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There are some naturally occurring pyrazines which carry hydroxyl groups on the alkyl side chains.¹⁻⁷⁾ Among these hydroxyalkylpyrazines, deoxyneo- β -hydroxyaspergillic acid (I),⁸⁾ *dl*-neohydroxyaspergillic acid (II)⁹⁾ and mutaaspergillic acid (IIIa)¹⁰⁾ have already been synthesized. In the case of the former two compounds, a hydroxyl group was introduced into the side chain by the reaction of 2-chloro-3,6-diisobutylpyrazine 1- and 4-oxides with acetic anhydride.^{8,9)} On the other hand, ring closure of 4-methyl-2-(3-hydroxy-3-methyl-2-oxobutylamino)valerohydroxamic acid was undertaken for the synthesis of IIIa, which carries a tertiary hydroxyl group on the isopropyl side chain.¹⁰⁾ In a previous communication,¹¹⁾ we described a hydroxylation of the α -position of the alkyl groups of alkylpyrazines by oxygen in the presence of a base, and a convenient synthesis of II. The present paper reports a continuation of our study on the hydroxylation of the side chain of alkylpyrazines and describes the syntheses of IIIa and *dl*-hydroxyaspergillic acid (IIIb).^{1,6)}

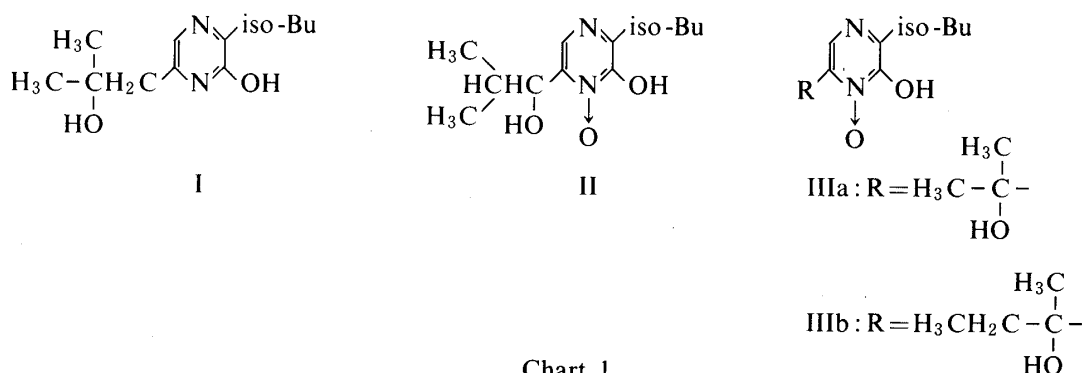


Chart 1

2-Chloro-3-isobutyl-6-isopropylpyrazine (Va) was obtained in the reported manner.¹²⁾ 2-Chloro-3-isobutyl-6-*sec*-butylpyrazine (Vb)¹³⁾ was similarly prepared by the reaction of DL-isoleucyl-leucyl anhydride (IVb)¹⁴⁾ with a mixture of phosphoryl chloride and phosphorus pentachloride, and purified by careful column chromatography on silica gel. Both 2-chloropyrazines were oxidized with persulfate in concentrated sulfuric acid¹⁵⁾ to afford the corresponding 1-oxides (VIIIa and VIIIb). In order to determine their structures, the products (VIIIa and VIIIb) were hydrolyzed in an alkaline medium to afford the corresponding hydroxypyrazines (IXa and IXb),¹⁵⁾ which gave a positive hydroxamic acid coloration with ferric chloride. Compounds IXa and IXb were shown to be identical with authentic specimens of 2-hydroxy-3-isobutyl-6-isopropylpyrazine 1-oxide¹²⁾ and *dl*-aspergillic acid¹³⁾ respectively, by

Experimental

Melting points were recorded on a Yanagimoto micro-melting point apparatus and are uncorrected. Boiling points are also uncorrected. Gas chromatograms were recorded on a Shimadzu GC-4B unit, ultraviolet (UV) spectra on a Hitachi 557 spectrophotometer, IR spectra on a Shimadzu IR-400 spectrometer, and ^1H -NMR spectra on a Varian EM-390 instrument with tetramethylsilane as an internal standard. Mass spectra (MS) were obtained with a Hitachi M-80 spectrometer.

