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117. Yoshio Arata : Constituents of Rhizoma Nupharis. XXI.*1 Structure of Dehydro-deoxynupharidine.

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The structure of a new alkaloid, dehydro-deoxynupharidine (I), isolated recently from the roots of *Nuphar japonicum* DC. was reported[1] briefly and its detail will be described in this report.

The base which was distilled between 110~120°/3 mm. Hg from the methanol extract of the roots of *Nuphar japonicum* DC. is composed mainly of deoxynupharidine.[2~5] The distillate is much unstable than (−)-deoxynupharidine (II), b.p_3 112~115°, $[\alpha]_D$ −112.5°, which was purified through the HCl salt, and it changed its color into brown when kept standing overnight. This fact suggested that the crude base might be contaminated with easily oxidizable bases and, therefore, an investigation of new bases was carried out.

The crude deoxynupharidine was once converted into its HCl salt, and the HCl salt of II was attempted to be separated as much as possible by recrystallization. The free base from the mothor liquor was afforded as a crystalline perchlorate, $C_{15}H_{21}ON \cdot HClO_4$, m.p. 159~161°, $[\alpha]_D^{29}$ +130.1°($CHCl_3$) and hydrobromide, $C_{15}H_{21}ON \cdot HBr$, m.p. 199.5~201°. The infrared spectrum of the perchlorate in chloroform showed at 877, 1510, and 3128 cm^{-1} which may be assigned to furan, and also at 1640 cm^{-1} which may be considered the iminium conjugated with furan ring. Similarly, the nuclear magnetic resonance spectrum indicated 3 signals (δ 8.09, 7.56, and 6.82) which corresponded to one proton each. These data assigned clearly to the furan ring.

The free base (I) derived from the salt, was so unstalbe colorless liquid, b.p_3 125° (bath temperature) that it changed its color into brown during the distillation. The new base, $C_{15}H_{21}ON$ (I) which gave perchlorate of m.p. 159~161°, had been named dehydro-deoxynupharidine.

The perchlorate of I was reduced by sodium borohydride to a colorless oily base, $C_{15}H_{23}ON$ (III), b.p_3 125° (bath temperature), $[\alpha]_D$ −114.1°($CHCl_3$), IR cm^{-1} : 2764, 2792 (*trans*-quinolizidine), 874, 1032, 1505, 3140 (furan), which gave picrate, m.p. 154.5~155.5° and perchlorate, m.p. 203~204.5°. No difference of the infrared spectra and no depression of melting point by admixture indicated that III was in accordance with II. Therefore, I is considered to be composed of having similar C-N-O skeleton to deoxynupharidine (II), though the position of one double bond being remained undecided.

The facts that the perchlorate of I prepared the dihydro compound (II) by sodium borohydride reduction, and that it indicated a negative reaction by the active hydrogen measurement of Zerewitinoff, suggested the enamine type formation of I, and also the iminium-type formation of the salt of I. On the other hand, the enamine had been already reduced also to dihydro derivative[5] by the reagent. The reduction of I by sodium borohydride was attempted and the production of the expected II was identified.

*1 Part XX : Rep. Pharm. Kanazawa, Japan, **12**, 39 (1962).
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1) Y. Arata : This Bulletin, **12**, 1394 (1964).
2) Y. Arata, *et al.* : Yakugaku Zasshi, **66 B**, 139 (1946).
3) Y. Arata : *Ibid.*, **76**, 1446 (1956); Y. Arata, *et al.* : *Ibid.*, **77**, 236 (1957).
4) Y. Arata, *et al.* : *Ibid.*, **80**, 855 (1960).
5) Y. Arata, *et al.* : This Bulletin, **10**, 676 (1962).

However, as to the enamine compounds which are premised to produce deoxynupharidine (II), the formation of the compounds (IV), (V) and (VI) might be possible to consider, so does the presence of IV', V', and VI' from their iminium salts, respectively.

On the other hand, the infrared spectrum of perchlorate of I showed at 1640 cm^{-1} which was supposed to be an iminium salt conjugated to aromatic ring, therefore, the structure of I and its salt could be presented as the formula (IV) and (IV').

