

coupling of **8** and glycine, methyl ester using 1-ethoxycarbonyl-2-ethoxy-1,2-dihydroquinoline [15] as a coupling agent gave methyl ester **9** which was further converted to

ethyl amide **10**. On the other hand, coupling of **8** and glycyl-*N*-methyl-L-phenylalaninamide [16] using the mixed anhydride coupling method [17] gave the heterocyclic tetrapeptide **11** in 42% yield. Finally, coupling of **8** and glycyl-L-phenylalanyl-*N*-ethyl-L-leucinamide afforded, in 62% yield, the pentapeptide **12** which represent a heterocyclic analog of the endogenous leucine-enkephalin.

EXPERIMENTAL

Melting points were determined in open capillaries on a Thomas Hoover apparatus and are uncorrected. Nuclear magnetic resonance spectra were recorded on Varian FT80A, EM390, and XL300 spectrometers. The chemical shifts are given in parts per million from tetramethylsilane as the internal standard. Mass spectra were obtained on a Finnigan 4600 mass spectrometer.

5,6-Dihydro-5,5-dimethyl-2-oxo-3-phenyl-1(2*H*)-pyrazine (**4**).

A solution of methyl phenylglyoxylate (24.0 g, 0.147 mole), 1,2-diamino-2-methylpropane (15.4 ml, 0.147 mole), and absolute ethanol (500 ml) was heated at reflux for 18 hours. The reaction was concentrated to approximately 250 ml and hot hexane (450 ml) was added. After several hours a beige precipitate (0.15 g) was removed by filtration and the filtrate was placed in the freezer. After 17 hours, 21.8 g (73%) of **4** was isolated as large colorless spars, mp 126-128°; ¹H nmr (deuteriochloroform): δ 1.33 (s, 6, geminal CH₃s), 3.22 (d, 2, CH₂, J = 3.8 Hz), 7.2-7.5 (m, 3, aromatic), 7.7-7.9 (m, 2, aromatic), 8.05 (broad s, 1, NH).

Anal. Calcd. for C₁₂H₁₄N₂O: C, 71.26; H, 6.98; N, 13.85. Found: C, 71.32; H, 6.87; N, 13.98.

5,6-Dihydro-5,5-dimethyl-2-oxo-3-phenyl-1(2*H*)-pyrazineacetic Acid, Methyl Ester (**6**).

To a stirred, room temperature solution of **4** (10.1 g, 0.050 mole) and dry tetrahydrofuran (80 ml) was added portionwise sodium hydride, 61% oil dispersion (1.32 g, 0.055 mole). The reaction was stirred 1 hour before methyl bromoacetate (4.6 ml, 0.055 mole) was added slowly *via* syringe. The reaction was stirred for 17 hours before being poured into water. The aqueous mixture was extracted several times with ether. The ethereal extracts were combined, washed with saturated aqueous sodium chloride, and dried over anhydrous sodium sulfate. The drying agent was removed by filtration and the filtrate was evaporated at reduced pressure affording an oil. Kugelrohr distillation at 165° (0.05 mm) gave 6.6 g (48%) of **6** as colorless prisms, mp 58-60°; ¹H nmr (deuteriochloroform): δ 1.37 (s, 6, geminal CH₃s), 3.41 (s, 2, ring CH₂), 3.72 (s, 3, ester CH₃), 4.20 (s, 2, glycine CH₂), 7.2-7.5 (m, 3, aromatic), 7.7-8.0 (m, 2, aromatic).

Anal. Calcd. for C₁₅H₁₈N₂O₃: C, 65.68; H, 6.61; N, 10.21. Found: C, 65.81; H, 6.63; N, 10.18.

N-Ethyl-5,6-dihydro-5,5-dimethyl-2-oxo-3-phenyl-1(2*H*)-pyrazineacetamide (**7**).

Methyl ester **6** (5.5 g, 0.020 mole) and anhydrous ethylamine (22.8 g, 0.506 mole) were stirred at room temperature in a sealed flask for 17 hours. The excess ethylamine was evaporated at reduced pressure leaving an oil which when triturated with ether gave a solid. Crystallization from toluene-hexane afforded 4.1 g (71%) of **7** as colorless needles, mp 113-115°; ¹H nmr (dimethylsulfoxide-*d*₆): δ 1.04 (t, 3, ethyl CH₃, J = 7.5 Hz), 1.28 (s, 6, geminal CH₃s), 3.12 (dq, 2, ethyl CH₂, J = 5.1 and 7.5 Hz), 3.47 (s, 2, ring CH₂), 4.06 (s, 2, glycine CH₂), 7.3-7.5 (m, 3, aromatic), 7.7-7.8 (m, 2, aromatic), 8.05 (t, 1, NH, J = 5.1 Hz).

