PYRIMIDINE NUCLEOSIDES OF THE FURANOSE FORM OF 2-AMINO-2-DEOXY-D-XYLOSE*

M. L. WOLFROM† AND P. J. CONIGLIARO
Department of Chemistry, The Ohio State University, Columbus, Ohio 43210 (U. S. A.)
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ABSTRACT

Periodate oxidation of ethyl 2-deoxy-1-thio-2-(trifluroacetamido)-α-D-glucofuranoside, followed by immediate reduction of the product with sodium borohydride, yielded ethyl 2-deoxy-1-thio-2-(trifluoroacetamido)-α-D-xylofuranoside, which was acetylated with acetic anhydride–pyridine to give ethyl 3,5-di-*O*-acetyl-2-deoxy-1-thio-2-(trifluoroacetamido)-α-D-xylofuranoside (**3**). Treatment of **3** with chlorine yielded 3,5-di-*O*-acetyl-2-deoxy-2-(trifluoroacetamido)-D-xylofuranosyl chloride (**4**), which was immediately condensed with bis(trimethylsilyl)thymine to give 1-[3,5-di-*O*-acetyl-2-deoxy-2-(trifluoroacetamido)-β-D-xylofuranosyl]thymine (**5**). Deacylation of **5** with methanolic ammonia yielded 1-(2-amino-2-deoxy-β-D-xylofuranosyl)thymine, isolated as the hydrochloride. Condensation of **4** with bis(trimethylsilyl)uracil gave 1-[3,5-di-*O*-acetyl-2-deoxy-2-(trifluoroacetamido)-β-D-xylofuranosyl]uracil, which was deacylated with methanolic ammonia to give 1-(2-amino-2-deoxy-β-D-xylofuranosyl)uracil, isolated as the hydrochloride.

INTRODUCTION

The synthesis of nucleosides of 2-amino-2-deoxy sugars has been of interest in this laboratory, because of the possible utility of these compounds as cancer chemotherapeutic agents. Because the sugar moieties of naturally occurring nucleosides are, for the most part, pentoses in their furanose form, the synthesis of nucleosides of the furanose form of 2-amino-2-deoxypentoses is of special interest. Thus far, the only nucleosides of this type to be synthesized are purine nucleosides of 2-amino-2-deoxy-D-ribose, namely, 9-(2-amino-2-deoxy-β-D-ribofuranosyl)-6-(dimethylamino)purine[1] and the anomers of 9-(2-amino-2-deoxy-D-ribofuranosyl)adenine[2]. In addition, a purine nucleoside of 2-amino-2,3-dideoxy-D-*erythro*-pentose, namely, 9-(2-amino-2,3-dideoxy-β-D-*erythro*-pentofuranosyl)adenine[3], has been described. We now report

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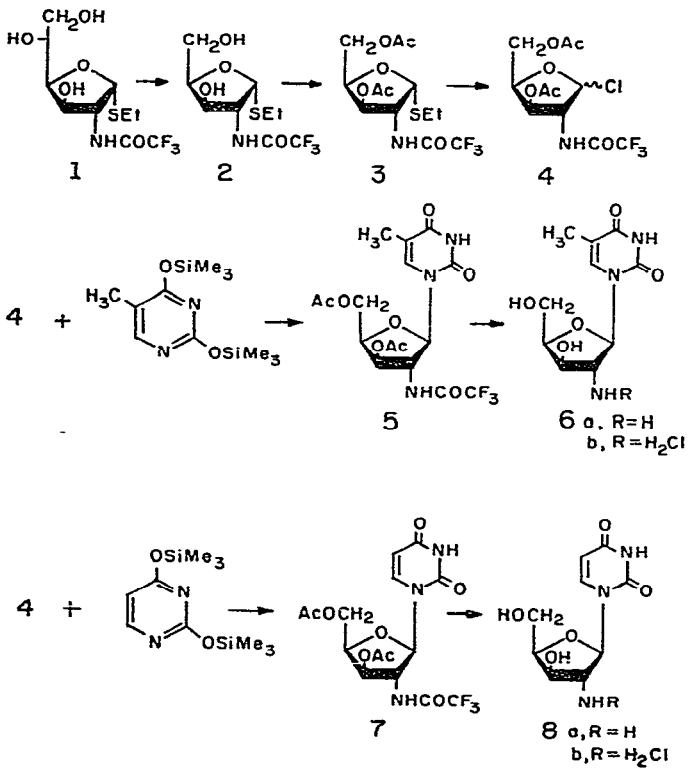
the synthesis of two pyrimidine nucleosides of the furanose form of 2-amino-2-deoxy-D-xylose: 1-(2-amino-2-deoxy-β-D-xylofuranosyl)thymine and 1-(2-amino-2-deoxy-β-D-xylofuranosyl)uracil. The trifluoroacetyl group was employed as the amino-protecting group.