2-Chloro-3-isobutyl-6-sec-butylpyrazine (Vb)—A mixture of DL-isoleucyl-leucyl anhydride¹⁴⁾ (20 g, 88.5 mmol), POCl_3 (50 ml), and PCl_5 (10 g) was heated at 130–140°C for 1 h in a sealed tube, then poured into ice-water, made alkaline with K_2CO_3 and extracted with Et_2O . The Et_2O layer was dried over Na_2SO_4 and the solvent was evaporated off to give a brown oil, which was dissolved in hexane. The hexane solution was extracted with conc. HCl. The HCl layer was made alkaline with K_2CO_3 and extracted with Et_2O to give a brown oil, which was applied to a silica gel (Wakogel C-200, 400 g) column and eluted with hexane containing increasing amounts of Et_2O . The hexane– Et_2O (58:1) fractions gave VIb as a colorless oil (7.1 g, 32%), bp 105–106°C (4 Torr). Further elution with the same developer gave a mixture of VIb and Vb (4.2 g) and then Vb as a colorless oil (4.7 g, 21%), bp 110–111°C (4 Torr). Both monochloropyrazines were shown to be identical with authentic samples¹³⁾ by a comparison of IR spectra.

The hexane layer was washed with H_2O and dried over Na_2SO_4 . Evaporation of the solvent gave VIIb as a pale yellow oil, which was purified by distillation to furnish a colorless oil, bp 115–116°C (4 Torr). *Anal.* Calcd for $\text{C}_{12}\text{H}_{18}\text{Cl}_2\text{N}_2$: C, 55.18; H, 6.95; N, 10.73. Found: C, 55.04; H, 6.95; N, 10.61. MS m/e : 264 ($\text{M}^+ + 4$), 262 ($\text{M}^+ + 2$), 260 (M^+). ^1H -NMR (CDCl_3) δ ppm: 0.86 [3H, t, $J=7.5$ Hz, $\text{CH}(\text{CH}_3)\text{CH}_2\text{CH}_3$], 0.97 [6H, d, $J=6$ Hz, $\text{CH}_2\text{CH}(\text{CH}_3)_2$], 1.26 [3H, d, $J=7$ Hz, $\text{CH}(\text{CH}_3)\text{CH}_2\text{CH}_3$], 1.45–1.94 [2H, m, $\text{CH}(\text{CH}_3)\text{CH}_2\text{CH}_3$], 2.01–2.46 [1H, m, $\text{CH}_2\text{CH}(\text{CH}_3)_2$], 2.77 [2H, d, $J=6$ Hz, $\text{CH}_2\text{CH}(\text{CH}_3)_2$], 3.05–3.56 [1H, m, $\text{CH}(\text{CH}_3)\text{CH}_2\text{CH}_3$]. UV $\lambda_{\text{max}}^{\text{ethanol}}$ nm (log ϵ): 221 (4.13), 285 (3.92, sh), 297 (3.99).

