The organic phase was then dried (sodium sulfate), filtered, and concentrated to a small volume. Column chromatography, as described in the isolation of **7**, using methylene chloride as an eluent, yielded **22** in 77% yield: mp 101–104 °C from ethyl acetate–hexane; mass spectrum, *m/e* (relative %) 318 (9), 276 (21), 235 (7), 234 (51), 193 (13), 192 (100), 191 (22), 177 (12), 174 (12), 91 (3), 43 (27); ^{1}H NMR δ 7.00 (1 H, aromatic), 6.22 (1 H, vinyl, d), 3.56 (1 H, methine, d of q), 2.26 (3 H, vinyl acyl methyl), 2.24 (6 H, acetoxy methyl), 2.18 (3 H, aromatic methyl), 1.24 (3 H, alkyl methyl, $J_{CH_3,H}$ = 7 Hz).

Acknowledgment. I thank William K. Rohwedder for the mass spectra and Larry J. Tjarks for the proton magnetic resonance data.

Registry No. (±)-1, 77028-39-8; (±)-2, 77028-40-1; (±)-3, 77028-41-2; (±)-4, 77028-42-3; *cis*-(±)-5, 77028-43-4; *trans*-(±)-5, 77028-44-5; (±)-6, 77028-45-6; (±)-7, 77028-46-7; (±)-8, 77028-47-8; (±)-9, 77028-48-9; **10**, 77028-49-0; **11**, 77028-50-3; **12**, 77028-51-4; **13**, 77028-52-5; **14**, 77028-53-6; **15**, 77028-54-7; **16**, 77028-55-8; **17**, 77028-56-9; **18**, 77028-57-0; **19**, 77028-58-1; **20**, 77028-59-2; **21**, 77028-60-5; (±)-**22**, 77028-61-6.

Pseudodipeptides: A Novel Route to Serine-Containing Diastereomeric Analogues

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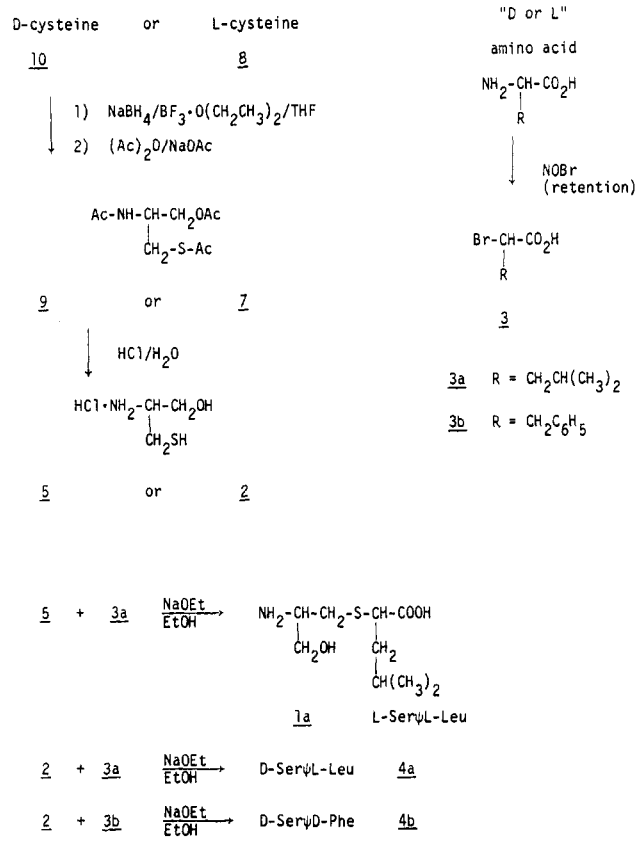
Received November 27, 1979