DISCUSSION

The starting compound for these syntheses was ethyl 2-deoxy-1-thio-2-(trifluoroacetamido)-α-D-glucofuranoside[4] (**1**). Treatment of **1** with slightly more than one molar equivalent of sodium metaperiodate, followed by immediate reduction of the product with sodium borohydride, gave crystalline ethyl 2-deoxy-1-thio-2-(trifluoroacetamido)-α-D-xylofuranoside (**2**) in 73% yield. The procedure followed was similar to that employed by Wolfrom and Anno[5] for the synthesis of ethyl 2-acetamido-2-deoxy-1-thio-α-D-xylofuranoside from ethyl 2-acetamido-2-deoxy-1-thio-α-D-glucofuranoside, but was modified as described by Wolfrom and Winkley[6]. Compound **2** was acetylated with acetic anhydride–pyridine, to give crystalline ethyl 3,5-di-*O*-acetyl-2-deoxy-1-thio-2-(trifluoroacetamido)-α-D-xylofuranoside (**3**) in 84% yield.



Treatment of a solution of **3** in dichloromethane with dry chlorine yielded syrupy 3,5-di-*O*-acetyl-2-deoxy-2-(trifluoroacetamido)-β-D-xylofuranosyl chloride (**4**).

which was immediately condensed with bis(trimethylsilyl)thymine[7,8] by the fusion technique[8,9]. The product was purified by preparative t.l.c., to give amorphous 1-[3,5-di-*O*-acetyl-2-deoxy-2-(trifluoroacetamido)-β-D-xylofuranosyl]thymine (**5**) in 61% yield from **3**. Examination of the crude product by t.l.c. indicated that no significant proportion of a second anomeric nucleoside derivative was present. Complete deacylation of **5** was effected with methanolic ammonia at room temperature, to give 1-(2-amino-2-deoxy-β-D-xylofuranosyl)thymine (**6a**), isolated as the crystalline hydrochloride (**6b**) in 86% yield from **5**. The anomeric assignments for **5** and **6** were made on the basis of optical rotatory dispersion (o.r.d.). The o.r.d. spectrum of **6b**

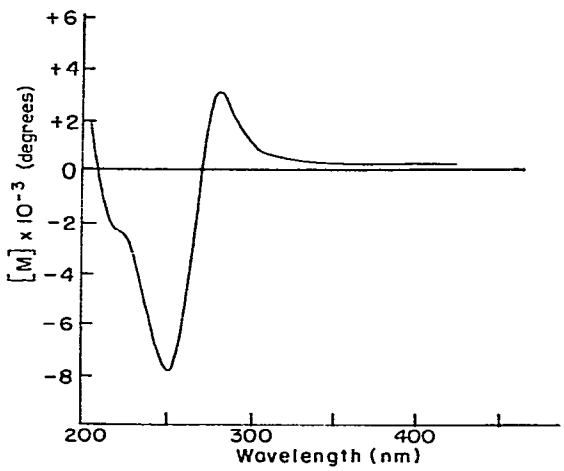


Fig. 1. Optical rotatory dispersion spectrum of 1-(2-amino-2-deoxy-β-D-xylofuranosyl)thymine hydrochloride (**6b**).

measured in water (see Fig. 1) exhibited a positive Cotton effect. This positive Cotton effect has been shown[10–12] to be characteristic of the β-D configuration in pyrimidine nucleosides of the furanose form.