2-Chloro-3-isobutyl-6-isopropylpyrazine 1-Oxide (VIIIa)— $\text{K}_2\text{S}_2\text{O}_8$ (7.7 g, 28 mmol) was added portionwise to a solution of Va (5.0 g, 23.5 mmol) in conc. H_2SO_4 (25 ml), over a period of 30 min under stirring at room temperature. The reaction mixture was stirred for a further 24 h, poured into ice-water (80 ml) and extracted with CHCl_3 . The CHCl_3 layer was washed successively with 10% KHCO_3 and water, and dried over Na_2SO_4 . Evaporation of the CHCl_3 gave a pale yellow oil (ca. 5 g), which was chromatographed on silica gel (Wakogel C-200, 55 g) with hexane– Et_2O (7:3) and Et_2O . From the former eluate, the starting material (2.8 g, 56%) was recovered. The Et_2O fractions gave VIIIa (2.2 g, 41%), which was distilled to furnish a colorless oil, bp 115°C (1 Torr). *Anal.* Calcd for $\text{C}_{11}\text{H}_{17}\text{ClN}_2\text{O}$: C, 57.76; H, 7.49; N, 12.25. Found: C, 57.58; H, 7.50; N, 12.10. MS m/e : 228 (M^+). ^1H -NMR (CDCl_3) δ ppm: 0.99 [6H, d, $J=6$ Hz, $\text{CH}_2\text{CH}(\text{CH}_3)_2$], 1.33 [6H, d, $J=6$ Hz, $\text{CH}(\text{CH}_3)_2$], 2.01–2.46 [1H, m, $J=6$ Hz, $\text{CH}_2\text{CH}(\text{CH}_3)_2$], 2.83 [2H, d, $J=6$ Hz, $\text{CH}_2(\text{CH}_3)_2$], 3.40–3.85 [1H, m, $J=6$ Hz, $\text{CH}(\text{CH}_3)_2$], 8.30 (1H, s, pyrazine H). UV $\lambda_{\text{max}}^{\text{ethanol}}$ nm (log ϵ): 232 (4.39), 270–272 (4.06), 299.5–302 (3.52), 311–312 (3.51, sh).

2-Chloro-3-isobutyl-6-sec-butylpyrazine 1-Oxide (VIIIb)—A solution of Vb (2.5 g, 11 mmol) in conc. H_2SO_4 (12 ml) was treated with $\text{K}_2\text{S}_2\text{O}_8$ (3.6 g, 13 mmol) and worked up as described for the synthesis of VIIIa. The products were chromatographed on Wakogel C-200 (30 g). The fractions eluted with hexane– Et_2O (7:3) gave the starting material (0.98 g, 40%) and those eluted with Et_2O gave VIIIb (1.5 g, 57%). The product (VIIIb) was purified by distillation to furnish a colorless oil, bp 130–131°C (4 Torr). *Anal.* Calcd for $\text{C}_{12}\text{H}_{19}\text{ClN}_2\text{O}$: C 59.37; H, 7.89; N, 11.54. Found: C, 59.65; H, 7.90; N, 11.51. MS m/e : 242 (M^+). ^1H -NMR (CDCl_3) δ ppm: 0.95 [3H, t, $J=7$ Hz, $\text{CH}(\text{CH}_3)\text{CH}_2\text{CH}_3$], 1.00 [6H, d, $J=7$ Hz, $\text{CH}_2\text{CH}(\text{CH}_3)_2$], 1.32 [3H, d, $J=7$ Hz, $\text{CH}(\text{CH}_3)\text{CH}_2\text{CH}_3$], 1.55–2.03 [2H, m, $J=7$ Hz, $\text{CH}(\text{CH}_3)\text{CH}_2\text{CH}_3$], 2.03–2.46 [1H, m, $J=7$ Hz, $\text{CH}_2\text{CH}(\text{CH}_3)_2$], 2.81 [2H, d, $J=7$ Hz, $\text{CH}_2\text{CH}(\text{CH}_3)_2$], 3.27–3.65 [1H, m, $J=7$ Hz, $\text{CH}(\text{CH}_3)\text{CH}_2\text{CH}_3$], 8.25 (1H, s, pyrazine H). UV $\lambda_{\text{max}}^{\text{ethanol}}$ nm (log ϵ): 307 (4.40), 270–271 (4.06), 301–302 (3.56), 311–312 (3.51, sh).

