and named nupharine. The compound (III) obtained by the catalytic reduction after the chlorination of I with thionyl chloride is completely in accordance with (–)-desoxy-nupharine, derived from nupharine (X).

By the catalytic reduction of I with palladium-carbon, dihydronupharine (V), (–)-deoxynupharidine (VI-A) and allodeoxynupharidine (VI-B) were prepared. VI-A and VI-B were also prepared from V. Judging from these reactions and the nuclear magnetic resonance spectra, the structure of I has been determined to be presented by the formula VIII. The steric chemical correlations among the alkaloids, (–)-desoxy-nupharidine (VI-A), (–)-nupharine (VIII) and (–)-nupharine (X) isolated from *Nuphar japonicum* have been also achieved.

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