Condensation of freshly prepared 3,5-di-*O*-acetyl-2-deoxy-2-(trifluoroacetamido)-D-xylofuranosyl chloride (**4**) with bis(trimethylsilyl)uracil[7,8] by the fusion technique[8,9] gave amorphous 1-[3,5-di-*O*-acetyl-2-deoxy-2-(trifluoroacetamido)-β-D-xylofuranosyl]uracil (**7**) in 48% yield from **3**. Compound **7** was deacylated with methanolic ammonia, to give 1-(2-amino-2-deoxy-β-D-xylofuranosyl)uracil (**8a**), isolated as the crystalline hydrochloride (**8b**) in 80% yield from **7**. The anomeric assignments for **7** and **8** were made on the basis of the o.r.d. spectrum of **8b**. This spectrum, measured in water (see Fig. 2), exhibited a positive Cotton effect, characteristic[10–12] of the β-D configuration of pyrimidine nucleosides of the furanose form.

EXPERIMENTAL

Melting points were determined with a Thomas–Hoover apparatus. Specific rotations were determined in a 2-dm polarimeter tube. I.r. spectra were recorded with

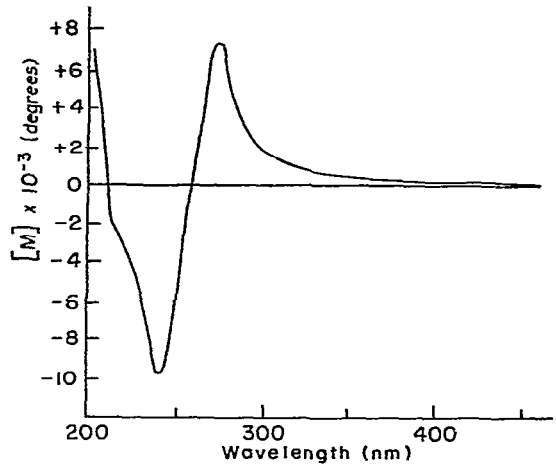


Fig. 2. Optical rotatory dispersion spectrum of 1-(2-amino-2-deoxy-β-D-xylofuranosyl)uracil hydrochloride (8b).

a Perkin–Elmer Infracord spectrometer. Ultraviolet spectra were recorded with a Bausch and Lomb Spectronic 505 spectrometer. O.r.d. spectra were recorded with a Jasco ORD/UV5 spectrometer. X-Ray powder diffraction data give interplanar spacings (Å) for CuKα radiation. Relative intensities were estimated visually: m, moderate; s, strong; v, very; w, weak. The stronger lines are numbered (1, strongest); multiple numbers indicate approximately equal intensities. T.l.c. was performed with Desaga equipment, by using Silica Gel G (E. Merck, Darmstadt, Germany), activated at 110°. Indication was effected by sulfuric acid, unless otherwise noted; proportions for developers are given by volume. Evaporations were conducted under diminished pressure (water aspirator). Microanalyses were made by W. N. Rond.

Ethyl 2-deoxy-1-thio-2-(trifluoroacetamido)-α-D-xylofuranoside (2). — To a cold (10°) solution of ethyl 2-deoxy-1-thio-2-(trifluoroacetamido)-α-D-glucofuranoside[4] (1, 1.91 g) in 50% methanol (40 ml) was added a solution of sodium metaperiodate (1.35 g, 1:1.05 molar ratio) in water (25 ml) at 10°. The solution was kept in the dark for 30 min at 10°, a solution of barium chloride dihydrate (0.77 g) in water (6 ml) was added, and the resulting precipitate of barium iodate was removed by filtration.

To the filtrate was added dropwise, with stirring, a solution of sodium borohydride (0.30 g) in water (6 ml) during 10 min. After being stirred for an additional 30 min, the pH of the solution was brought to 7 with 0.5M sulfuric acid, the solution was concentrated to a small volume, and the water remaining was removed by repeated addition and evaporation of absolute ethanol. The resulting solid residue was extracted with acetone (100 ml), the extract was evaporated, and the residue was chromatographed through a column (2.3 × 35 cm) of silica gel*, with ethyl acetate as the developer. After the first 50 ml of eluate had been discarded, 1 liter

*Grade 950, 60–200 mesh; W. R. Grace, Division of Davison Chemical Co., Baltimore, Md.

of eluate was collected, and evaporated to dryness. The residue was crystallized from acetone; yield 1.26 g (73%), m.p. 139–141°, $[\alpha]_D^{20}$ +242 ±1° (*c* 2.5, methanol); λ_{max}^{KBr} 3.0–3.1 (OH, NH), 5.88 (*N*-trifluoroacetyl carbonyl) 6.45 (NH), 8.62 (CF), 3.42, 6.8, 7.3, 7.5, 7.68, 7.96, 8.2–8.4, 9.1, 9.56, 9.7, 9.82, 10.85, 11.52, 12.1, 12.75, 13.08, and 13.6 μm; X-ray powder diffraction data: 7.44 s (2), 5.01 s (3), 4.31 vs (1), 4.10 m, 4.03 m, 3.90 w, 3.71 m, 3.26 w, 3.11 w, 2.95 s, 2.81 m, 2.69 w, 2.55 w, 2.42 w, 2.34 w, 2.28 w, 2.22 w, 2.12 w, 2.06 vw, and 1.97 w.