Continued interest in peptide analogues which contain methylenethio groups substituted for the peptide bond atoms has led us to explore synthetic routes to dipeptide analogues of the type SerψX (1) where ψ = CH_2S substituted for CONH and X may be one of a variety of amino acids. We have recently reported synthetic routes leading to XψGly pseudodipeptides[1] but this report represents the first example of a dipeptide analogue in which neither portion is formally derived from glycine. The presence of a serine side chain is of considerable interest in view of the many peptides and proteins in which serine residues perform important and apparently essential roles. Previous studies on a GlyψLeu analogue, first prepared by Yankeelov et al.,[2] have demonstrated that this analogue binds several times more tightly to aminopeptidase M than the natural substrate. Model building as well as enzymatic studies indicate that such analogues may satisfy binding requirements while being resistant to enzymatic hydrolysis.[2]

The serine-containing analogues can be easily prepared by utilizing the sulfur of cysteinol (**2**) as the nucleophile to displace the bromide of α-bromo acids (**3**). The cysteine side chain thus effectively undergoes a role reversal with the carboxyl-derived hydroxymethyl group and becomes part of the modified dipeptide linkage. Thus the resulting inversion of the chiral center forms the D-serine analogue (**4**) from L-cysteinol (**2**). It is of interest that a similar but inverse role reversal was utilized by Patchornik and coworkers in the synthesis of cysteine derivatives from serine analogues.[3]

Scheme I



The diastereomeric analogues L-SerψL-Leu (**1a**) and D-SerψL-Leu (**4a**) were prepared by the treatment of (*R*)-2-bromo-4-methylpentanoic acid (**3a**) with D-cysteinol (**5**) and L-cysteinol (**2**), respectively. The two diastereomers (**1a** and **4a**) could be completely resolved by reversed-phase high-performance liquid chromatography (HPLC) with retention times of 10.95 and 9.38 min, respectively. The HPLC chromatograms showed that neither analogue was contaminated by its diastereomer. To ensure that one diastereomer was not selectively crystallized from the other, chromatograms were obtained with the crude fraction prior to crystallization. The syntheses therefore proceed without racemization in either the formation of the bromo acid or the displacement of the bromide by the sulfur nucleophile, since racemization at *either* step would result in the formation of diastereomeric compounds.

The (*tert*-butyloxy)carbonyl (Boc)-derivatized dipeptide analogues can be prepared by standard procedures using di-*tert*-butyl dicarbonate,[4] and the analogues can be quantified by normal amino acid analysis. The preparation of peptides containing these pseudodipeptide units blocked as their *N*-α-*t*-Boc derivatives can proceed according to established methods of solution- and solid-phase peptide synthesis. By use of the latter method, analogues of LH–RH containing the dipeptide surrogates GlyψLeu and D-SerψLeu were recently prepared.[5] Thus, the availability of synthetic routes leading to XψY pseudodipeptides (Scheme I) should permit their facile incorporation into a wide variety of peptides of biological interest.

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0022-3263/81/1946-2393$01.25/0

Experimental Section

Nuclear magnetic resonance (NMR) data were obtained on a Perkin-Elmer R12A spectrometer and high-performance liquid chromatography (HPLC) data were obtained on a Dupont Model 850 chromatograph under reversed-phase conditions on a Zorbax ODS column, 4.6 mm × 25 cm, using a 0.25 M triethylammonium phosphate (TEAP, pH 3.5)/CH_3CN (97:3) system and monitoring at 230 nm. Microanalyses were performed by Galbraith Laboratories, Inc. Chemical-ionization mass spectrometric analyses were performed on a Finnigan 3200 quadrupole mass spectrometer at the University of Virginia, using methane as the reagent gas.

(*R*)-2-Bromo-4-methylpentanoic Acid (3a). Treatment of D-leucine (**6a**, 16.4 g, 125 mmol) with nitrosyl bromide,[6] followed by distillation of the crude product, gave 15.1 g (62%) of purified **3a**: bp 97–98 °C (0.25 mm); $[\alpha]^{22}_D$ +38.2 (*c* 2, methanol) [lit.[7] (*S* isomer) $[\alpha]^{27}_D$ −34° (methanol)].

(*S*)-2-Bromo-3-phenylpropanoic Acid (3b). Compound **3b** was prepared in the same manner as **3a** in 63% yield: bp 144–145 °C (0.35 mm); $[\alpha]^{25}_D$ −10.0° (*c* 2, methanol) [lit.[8] $[\alpha]^{20}_D$ −9.9° (*c* 5, absolute ethanol)]; ^1H NMR ($CDCl_3$) δ 3.36 (m, 2 H), 4.45 (m, 1 H), 7.29 (s, 5 H), 10.20 (s, 1 H).

