Aporphine Alkaloids from *Parabenzozin praecox* (SIEB. et ZUCC.) NAKAI

MUTSUO KOZUKA,* AKIRA INADA,  TAKAO KONOSHIMA, and TOKUNOSUKE SAWADA

*Kyoto Pharmaceutical University, Nakauchi-cho, Misasagi, Yamashina-ku, Kyoto 607, Japan*

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A new aporphine alkaloid, praecoxine (1), was isolated from the root of *Parabenzozin praecox* (SIEB. et ZUCC.) NAKAI. The alkaloid was shown to be identical with N-methylhermine, (S)-(+)-1,2,11-trimethoxy-10-hydroxyaporphine (I), on the basis of chemical transformations and spectral evidence. The occurrence of nandigerine (10) in the root and the stem wood was also proved.

**Keywords**— *Parabenzozin praecox*; Lauraceae; aporphine alkaloid; praecoxine; nandigerine; \(^1^H\)-NMR; chemical transformation; (S)-(+)-1,2,11-trimethoxy-10-hydroxyaporphine

*Parabenzozin praecox* (SIEB. et ZUCC.) NAKAI (Lauraceae) is a tree occurring in Kyushu, Shikoku, and Honshu. The insect antifeedant substances, and the essential oil, especially mono- and sesquiterpenoids, from this plant have been investigated. However, no study on alkaloidal constituents of the plant seems to have been published. As a part of our continuing work on alkaloids of Lauraceous plants, we report here the isolation and characterization of a new aporphine alkaloid, that has been named praecoxine\(^1\) (1), from the root of *P. praecox*.

Praecoxine (1) was isolated as a colorless crystalline compound, which decomposes at 230 °C, \([\alpha]_D^\infty +270^\circ\) (methanol), infrared (IR) spectrum (CHCl\(_3\)) cm\(^{-1}\): 3500 (OH). The molecular formula C\(_{26}\)H\(_{23}\)NO\(_4\) was derived by high resolution mass spectroscopy. The ultraviolet (UV) spectrum of the base was clearly of a 1,2,10,11-tetrasubstituted aporphine\(^6\) and exhibited a bathochromic shift in alkaline ethanol solution. Its \(^1^H\)-nuclear magnetic resonance (\(^1^H\)-NMR) spectrum showed an N-methyl group, three methoxyl groups, a hydroxyl group, and three aromatic protons. Two of the three aromatic protons appeared as overlapped singlets at \(\delta 6.87\) [C(8)- and C(9)-H].\(^7\)

Methylation of praecoxine (1) with diazomethane afforded amorphous *O*-methylpraecoxine (7). Its \(^1^H\)-NMR spectrum showed an N-methyl group, four methoxyl groups, and three aromatic protons.

Praecoxine (1) reacts with methyl iodide and KOH in methanol to give *O*-methylpraecoxine methiodide (8), mp 249 °C (dec.), \([\alpha]_D^\infty +147^\circ\) (ethanol), which was identical with authentic *O*,*O*-dimethylcoryctuberine methiodide\(^8\) (8) on the basis of mixed melting point determination and IR spectral comparison.

The position of the phenolic hydroxyl group in praecoxine (1) was proven in the following manner. Praecoxine (1) was heated in deuterium oxide containing sodium hydroxide to give a monodeuterio derivative (M\(^+\), m/z 342). The \(^1^H\)-NMR spectrum of the monodeuterated base (3) differed from that of the non-deuterated compound (1) only in the intensity of the \(\delta 6.87\) aromatic proton signal, which was found to be decreased to one-half by the deuteration, while the \(\delta 6.68\) signal due to 1H at C-3 was unchanged. Since the formula 4 was excluded,\(^9\) the site of deuteration exchange must be C-9, and the position of the phenolic hydroxyl in praecoxine (1) at C-10 was established.
On the basis of the foregoing and other results, it was concluded that praecoxine is (S)-(-)-1,2,11-trimethoxy-10-hydroxyaporphine (I).

![Chemical structures](image)

1: \( R_1 = R_2 = R_3 = CH_3, \quad R_4 = H \)
2: racemic form of 1
3: 1 [C(9) = D instead of C(9) = H]
4: \( R_1 = H, \quad R_2 = R_3 = R_4 = CH_3 \)
5: \( R_1 = R_3 = R_4 = CH_3, \quad R_2 = H \)
6: \( R_1 = R_2 = R_4 = CH_3, \quad R_3 = H \)
7: \( R_1 = R_2 = R_3 = R_4 = CH_3 \)

**Chart 1**

Syntheses of the racemic form (2) have already been reported.\(^{10,11b}\) Further, a new aporphine alkaloid which has the N-norpraecoxine structure, named hernagine (9), was isolated from *Hernandia nymphaeifolia*\(^{11a}\) and *H. cordigera*.\(^{11b}\) N-Methylhernagine was obtained from hernagine (9), and the structure 1 was confirmed by the spectral data.\(^{11a}\) The IR and \(^1\)H-NMR spectra of the racemic base \(^{10b}\) (2), and N-methylhernagine \(^{11a}\) (1) were compared with those of praecoxine (1), and they were found to be identical.