Anal. Calc. for $C_9H_{14}F_3NO_4S$: C, 37.37; H, 4.88; N, 4.48; S, 11.08. Found: C, 37.64; H, 5.17; N, 5.16; S, 11.53.

This compound was homogeneous by t.l.c. with 1:1 chloroform–acetone or ethyl acetate as the developer.

*Ethyl 3,5-di-*O*-acetyl-2-deoxy-1-thio-2-(trifluoroacetamido)-α-*D*-xylofuranoside* (**3**). — Compound **2** (2.2 g) was added to a mixture of acetic anhydride (6 ml) and pyridine (10 ml). After 24 h at room temperature, the solution was poured, with stirring, into ice and water (30 ml), and the mixture was extracted with dichloromethane (100 ml). The extract was washed with water, dried (sodium sulfate), and evaporated to a colorless syrup which was crystallized from ether–hexane; yield 2.38 g (84%), m.p. 60–61.5°, $[\alpha]_D^{21}$ +159 ±1° (*c* 3.3, chloroform); λ_{max}^{KBr} 3.1 (NH), 5.75 (*O*-acetyl carbonyl), 5.85 (*N*-trifluoroacetamido carbonyl), 6.42 (NH), 8.0–8.2 (ester), 8.65 (CF), 7.28, 8.28, 8.43, 9.03, 9.55, 9.8, 10.6, 11.36, 12.05, and 13.5 μm; X-ray powder diffraction data: 9.41 s (1), 6.37 w, 5.44 w, 4.89 s (3,3), 4.58 s (2), 4.33 m, 4.10 s (3,3), 3.93 m, 3.74 m, 3.58 w, 3.45 w, 3.24 m, 3.04 m, 2.91 vw, 2.84 vw, 2.71 w, 2.64 w, 2.53 m, 2.34 w, and 2.24 w.

Anal. Calc. for $C_{13}H_{18}F_3NO_6S$: C, 41.82; H, 4.86; N, 3.75; S, 8.59. Found: C, 41.86; H, 4.83; N, 4.03; S, 9.04.

This compound was homogeneous by t.l.c. with 2:1 ether–hexane as the developer.

*1-[3,5-Di-*O*-acetyl-2-deoxy-2-(trifluoroacetamido)-β-*D*-xylofuranosyl]thymine* (**5**). — Compound **3** (1.35 g) was dissolved in dichloromethane (20 ml), and dry chlorine was passed through the solution for 15 min. Evaporation of the solvent at 20° gave a pale-yellow syrup; this was redissolved in dichloromethane (10 ml) and a few drops of cyclohexene were added. After evaporation of the solvent at 20°, the resulting syrup was dissolved in chloroform, bis(trimethylsilyl)thymine[7,8] (2.0 g) was added and the mixture was stirred until homogeneous. The solvent was then evaporated, and the residue was heated for 15 min at 110–120° under diminished pressure (water aspirator), and cooled to room temperature. Ethanol (80%, 30 ml) was added and the mixture was heated for 15 min at 60°, with stirring. The solvent was then evaporated, the residue was extracted with chloroform (200 ml), and the extract was washed with water, dried (sodium sulfate), and evaporated, to yield a pale-amber glass (1.6 g). T.l.c. of the crude product with 3:2 chloroform–acetone as the developer revealed two major components (R_F 0.85 and 0.55). The crude product was resolved on 24 chromatoplates (200 × 200 × 1 mm) with 3:2 chloroform–acetone as the developer, and indication by u.v. light. The two major zones were removed, and extracted with

acetone. Evaporation of the extract of the slower-moving zone (R_F 0.55) gave a clear glass (1.05 g). Attempted crystallization from chloroform–isopropyl ether gave compound **5** as a white, amorphous* material; yield 0.96 g (61%), m.p. 138–145° (softening above 85°), $[\alpha]_D^{21}$ −41 ±1° (*c* 3.1, chloroform); λ_{max}^{KBr} 3.1 (NH), 5.7–5.8 (*O*-acetyl and *N*-trifluoroacetyl carbonyl), 5.9, 6.42, 6.8 (NH, thymine), 8.05–8.2 (ester), 8.6 (CF), 7.3, 8.45, 8.96, 9.5, 11.08, and 12.7 μm; λ_{max}^{EtOH} 208 (ε 12,000) and 265 nm (ε 9,650).