Anal. Calcd. for C₁₆H₂₁N₃O₂: C, 66.88; H, 7.37; N, 14.62. Found: C, 67.01; H, 7.32; N, 14.63.

5,6-Dihydro-5,5-dimethyl-2-oxo-3-phenyl-1(2*H*)-pyrazineacetic Acid (**8**).

A mixture of methyl ester **6** (6.1 g, 0.022 mole), 1 molar aqueous lithi-

um hydroxide (24 ml, 0.024 mole), and methanol (50 ml) was stirred at room temperature for 2 hours. Most of the methanol was evaporated at reduced pressure and the aqueous concentrate was acidified by the addition of 1 molar hydrochloric acid (50 ml, 0.050 mole). The acidified mixture was extracted several times with ethyl acetate. The organic extracts were combined, washed with saturated aqueous sodium chloride, and dried over anhydrous sodium sulfate. The drying agent was removed by filtration and the filtrate was evaporated at reduced pressure leaving a solid. Crystallization from ethyl acetate-hexane gave 4.0 g (69%) of **8** as colorless irregular prisms, mp 159-167°; ¹H nmr (dimethylsulfoxide-*d*₆): δ 1.29 (s, 6, geminal CH₃s), 3.48 (s, 2, ring CH₂), 4.16 (s, 2, glycine CH₂), 7.2-7.5 (m, 3, aromatic), 7.6-7.9 (m, 2, aromatic).

Anal. Calcd. for C₁₄H₁₆N₂O₃: C, 64.60; H, 6.20; N, 10.76. Found: C, 64.42; H, 6.17; N, 10.57.

N-[(5,6-Dihydro-5,5-dimethyl-2-oxo-3-phenyl-1(2*H*)-pyrazinyl)acetyl]glycine, Methyl Ester (**9**).

Carboxylic acid **8** (6.8 g, 0.026 mole), glycine, methyl ester, hydrochloride (3.3 g, 0.026 mole), and 1-ethoxycarbonyl-2-ethoxy-1,2-dihydroquinoline (6.5 g, 0.026 mole) were stirred in dichloromethane (100 ml) for 17 hours. The reaction mixture was washed with 0.5 molar hydrochloric acid, saturated aqueous sodium bicarbonate, and saturated aqueous sodium chloride before being dried over anhydrous sodium sulfate. The drying agent was removed by filtration and the filtrate was evaporated at reduced pressure leaving an oil which slowly solidified. Crystallization from ethyl acetate-hexane afforded 5.4 g (62%) of **9** as colorless needles, mp 99-101°; ¹H nmr (dimethylsulfoxide-*d*₆): δ 1.27 (s, 6, geminal CH₃s), 3.43 (s, 2, ring CH₂), 3.59 (s, 3, ester CH₃), 3.87 (d, 2, 3-glycine CH₂, J = 6.0 Hz), 4.12 (s, 2, 2-glycine CH₂), 7.2-7.5 (m, 3, aromatic), 7.6-7.9 (m, 2, aromatic), 8.45 (t, 1, NH, J = 6.0 Hz).

Anal. Calcd. for C₁₇H₂₁N₃O₄: C, 61.62; H, 6.39; N, 12.68. Found: C, 61.55; H, 6.39; N, 12.56.

N-[(5,6-Dihydro-5,5-dimethyl-2-oxo-3-phenyl-1(2*H*)-pyrazinyl)acetyl]-*N*-ethylglycinamide (**10**).