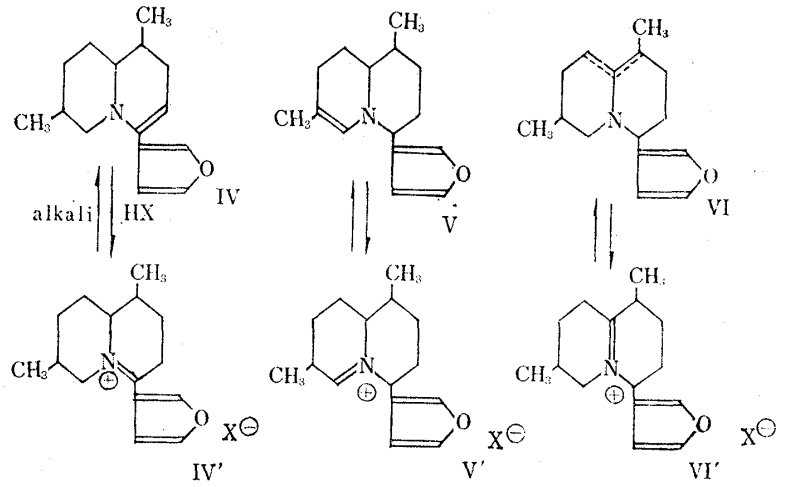

Chart 1.

As it will be shown in Chart 2, it has been confirmed further by the syntheses.

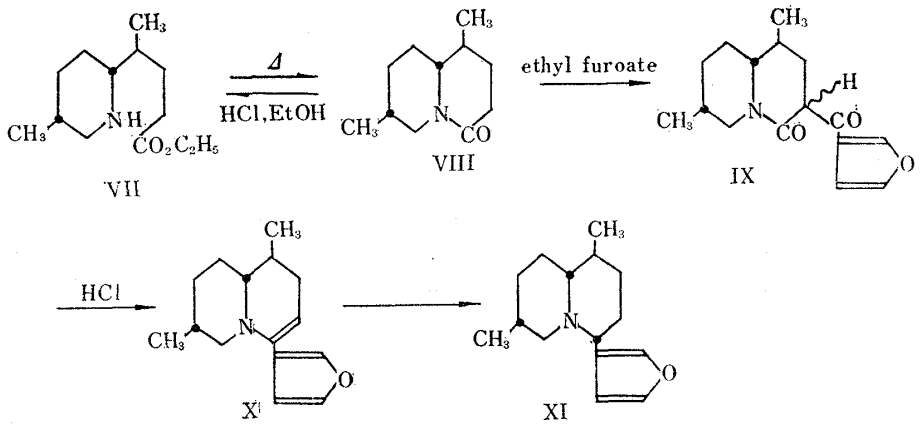

Chart 2.

The (±)-dimethylo-4-quinolizidinone[5] synthesized previously, is composed of a mixture of four kinds of racemate,*3 based on the asymmetric carbon atoms, and the picrolonate of ethyl 4-(5-methyl-2-piperidyl)valerate derived above from the mixture, gave two kinds of substances[4] of m.p. 163~164° and 156~158° by recrystallization. The synthesis of (±)-deoxynupharidine from the starting material of picrolonate of VII had already been reported.[4]

The derived lactam (VIII) from VII by heating produced IX, being colored in dark green with ferric chloride solution, by the condensation with ethyl 3-furoate in the presence of sodium hydride. IX was hydrolyzed in dil. hydrochloric acid to give enamine (X), b.p_2 120~130° (bath temperature) which formed perchlorate, m.p. 145.5~147°.

*3 Bohlmann, *et al.* (Chem. Ber., **94**, 3151 (1961)) separated 4 kinds of substances by chromatography.

The infrared spectra of the perchlorates synthesized and the one obtained from the natural product (I) in chloroform did not show any difference (Fig. 1).

The reduction of perchlorate of X by sodium borohydride prepared (±)-deoxynupharidine (XI), IR cm^{-1}: 872, 1028, 1500, 3100 (furan), 2770, 2795 (*trans*-quinolizidine) and its perchlorate, m.p. 201~203° did not show any melting point depression by the admixture with that of (±)-deoxynupharidine,[5] previously synthesized. The infrared spectra of both XI and II in carbon tetrachloride were completely in coincident.

As the absolute configuration[6] of (−)-deoxynupharidine (II) had been already defined, the formula (I) was presented as the structure and the absolute configuration of dehydro-deoxynupharidine (Chart 3).