Anal. Calc. for $C_{16}H_{18}F_3N_3O_8$: C, 43.94; H, 4.15; N, 9.61. Found: C, 44.07; H, 4.51; N, 9.40.

This compound was homogeneous by t.l.c. with 3:2 chloroform–acetone or 5:2 ethyl acetate–benzene as the developer.

Evaporation of the extract from the faster-moving zone (R_F 0.85) gave a clear glass (0.28 g). The i.r. spectrum of this substance resembled that of the starting material **3**; it lacked a strong absorption in the 5.9–6.2-μm range. The u.v.spectrum of this substance showed no absorption bands above 210 nm. It was, therefore, concluded that this substance was not a nucleoside derivative, and it was not investigated further. Several other minor components of the crude product were observed, but were not isolated. It was estimated that none of these compounds were present in greater than 5–8% yield.

*1-(2-Amino-2-deoxy-β-*D*-xylofuranosyl)thymine hydrochloride* (**6b**). — Compound **5** (0.40 g) was dissolved in methanol presaturated at 0° with ammonia (50 ml). After 7 days at room temperature, the solution was concentrated to ~5 ml, and ether (60 ml) was added. The resulting, flocculent precipitate was filtered off, and washed with ether, to give crude **6a** as an amorphous material, attempted crystallization of which from methanol–ether was unsuccessful.

Crude **6a** was dissolved in methanol, and 2M hydrochloric acid (0.8 ml) was added. The solvent was evaporated, and the excess of hydrogen chloride was removed by repeated addition and evaporation with ethanol. The residue was dissolved in methanol (15 ml), and ether (50 ml) was added. The resulting precipitate was filtered off, washed with ether, and crystallized from ethanol–ether; yield 0.23 g (86%), m.p. 226–227° (dec.), $[\alpha]_D^{21}$ +18 ±1° (*c* 1.2, water); o.r.d. spectrum (see Fig. 1): $[M]_{283}^{20}$ +4,400 ±500° (peak) and $[M]_{252}^{20}$ −8,000 ±600° (trough) (*c* 0.09, water); λ_{max}^{KBr} 2.9–3.0 (OH), 3.15–3.4 (NH_3^+), 5.85, 6.65, 6.83 (thymine), 7.12, 7.28, 7.78, 8.32, 8.7, 9.10, 9.5, 9.82, 10.2, 10.8, 12.3, 12.7, 12.9, and 13.7 μm; $\lambda_{max}^{H_2O}$ 207 (ε 9,080) and 267 nm (ε 9,920); X-ray powder diffraction data: 10.22 vw, 8.27 vs (1), 6.13 m, 5.79 m, 5.43 m, 4.87 m, 4.53 s, 4.19 s (3), 3.90 s, 3.69 s, 3.48 s, 3.27 s (2,2), 3.16 s(2,2), 3.04 vw, 2.93 m, 2.84 m, 2.65 m, 2.54 m, 2.51 w, 2.45 w, 2.39 w, 2.32 m, 2.22 w, 2.12 w, 1.99 w, and 1.87 w.

Anal. Calc. for $C_{10}H_{16}ClN_3O_5$: C, 40.88; H, 5.49; Cl, 12.07; N, 14.30. Found: C, 41.07; H, 5.30; Cl, 11.79; N, 14.32.

*1-[3,5-Di-*O*-acetyl-2-deoxy-2-(trifluoroacetamido)-β-*D*-xylofuranosyl]uracil* (**7**).

*It was concluded that this compound was amorphous because it had a blank, or foggy, X-ray powder diffraction pattern.