2-Hydroxy-3-isobutyl-6-isopropylpyrazine 1-Oxide (IXa)—A solution of VIIIa (83 mg, 0.36 mmol) in a mixture of 20% KOH (3 ml) and MeOH (3 ml) was refluxed for 2 h, then the MeOH was removed by distillation *in vacuo*. The residual alkaline solution was extracted with Et_2O . The water layer was acidified with 2N HCl and extracted with CH_2Cl_2 . The extract was dried over Na_2SO_4 , and the solvent was distilled off to give IXa as a pale yellow solid (64 mg, 84%), which was recrystallized from MeOH– H_2O to furnish pale yellow needles, mp 88–91°C, undepressed on admixture with an authentic sample (mp 92–93°C).¹²⁾

dl-Aspergillilic Acid (IXb)—A solution of VIIIb (46 mg, 0.19 mmol) in a mixture of 20% KOH (3 ml) and MeOH (3 ml) was worked up as described in the above-mentioned experiment, to afford IXb (36 mg, 86%) as pale yellow needles, mp 92–94°C, undepressed on admixture with an authentic sample (mp 93–94.5°C).¹³⁾

Hydroxylation of 2-Chloro-3-isobutyl-6-isopropylpyrazine 1-Oxide (VIIIa)—Oxygen was bubbled into a THF (30 ml) solution of VIIIa (490 mg, 2.1 mmol) and LDA [prepared from 4.1 ml (2.6 mmol) of a BuLi–hexane solution (0.64 M) and 260 mg (2.6 mmol) of diisopropylamine] for 5 h at –78°C, then the solvent was distilled off under reduced pressure. The resulting brown oily substance was triturated with water (30 ml) and extracted with Et_2O . The water layer was acidified with 2N HCl and extracted with CH_2Cl_2 .

The Et_2O extract was dried over Na_2SO_4 and concentrated to give an oily residue (ca. 500 mg), which was

applied to a silica gel (Wakogel C-200, 50 g) column and eluted with benzene containing increasing amounts of Me₂CO. The benzene–Me₂CO (80:1) eluate gave the starting material (106 mg, 22%). From the benzene–Me₂CO (30:1) fractions, Xa (98 mg, 19%) was obtained as a colorless oil, bp 120–125°C (3 Torr) (oil bath temp.).

The CH₂Cl₂ extract was shaken with 0.05 M NaHCO₃. The organic layer was dried over Na₂SO₄, and the solvent was evaporated off to give IXa (40 mg, 9%) as a pale yellow solid. The aqueous layer was acidified with 2 N HCl and extracted with CHCl₃. The extract was dried over Na₂SO₄. Evaporation of the solvent afforded IIIa (28 mg, 6%) as pale yellow crystals, which were recrystallized from MeOH–H₂O to furnish yellow prisms, mp 170–173°C (lit.⁶⁾ mp 173–174°C, lit.¹⁰⁾ mp 167–168°C).

IIIa: MS *m/e*: 226 (M⁺), 211 (M⁺–CH₃), 209 (M⁺–OH). ¹H-NMR (CDCl₃) δ ppm: 0.97 [6H, d, *J*=6 Hz, CH₂CH(CH₃)₂], 1.70 [6H, s, C(OH)(CH₃)₂], 1.90–2.55 [1H, m, *J*=6 Hz, CH₂CH(CH₃)₂], 2.75 [2H, d, *J*=6 Hz, CH₂CH(CH₃)₂], 5.85–6.26 [2H, br s, C(OH)(CH₃)₂ and pyrazinol-OH], 7.84 (1H, s, pyrazine H). UV λ_{max}^{ethanol} nm (log ε): 235.5 (3.89), 338–339 (3.66).

Xa: High resolution MS Calcd for C₁₁H₁₇ClN₂O₂: 244.0978. Obsd: 244.0981. MS *m/e*: 244 (M⁺), 229 (M⁺–CH₃). ¹H-NMR (CDCl₃) δ ppm: 1.00 [6H, d, *J*=6 Hz, CH₂CH(CH₃)₂], 1.84 [6H, s, C(OH)(CH₃)₂], 2.04–2.50 [1H, m, *J*=6 Hz, CH₂CH(CH₃)₂], 2.85 [2H, d, *J*=6 Hz, CH₂CH(CH₃)₂], 6.05 [1H, br s, C(OH)(CH₃)₂], 8.50 (1H, s, pyrazine H). UV λ_{max}^{ethanol} nm (log ε): 231 (4.27), 271 (3.96), 297–301 (3.48, sh), 311 (3.41, sh). IR (liq. film) cm^{–1}: 3430 (OH).