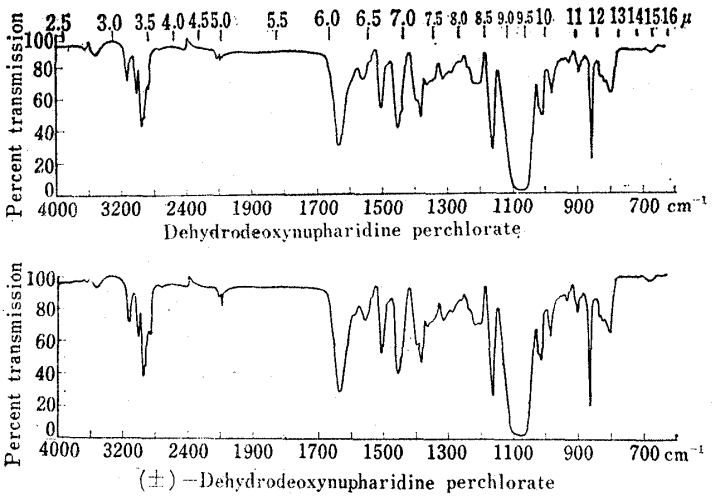

Fig. 1.

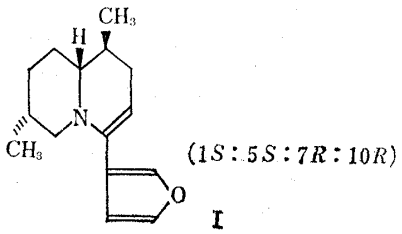

Chart 3.

Experimental[4]*

Dehydro-deoxynupharidine (I)——According to the methods by Arima, *et al.*[7] and Arata, *et al.*,[8] total alkaloids obtained from the MeOH extracts from roots (50 kg.) of dried *Nuphar japonicum* DC., were converted to the picrate, which was recrystallized from EtOH. The nupharidine picrate deposited was separated from the mother liquor, which gave the free base by the distillation under the reduced pressure.

The distillate of b.p. 110~125°/3 mm. Hg was collected, converted once into HCl salt and recrystallized from water. The deoxynupharidine (II) HCl salt deposited was separated and the mother liquor was condensed to remove the HCl of II, deposited again from the mother liquor. It was made alkaline by Na_2CO_3 and the oily substance deposited was extracted with ether. After being evaporated, the distillate, b.p. 110~120°/3 mm. Hg, was collected from the residue, converted once to HCl salt, and treated similary with water to the procedure mentioned above in order to remove the HCl salt of II as much as possible. The mother liquor was extracted with ether in Na_2CO_3 solution. The ether layer was washed with water and desiccated, and then the solvent was evaporated. From the residue, unstable brown distillate 6 g. of b.p. 110~120°/3 mm. Hg was obtained. The perchlorate of this distillate was evaporated *in vacuo* and the insoluble precipitate obtained from the perchlorate, treated with a small amount of AcOEt, was recrystallized from MeOH, to give colorless needles, or prisms, m.p. 159~161°, $[\alpha]_D^{29}$ +130.1° (Sub. 0.0907 g. in $CHCl_3$, 2 ml., α +5.90°). IR cm^{-1}: 877, 1510, 3128 (furan), 1640 (conjugate iminium) (in $CHCl_3$). No CH_4 generation was observed by the active hydrogen measurement according to Zerewitinoff. *Anal.* Calcd. for $C_{15}H_{22}O_5NCl$: C, 54.28; H, 6.69; N, 4.22. Found: C, 54.19; H, 6.77; N, 4.41.

*4 All melting points were measured with a micro-melting point apparatus, the Yanagimoto Mfc. Co., and the IR spectra were measured with a Infrared Spectrophotometer S and DS-402G, the Japan Spectroscopic Co., Ltd.

6) Y. Arata, *et al.*: Yakugaku Zasshi, **82**, 326 (1962); Rep. Pharm. Kanazawa, Japan, **12**, 39 (1962); M. Kotake, *et al.*: Bull. Chem. Soc. Japan, **35**, 1335 (1962); F. Bohlmann, *et al.*: Chem. Ber., **94**, 3151 (1961).
7) Z. Arima, *et al.*: Nippon Kagaku Zasshi, **52**, 815 (1931).
8) Y. Arata, *et al.*: Yakugaku Zasshi, **66 B**, 138 (1946); **77**, 792 (1957).