— Compound **3** (2.0 g) was dissolved in dichloromethane (20 ml), and dry chlorine was passed through the solution for 15 min. Evaporation of the solvent at 20° gave a pale-yellow syrup; this was redissolved in dichloromethane (10 ml) and a few drops of cyclohexene were added. After evaporation of the solvent at 20°, the residual syrup was dissolved in dichloromethane (10 ml), bis(trimethylsilyl)uracil[7,8] (3.5 g) was added, and the mixture was stirred until homogeneous. The solvent was evaporated, the residue was heated for 15 min at 110–120° under diminished pressure (water aspirator), and cooled to room temperature. The crude product was added to methanol (50 ml), and the mixture was heated for 15 min at 60°, with stirring. The solvent was then evaporated, the residue was extracted with chloroform (200 ml), and the extract was washed with water, dried (sodium sulfate), and evaporated to an amber glass (2.1 g). T.l.c. of the crude product with 3:2 chloroform–acetone as the developer revealed two major components (R_F 0.84 and 0.50). The crude product was resolved on 24 chromatoplates (200 × 200 × 1 mm) with 3:2 chloroform–acetone as the developer and indication by u.v. light. The zone having R_F 0.50 was removed, and extracted with acetone. Evaporation of the extract gave a clear glass (1.2 g). Attempted crystallization from chloroform–isopropyl ether gave **7** as a white, amorphous material; yield 1.09 g (48%), m.p. 127–135° (softening above 92°), $[\alpha]_D^{23}$ +6 ±1° (*c* 1.7, chloroform); λ_{max}^{KBr} 3.1 (NH), 5.7–5.8 (*O*-acetyl and *N*-trifluoroacetyl carbonyl), 5.92, 6.46, 6.78 (NH, uracil), 8.2 (ester), 8.66 (CF), 7.3, 7.95, 8.5, 8.66, 9.0, 9.55, 11.3, 12.25, 13.1, and 14.0 μm; λ_{max}^{EtOH} 208 (ε 10,800) and 262 nm (ε 9,660).

Anal. Calc. for $C_{15}H_{16}F_3N_3O_8$: C, 42.56; H, 3.81; N, 9.93. Found: C, 42.71; H, 3.97; N, 9.73.

This compound was homogeneous by t.l.c. with 3:2 chloroform–acetone or 5:2 ethyl acetate–benzene as the developer.

*1-(2-Amino-2-deoxy-β-*D*-xylofuranosyl)uracil hydrochloride* (**8b**). — Compound **7** (0.30 g) was deacylated with methanol presaturated at 0° with ammonia, and the crude, free nucleoside (**8a**) was converted into the hydrochloride (**8b**) by the procedure described for the preparation of **6b**. The yield, after crystallization from methanol–isopropyl ether, was 0.16 g (80%); m.p. 249–252° (dec.), $[\alpha]_D^{21}$ +38 ±2° (*c* 0.7, water); o.r.d. spectrum (see Fig. 2): $[M]_{280}^{23}$ +7,500 ±500° (peak) and $[M]_{244}^{23}$ −10,100 ±600° (trough) (*c* 0.01, water); λ_{max}^{KBr} 2.9, 3.05 (OH), 3.2–3.6 (NH_3^+), 5.85–5.9, 6.05, 6.28, 6.35, 6.6, 6.85 (uracil), 7.05, 7.2, 7.55, 7.7, 7.8, 8.0, 8.26, 8.4, 8.78, 9.02, 9.2, 9.6, 9.94, 11.1, 12.06, 12.43, 12.86, 13.2, and 13.78 μm; λ_{max}^{EtOH} 207 (ε 8,730) and 267 nm (ε 10,235); X-ray powder diffraction data: 10.28 w, 9.17 s, 7.31 w, 6.17 m, 5.83 s, 5.44 w, 5.07 m, 4.85 w, 4.67 vs (2), 4.51 m, 4.27 s, 4.00 vs (3,3), 3.90 vs (3,3), 3.77 m, 3.65 s, 3.49 w, 3.33 s, 3.22 m, 3.18 m, 3.05 vs (1), 2.99 m, 2.91 s, 2.74 w, 2.63 s, 2.55 m, 2.47 s, 2.42 s, 2.37 w, 2.33 m, and 2.27 s.

Anal. Calc. for $C_9H_{14}ClN_3O_5$: C, 38.65; H, 5.05; Cl, 12.68; N, 15.03. Found: C, 38.46; H, 5.18; Cl, 12.47; N, 14.99.

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