The isolation and characterization of praecoxine (1) thus provide further support for the structures of hernagine (9) and its N-methyl derivative (1). This paper is the first report of isolation of N-methylhernagine (= praecoxine) from a natural source.

Further, a major alkaloidal constituent of the root and the stem wood of *P. praecox* was found to be nandinigerine (10), which was identified by direct comparison with an authentic specimen.\(^{12b}\)

### Experimental

All melting points were taken on a Yanagimoto micro melting point apparatus and are uncorrected. UV spectra were obtained on a Hitachi EPS-2 spectrophotometer and IR spectra were recorded on a Shimadzu IR-27C spectrophotometer. \(^1\)H-NMR spectra were recorded on a Varian A-60A spectrometer with tetramethylsilane as an internal standard. Mass spectra (MS) were taken with Hitachi RMU-7 and RMU-6E machines. Optical rotations were measured on a Rex photoelectric polarimeter. Column chromatography was carried out with silica gel (Wakogel C-200, 100–200 mesh) or with alumina (Aluminiumoxid nach Brockmann, Merck). Thin-layer chromatography (TLC) was performed on Aluminiumoxid G (Merck) in CHCl\(_3\)-acetone (1:1, v/v) unless otherwise stated, and the developed spots were detected with iodine vapor and Dragendorff’s reagent.

**Material and Extraction**—Root and stem wood of *Parabenzoin praecox* (Sieb. et Zucc.) Nakai were collected in Sakyo-ku, Kyoto, and in Hyogo Prefecture in November 1968. Air-dried and cut materials (root: 28.3 kg; stem wood: 21.0 kg) were extracted with hot MeOH and the MeOH was evaporated off under reduced pressure to leave a residue (root, ca. 600 g; stem wood, ca. 530 g). The residue was dissolved in 3% aqueous citric acid, and the solution was filtered, and washed with Et\(_2\)O to remove the non-basic substances. The acidic solution was made alkaline with conc. NH\(_4\)OH and extracted with Et\(_2\)O. The Et\(_2\)O solution was shaken with 4% NaOH solution, and the NaOH solution was made ammoniacal with NH\(_4\)Cl. The NH\(_4\)OH-ammoniacal solution was further extracted with Et\(_2\)O, and the Et\(_2\)O solution was washed with H\(_2\)O, then dried over anhyd. MgSO\(_4\). Evaporation of the Et\(_2\)O gave the crude phenolic alkaloid mixture (root, 6.5 g; stem wood, 6.1 g). The Et\(_2\)O solution freed of phenolic alkaloids was washed with H\(_2\)O, dried over anhyd. K\(_2\)CO\(_3\), and evaporated to leave a small amount of crude non-phenolic
bases (root, 0.562 g; stem wood, 0.310 g).

Isolation of Paeonoxine (1) and Nandigerine (10) from the Root — The crude phenolic alkaloid mixture from the root (6.5 g) was dissolved in a small amount of CHCl₃, and a saturated CHCl₃ solution of picrolonic acid was added to yield crystalline nandigerine (10) picrolonate (0.613 g). The CHCl₃ was removed from the mother liquor of this picrolonate. The residue was dissolved in acetonitrile, and poured into 3% aqueous HCl solution. The resulting suspension was washed with Et₂O, then made alkaline with conc. NH₄OH and extracted with Et₂O. The Et₂O was evaporated off and the residue was chromatographed on silica gel. The fraction eluted with aceton–CHCl₃ (5:95, v/v) gave crude paeonoxine (1) (110.8 mg), while the fraction eluted with aceton–CHCl₃ (1:9) contained crude nandigerine (10) (1.4 g).

Paeonoxine (1) — Colorless crystals (from acetonitrile), decomposes at 230 °C, [α]D²⁰ +270° (c = 0.1, MeOH). IR (CHCl₃) cm⁻¹: 3500 (OH). UV λmax (EtOH) nm (ε): 271 (12380), 302 (5000). ¹H-NMR (CDCl₃) δ ppm: 2.55 (3H, s, NCH₃), 3.50 (3H, s, OCH₃ at C-1), 3.54 (3H, s, OCH₃ at C-11), 3.87 (3H, s, OCH₃ at C-2), 4.87—5.60 (1H, br. exchanged by D₂O, OH), 6.68 (1H, s, C-3 aromatic H), 6.87 (2H, overlapped s, C-8 and C-9 aromatic H). MS m/z: 341 (M⁺), 310 (base peak). C₁₀H₁₃NO₄ (M⁺). Calculated m/z: 341.1627; Found m/z: 341.1628.