Dehydro-deoxynupharidine (I) derived from purified perchlorate, is an unstable liquid, distilled at $b.p_3$ 125° (bath temperature) and turned into brown during the distillation. Hydrobromide was recrystallized from a mixture of EtOH and AcOEt (1:4) to give colorless prism, m.p. 199.5~201°. *Anal.* Calcd. for $C_{15}H_{22}ONBr$: C, 57.67; H, 7.10; N, 4.49. Found: C, 57.69; H, 7.07; N, 4.80.

Preparation of (−)-Deoxynupharidine (II)——1) Reduction of dehydro-deoxynupharidine perchlorate by sodium borohydride: 0.5 grams of dehydro-deoxynupharidine perchlorate was dissolved in 10 ml. of MeOH, to this was added 0.35 g. of $NaBH_4$ under cooling in a small portion. In this case, severe foaming was observed exothermically. After the reaction, the solution was kept standing overnight and the solvent was evaporated *in vacuo*. The residue was made alkaline with NaOH and the precipitates deposited were extracted with ether, which was washed with water, desiccated and then evaporated. The residue (0.4 g.) is a colorless, stable liquid, $b.p_3$ 125° (bath temperature). The base (III) purifiedt hrough the perchlorate, showed $[\alpha]_D^{27}$ −114.1° (Sub. 0.217 g. in $CHCl_3$ 2 ml., α −12.38°), IR cm^{-1}: 874, 1032, 1505, 3140 (furan), 2764, 2792 (*trans*-quinolizidine) and its IR spectrum was completely in accordance with that of (−)-deoxynupharidine (II), obtained from the roots of *Nuphar japonicum* DC.

Perchlorate: Recrystallized from MeOH to give colorless long plate crystals, m.p. 203~204.5°. No depression of melting point was observed by the admixture of this perchlorate with that of II. *Anal.* Calcd. for $C_{15}H_{24}O_5NCl$: C, 53.49; H, 7.18; N, 4.16. Found: C, 53.58; H, 7.33; N, 4.11. IR cm^{-1}: 3075, 1505, 875 (furan) (in Nujol).

Picrate: Recrystallized from EtOH to give yellow needles, m.p. 154.5~155.5°. No depression of melting point was observed by the admixture of picrates of both this compound and II. *Anal.* Calcd. for $C_{21}H_{26}O_8N_4$: C, 54.52; H, 5.67; N, 12.12. Found: C, 54.68; H, 5.67; N, 12.27.

2) Reduction of dehydro-deoxynupharidine (I) by sodium borohydride: To 5 ml. of MeOH solution of dehydro-deoxynupharidine (I) (0.05 g.), was added 0.05 g. of $NaBH_4$ and it was kept overnight. Following to the similar treatment to the one mentioned above, the reduced compound was converted to perchlorate which was recrystallized from MeOH, giving colorless long plate crystals, m.p. 202~204.5°. IR cm^{-1}: 3075, 1505, 875 (furan) (in Nujol). *Anal.* Calcd. for $C_{15}H_{24}O_5NCl$: C, 53.49; H, 7.18; N, 4.16. Found: C, 53.61; H, 7.30; N, 4.14. No depression of melting point was observed by the admixture of the perchlorate of this compound with that of II. The IR spectra of both compounds in Nujol were completely in accordance.

Ethyl 4-(5-methyl-2-piperidyl)valerate (VII)——(±)-1,7-Dimethyl-4-quinolizidinone obtained previously by Arata, *et al.*[4] was considered to be a mixture of four recemates and 7.2 g. of the material was heated for 10 hr. in 50 ml. of 15% HCl, followed by an evaporation under the reduced pressure. To the residue, 50 ml. of EtOH was added, warmed on a water-bath for one hour at 60~65° and it was kept for two all nights. To this an excess of silver carbonate was added and separated after well-mixing. To the fitrate, hydrogen sulfide was introduced to separate the Ag salt as a deposit. To the filtrate, a calculated amount of picrolonic acid was added and dissolved by warming. Then, it was evaporated to dryness *in vacuo*. A repeated recrystallization from 60% EtOH gave yellow cube (1.7 g.), m.p. 156~158° (picrolonate of VII). The mother liquor was also evaporated and the residue was recrystallized from 60% EtOH to give yellow powder crystals in upper layer and yellowish brown prisms at the bottom. After the separation of both crystals, a recrystallization from 80% EtOH gave yellow powder crystal (0.3 g.), m.p. 163~164° from the former and cubic crystals, 0.6 g., m.p. 156~158° from the latter. The latter showed no depression of melting point by the admixture of picrolonate of VII, however, the former substance showed the depression of melting point. *Anal.* Calcd. for $C_{23}H_{33}O_7N_5$ (m.p. 156~158°): C, 56.20; H, 6.77; N, 14.25. Found: C, 56.46; H, 7.05; N, 14.10. *Anal.* Calcd. for $C_{23}H_{33}O_7N_5$ (m.p. 163~164°) C, 56.20; H, 6.77; N, 14.25. Found: C, 56.53; H, 6.76; N, 14.26.