***N,O,S*-Triacetyl-L-cysteinol (7).** L-Cysteine (**8**, 10.0 g, 83 mmol) was reduced by the procedure of Anhoury et al.[9] The crude cysteinol obtained was acetylated with acetic anhydride/sodium acetate to give 7.51 g (39%) of **7**: mp 95–97 °C; $[\alpha]^{25}_D$ −42.6° (*c* 2, water) [lit.[10] mp 101–102 °C, $[\alpha]^{25}_D$ −45° (*c* 1.9, water)]; ^1H NMR ($CDCl_3$) δ 1.95 (s, 3 H), 2.06 (s, 3 H), 2.35 (s, 3 H), 3.11 (m, 2 H), 4.15 (m, 3 H), 6.90 (br, 1 H).

***N,O,S*-Triacetyl-D-cysteinol (9).** Compound **9** was prepared in the same manner as described for its enantiomer **7**: mp 96–97 °C, $[\alpha]^{25}_D$ +43.7° (*c* 2, water).

D-Serψ(CH$_2$NH)L-Leu (4a). Compound **7** (1.50 g, 6.44 mmol) was hydrolyzed in 60 mL of 1.7 N HCl at 90 °C under an atmosphere of nitrogen for 72 h. The solvent was removed under reduced pressure and residual water was removed by azeotropic distillation with absolute ethanol. The crude cysteinol hydrochloride (**2**) was dissolved in 20 mL of absolute ethanol containing NaOEt (17.34 mmol). To this solution was added **3a** (0.38 g, 4.46 mmol) in 5 mL of absolute ethanol. The reaction mixture was stirred for 72 h at room temperature followed by evaporation of the solvent. The residue was dissolved in water, acidified with 6 N HCl, and extracted with ether. The aqueous portion was neutralized with 2 N NaOH and diluted to 150 mL with deionized water. This solution was desalted on a 2 × 20 cm column of Dowex 1 × 8, according to the procedure of Drêze et al.[11] The ninhydrin-positive fractions were pooled and the solvent was removed under reduced pressure. The residue was crystallized from 47.5% ethanol to give 0.70 g (70%) of D-Serψ(CH$_2$NH)L-Leu (**4a**): mp 179–180 °C; $[\alpha]^{22}_D$ −53.7° (*c* 2.5, water); ^1H NMR (D_2O) δ 0.90 (d, 6 H), 1.60 (m, 3 H), 2.95 (m, 2 H), 3.48 (m, 2 H), 3.78 (m, 2 H); CI/CH_4 mass spectrum, (M + 1)/*e* (relative intensity) 222 (100), 205 (50), 204 (39), 159 (46), 149 (27), 117 (49). Anal. Calcd for $C_9H_{19}NO_3S$: C, 48.83; H, 8.67; N, 6.33; S, 14.48. Found: C, 48.68; H, 8.77; N, 6.33; S, 14.38. Compound **4a** elutes near the position of phenylalanine during automatic amino acid chromatography of the single column type (ninhydrin constant is 0.45 times that of leucine).

Compounds **1a** and **4b** were obtained in the same manner.

L-Serψ(CH$_2$NH)L-Leu (1a): mp 180–181 °C; $[\alpha]^{22}_D$ −5.4° (*c* 1.5, water); ^1H NMR (D_2O) δ 0.91 (d, 6 H), 1.62 (m, 3 H), 2.95 (m, 2 H), 3.50 (m, 2 H), 3.80 (m, 2 H); CI/CH_4 mass spectrum, (M + 1)/*e* (relative intensity) 222 (100), 205 (36), 204 (22), 159 (36), 149 (14), 117 (13). Compound **1a** elutes near the position of phenylalanine during automatic amino acid chromatography of the single column type (ninhydrin constant is 0.54 times that of leucine).