Hydrolysis of 2-Chloro-3-isobutyl-6-(α-hydroxy)isopropylpyrazine 1-Oxide (Xa)—A solution of Xa (94 mg, 0.38 mmol) in a mixture of MeOH (3 ml) and 20% KOH (3 ml) was refluxed for 2 h, then worked up as described for the hydrolysis of VIIIa to give IIIa (80 mg, 92%) as pale yellow prisms.

Hydroxylation of 2-Chloro-3-isobutyl-6-sec-butylpyrazine 1-Oxide (VIIIb)—A THF (30 ml) solution of VIIIb (508 mg, 2.1 mmol) was treated with oxygen in the presence of LDA [prepared from 3.9 ml (2.5 mmol) of a BuLi–hexane solution (0.64 M) and 248 mg (2.5 mmol) of diisopropylamine]. The reaction mixture was worked up as described for the hydroxylation of VIIIa to afford Xb (110 mg, 20%) as a colorless oil, bp 120–121°C (3 Torr) (oil bath temp.), IIb (44 mg, 9%) as pale yellow prisms (recrystallized from hexane), mp 148–151°C (lit.¹¹⁾ mp 149°C, lit.⁶⁾ mp 155°C), and IXb (68 mg, 15%) as pale yellow prisms.

IIIb: MS *m/e*: 240 (M⁺), 224 (M⁺–O), 223 (M⁺–OH), 211 (M⁺–C₂H₅). ¹H-NMR (CDCl₃) δ ppm: 0.90 [3H, t, *J*=8 Hz, C(OH)(CH₃)CH₂CH₃], 0.98 [6H, d, *J*=6 Hz, CH₂CH(CH₃)₂], 1.67 [3H, s, C(OH)(CH₃)–CH₂CH₃], 1.91–2.46 [3H, m, CH₂CH(CH₃)₂], C(OH)(CH₃)CH₂CH₃], 2.76 [2H, d, *J*=6 Hz, CH₂CH(CH₃)₂], 5.30–5.80 [2H, br s, C(OH)(CH₃)CH₂CH₃, pyrazinol-OH], 7.75 (1H, s, pyrazine H). UV λ_{max}^{ethanol} nm (log ε): 235 (4.16), 333 (3.84).

Xb: High resolution MS Calcd for C₁₂H₁₉ClN₂O₂: 258.1135. Obsd: 258.1104. MS *m/e*: 243 (M⁺–CH₃), 241 (M⁺–OH), 229 (M⁺–C₂H₅). ¹H-NMR (CDCl₃) δ ppm: 0.92 [3H, t, *J*=7.5 Hz, C(OH)(CH₃)CH₂CH₃], 1.00 [6H, d, *J*=6 Hz, CH₂CH(CH₃)₂], 1.65 [3H, s, C(OH)(CH₃)CH₂CH₃], 2.10 [2H, q, *J*=7.5 Hz, C(OH)(CH₃)CH₂CH₃], 2.08–2.50 [1H, m, CH₂CH(CH₃)₂], 2.87 [2H, d, *J*=7.5 Hz, CH₂CH(CH₃)₂], 5.93 [1H, s, C(OH)(CH₃)CH₂CH₃], 8.43 (1H, s, pyrazine H). UV λ_{max}^{ethanol} nm (log ε): 232.5 (4.22), 271–273 (3.86), 300–302 (3.41, sh).

Hydrolysis of 2-Chloro-3-isobutyl-6-(α-hydroxy)-sec-butylpyrazine 1-Oxide (Xb)—Compound Xb (29 mg, 0.11 mmol) was hydrolyzed in a mixture of MeOH (2 ml) and 20% KOH (2 ml), as described for the hydrolysis of VIIIa, to give IIIb (23 mg, 89%) as pale yellow needles.

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