O-Methylpaeonoxine (7) — An excess of Et₂O solution of diazomethane was added to a solution of paeonoxine (1) (20 mg) in MeOH, and the mixture was allowed to stand for 2 d at room temperature. The solvent was evaporated off and the residue was dissolved in 3% aqueous citric acid. The acidic solution was washed with Et₂O, made alkaline with NH₄OH and extracted with Et₂O. The combined Et₂O extract was washed successively with 5% NaOH solution and with H₂O, then dried over anhyd. K₂CO₃ and the solvent was evaporated off. The oily product (21 mg) was chromatographed on alumina with benzene to give 7. TLC: 1 spot. ¹H-NMR (CDCl₃) δ ppm: 2.53 (3H, s, NCH₃), 3.64, 3.72, 3.87, and 3.88 (3H × 4, each s, four OCH₃). 6.68 (1H, s, aromatic H), 6.81 and 6.96 (each 1H, a pair of d, J = 9 Hz, aromatic H).

O-Methylpaeonoxine Methiodide (O,O-Dimethyloctopamine Methiodide) (8) — A manesolanic solution of KOH (0.04 g) and an excess (0.5 ml) of Me₂ was added to a solution of paeonoxine (1) (38.7 mg) in MeOH. After this mixture had been heated under reflux for 2h, most of the portions of the manesolanic KOH (0.04 g) and Me₂ (0.5 ml) were added, and the heating was continued for 2.5 h. The process was repeated again and the mixture was further refluxed for 4 h. The solution was evaporated in vacuo, and the residue was extracted with a large amount of CHCl₃. The CHCl₃ solution was dried over anhyd. MgSO₄ and the solvent was evaporated off. Recrystallization of the residue from MeOH–acetic acid furnished 19.2 mg of colorless needles, mp 249 °C (dec.), [α]D³⁰ +147° (c = 0.16, EtOH). This compound was identified as O.O-dimethyloctopamine methiodide (8) by melting point determination and by comparison of the IR spectra and behavior on TLC.¹³⁰

9-Monodeuteriopaeonoxine (3) — A solution of 1 (64.6 mg) and NaOH (25 mg) in D₂O (3.5 ml) was heated in a sealed tube at 100 °C for 50 h. The alkaline solution was acidified with 3% aqueous HCl, washed with Et₂O, made alkaline with NH₄OH, and extracted with Et₂O. The Et₂O extract was dried over anhyd. MgSO₄ and the solvent was evaporated off to give 51 mg of 3 as an oily mass. MS m/z: 342 (M⁺), 311 (base peak). ¹H-NMR (CDCl₃) δ ppm: 2.53 (3H, s, NCH₃), 3.49, 3.53, 3.86 (3H × 3, each s, three OCH₃), 5.00—5.83 (1H, br. exchanged by D₂O, OH), 6.68 and 6.87 (1H × 2, each s, two aromatic H).

Nandigerine (10) — Light green crystals, mp 122—124 °C (from MeOH) [lit.,¹²⁷ solvent-free base: mp 176—177 °C (from MeOH); MeOH solvate: mp 99—100 °C]. [α]D³⁰ +380° (c = 0.25, EtOH). The UV, IR, and ¹H-NMR spectra were indistinguishable from those of an authentic sample of nandigerine (10). Picrolonate: decomposes at 218—219 °C (from MeOH). Anal. Calcd for C₁₅H₁₀N₂O₄·C₁₀H₃NO₃·C: 58.43; H, 4.38; N, 12.17. Found: C, 58.66; H, 4.46; N, 12.22.

Isolation of Nandigerine (10) from the Stem Wood — The crude phenolic bases (6.1 g) from the stem wood were treated with picrolonic acid in CHCl₃, but the picrolonate was not obtained in a crystalline form. The oily picrolonate and the mother liquor were combined and free bases were obtained by the procedure described above. The free bases were separated by differential pH extraction¹⁴ (McIlvaine buffer solution, double strength, pH 5.8 and 4.8) and by the use of 0.2 m citric acid to yield 3.255, 0.437 and 0.113 g, respectively, of crude bases. The crude alkaloids (2.578 g) from the pH 5.8 extract were dissolved in a small amount of acetone and an excess of saturated solution of p-nitrobenzoic acid in acetone was added to yield crystalline nandigerine (10) p-nitrobenzoate (2.213 g). The salt was recrystallized from MeOH to give pale yellow crystals, mp 193—194 °C. Nandigerine (10) (0.506 g) was liberated from the salt. The UV, IR, and ¹H-NMR spectra were superimposable on those of an authentic sample of nandigerine (10).

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References and Notes

1) A part of this work was presented at The 20th Annual Meeting of the Kinki Branch, Pharmaceutical Society of Japan, Kobe, November 1970 (M. Kozuka, A. Inada, T. Konoshima, and T. Sawada, Abstracts of Papers, p. 27).
2) Present address: Faculty of Pharmaceutical Sciences, Setsunan University, 45-1 Nagao Tohge-cho, Hirakata 573-01, Japan.
8) The sample was kindly donated by Professor J. Kunitomo.
9) Praeoxine was not identical with corydine (5) or isocorydine (6).
13) Kieselgel G (Merck) in acetone–MeOH (1:1, v/v).