3-(3-Furoyl)-1,7-dimethyl-4-quinolizidinone (IX)——The compound (VII), made free from the picrolonate of VII mentioned above, gave colorless liquid of 1,7-dimethyl-4-quinolizidinone (VIII), $b.p_2$ 130~135° (bath temperature), by the distillation under the reduced pressure. 0.57 g. of VIII was dissolved in 5 ml. of benzene, to this was added 0.135 g. of 56% NaH in oil, then 0.5 g. of ethyl 3-furoate and they were heated for 7 hr. at 80~90° with stirring. The reaction solution was acidified with HCl and extracted with ether, which was washed with water and desiccated, followed by the evaporation of the solvent. The residue was distilled to give 0.6 g. of milky viscous liquid, $b.p_3$ 163~215° (bath temperature), which contained the oil in NaH used. The EtOH solution of IX showed a dark green coloration by ferric chloride solution.

(±)-Dehydro-deoxynupharidine (X)——A mixture of the furoyl compound of IX (0.55 g.), 10% HCl (7.5 ml.) and AcOH (2.5 ml.) was heated for 10 hr. The reaction solution was evaporated to dryness *in vacuo* and the residue was shaken with ether and water. The water layer was made alkaline with K_2CO_3 and shaken again with ether, which was washed with water and desiccated. The residue evaporated from the ether extract afforded 0.3 g. of brownish liquid, $b.p_2$ 120~130° (bath temperature). It turned brown by rapid oxidation when kept standing.

Perchlorate: Recrystallized from a mixture of MeOH and AcOEt (1:6) to give colorless scaly crystals, m.p. 145.5~147°. *Anal.* Calcd. for $C_{15}H_{22}O_5NCl$: C, 54.28; H, 6.69; N, 4.22. Found: C, 54.45; H, 6.78: N, 4.19. IR cm^{-1}: 877, 1510, 3128 (furan), 1640 (conjugate iminium) (in $CHCl_3$). The IR spectra of the

perchlorate obtained here and that of dehydro-deoxynupharidine (I) in $CHCl_3$ were completely coincident (Fig. 1).

(±)-Deoxynupharidine (XI)——0.1 gram of perchlorate of X was dissolved in 2 ml. of MeOH, and to this was added 0.05 g. of $NaBH_4$. After it was kept standing overnight, the solvent was removed under the reduced pressure. The residue was made alkaline with NaOH and the deposits were extracted with ether, which was washed with water, and then desiccated. The residue evaporated from ether gave colorless liquid (XI), $b.p_3$ 120~130° (bath temperature). The IR absorption spectra of this compound and II were completely coincident in CCl_4. IR cm^{-1}: 872, 1028, 1500, 3100 (furan), 2770, 2795 (*trans*-quinolizidine) (in CCl_4).

Perchlorate: Recrystallized from 50% MeOH to give white needles, m.p. 201~203°. *Anal.* Calcd. for $C_{15}H_{24}O_5NCl$: C, 53.96; H, 7.25; N, 4.20. Found: C, 54.10; H, 7.15; N, 4.01.

No depression of melting point was observed by the admixture of this compound with (±)-deoxynupharidine,[5] synthesized previously.

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Summary

A new base, dehydro-deoxynupharidine (I), $C_{15}H_{21}ON$, was isolated from the roots of *Nuphar japonicum* DC. The infrared spectra of perchlorate of I showed at 1640 cm^{-1} which correspond to the iminium conjugated to aromatic ring. It was converted to (−)-deoxynupharidine (III) by sodium borohydride reduction. Therefore, the structure and the absolute configuration of I were represented by formula I, which was confirmed by the syntheses.

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