D-Serψ(CH$_2$NH)D-Phe (4b): mp 173–174 °C; $[\alpha]^{25}_D$ −13.3° (*c* 2, water); ^1H NMR (D_2O) δ 2.82 (m, 2 H), 3.13 (m, 2 H), 3.50 (m, 2 H), 3.78 (m, 2 H), 7.40 (s, 5 H); CI/CH_4 mass spectrum, (M + 1)/*e* (relative intensity) 256 (77), 238 (100), 193 (21), 175 (19), 149 (23), 133 (26), 105 (25). Anal. Calcd for $C_{12}H_{17}NO_3S$: C, 56.44; H, 6.72; N, 5.49; S, 12.55. Found: C, 56.19; H, 6.94; N, 5.53; S, 12.33. Compound **4b** elutes 17 min after phenylalanine during automatic amino acid analysis of the single column type (ninhydrin constant is 0.52 times that of leucine).

Boc-D-Serψ(CH$_2$NH)L-Leu (11). D-Serψ(CH$_2$NH)L-Leu (**4a**) was converted to **11** by standard procedures using di-*tert*-butyl dicarbonate:[4] mp 149–150 °C; $[\alpha]^{25}_D$ −88.6° (*c* 2, methanol); ^1H NMR ($CDCl_3$/Me_2SO-d_6) δ 0.90 (d, 6 H), 1.45 (s, 9 H), 1.69 (m, 3 H), 2.80 (m, 2 H), 3.34 (m, 1 H), 3.65 (m, 3 H), 5.62 (br, 1 H). Anal. Calcd for $C_{14}H_{27}NO_5S$: C, 52.32; H, 8.47; N, 4.36; S, 9.96. Found: C, 52.50; H, 8.63; N, 4.23; S, 9.80.

Boc-D-Serψ(CH$_2$NH)D-Phe (12). Compound **12** was prepared in the same manner as **11**: mp 120–121 °C; $[\alpha]^{25}_D$ +39.9° (*c* 2, methanol); ^1H NMR ($CDCl_3$) δ 1.42 (s, 9 H), 2.80 (m, 2 H), 3.10 (m, 2 H), 3.45 (m, 1 H), 3.65 (m, 2 H), 3.72 (m, 1 H), 5.25 (br, 1 H), 5.93 (br, 2 H), 7.23 (s, 5 H). Anal. Calcd for $C_{17}H_{25}NO_5S$: C, 57.44; H, 7.10; N, 3.94; S, 9.01. Found: C, 57.21; H, 7.18; N, 3.79; S, 9.14.

Acknowledgment. This work was supported by Contract HD-8-2830 from the Contraceptive Development Branch of the National Institute of Child Health and Human Development.

Registry No. **1a**, 76999-47-8; **2**, 77057-91-1; **3a**, 42990-28-3; **3b**, 35016-63-8; **4a**, 76999-48-9; **4b**, 76999-49-0; **5**, 77057-92-2; **6a**, 328-38-1; **6b**, 61-90-5; **7**, 76999-50-3; **8**, 52-90-4; **9**, 76999-51-4; **10**, 921-01-7; **11**, 76999-52-5; **12**, 76999-53-6.

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Synthesis of Angular Ring Methoxy-5-methylchrysenes and 5-Methylchrysenols[1]

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Received October 20, 1980

Methylated chrysenes contribute to the tumor initiation and complete carcinogenic activity of tobacco smoke and have also been detected in coal-liquefaction products.[2,3] Their concentration in the latter exceeds that of chrysene and is much higher than observed in tobacco smoke. Methylated chrysenes may also contribute to the mutagenicity of other energy-related materials.[4] Among the six possible monomethylchrysene isomers, 5-methylchrysene (**1**) is a potent carcinogen with activity approximately equal to that of benzo[*a*]pyrene. By comparison, chrysene is only weakly carcinogenic.[3,5]

The formation of vicinal dihydrodiol epoxides in the angular rings (positions 1–4 and 7–10 of **1**) of polynuclear aromatic hydrocarbons (PAH) appears to be a major activation process.[6-10] A related metabolic pathway, gen-

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