Methyl ester **9** (3.4 g, 0.010 mole) and anhydrous ethylamine (17.5 g, 0.388 mole) were stirred at room temperature in a sealed flask for 7 hours. The excess ethylamine was evaporated at reduced pressure leaving a solid. Two crystallizations from ethyl acetate gave 1.9 g (54%) of **10** as colorless plates, mp 158-162°; ¹H nmr (dimethylsulfoxide-*d*₆): δ 1.01 (t, 3, ethyl CH₃, J = 7.5 Hz), 1.28 (s, 6, geminal CH₃s), 3.10 (dq, 2, ethyl CH₂, J = 5.3 and 7.5 Hz), 3.49 (s, 2, ring CH₂), 3.71 (d, 2, 3-glycine CH₂, J = 5.8 Hz), 4.15 (s, 2, 2-glycine CH₂), 7.3-7.5 (m, 3, aromatic), 7.7-7.8 (m, 2, aromatic), 7.84 (t, 1, ethyl amide NH, J = 5.3 Hz), 8.38 (t, 1, 3-glycine NH, J = 5.8 Hz).

Anal. Calcd. for C₁₈H₂₄N₄O₃: C, 62.77; H, 7.02; N, 16.27. Found: C, 62.46; H, 7.07; N, 16.24.

N-[(5,6-Dihydro-5,5-dimethyl-2-oxo-3-phenyl-1(2*H*)-pyrazinyl)acetyl]glycyl-*N*-methyl-L-phenylalaninamide (**11**).

To a stirred, -10° solution of carboxylic acid **8** (2.6 g, 0.010 mole) and dry tetrahydrofuran (30 ml) was added in turn triethylamine (1.5 ml, 0.011 mole) and isobutyl chloroformate (1.4 ml, 0.011 mole). After 12 minutes, a freshly neutralized solution of glycyl-*N*-methyl-L-phenylalaninamide, hydrochloride (3.0 g, 0.011 mole), triethylamine (2.8 ml, 0.020 mole), tetrahydrofuran (30 ml), and water (15 ml) was added in one portion. The cooling bath was removed and the reaction warmed to room temperature. After 17 hours, the reaction was poured into a separatory funnel containing 0.5 molar hydrochloric acid. The aqueous mixture was extracted two times with ether and two times with dichloromethane. The ethereal extracts were combined and washed with both saturated aqueous sodium bicarbonate and saturated aqueous sodium chloride. The dichloromethane extracts were combined and washed similarly. The ethereal and dichloromethane extracts were combined and dried over anhydrous sodium sulfate. The drying agent was removed by filtration and the filtrate was evaporated at reduced pressure leaving a solid. Crystallization from isopropanol gave 2.0 g (42%) of **11** as colorless irregular prisms, mp 194-204°; ¹H nmr (dimethylsulfoxide-*d*₆): δ 1.28 (s, 6, geminal

CH₃s), 2.57 (d, 3, amide CH₃, J = 4.4 Hz), 2.77 (dd, 1, phenylalanine CH₂, J_{vic} = 10.4 Hz, J_{gem} = 13.8 Hz), 3.00 (dd, 1, phenylalanine CH₂, J_{vic} = 4.8 Hz, J_{gem} = 13.8 Hz), 3.47 (s, 2, ring CH₂), 3.65 (dd, 1, 3-glycine CH₂, J_{vic} = 6.3 Hz, J_{gem} = 16.6 Hz), 3.82 (dd, 1, 3-glycine CH₂, J_{vic} = 6.0 Hz, J_{gem} = 16.6 Hz), 4.14 (s, 2, 2-glycine CH₂), 4.44 (m, 1, phenylalanine CH), 7.1-7.8 (m, 10, aromatic), 7.92 (q, 1, methyl amide NH, J = 4.4 Hz), 8.18 (d, 1, phenylalanine NH, J = 8.1 Hz), 8.35 (t, 1, 3-glycine NH, J = 6.2 Hz); ms: 477 (M⁺, 15).

Anal. Calcd. for C₂₆H₃₁N₅O₄: C, 65.39; H, 6.54; N, 14.66. Found: C, 65.30; H, 6.59; N, 14.39.

N-[(5,6-Dihydro-5,5-dimethyl-2-oxo-3-phenyl-1(2H)-pyrazinyl)acetyl]glycyl-L-phenylalanyl-N-ethyl-L-leucinamide (**12**).

To a stirred, room temperature mixture of carboxylic acid **8** (5.2 g, 0.020 mole, glycyl-L-phenylalanyl-N-ethyl-L-leucinamide, hydrochloride, hydrate (8.0 g, 0.020 mole), 1-ethoxycarbonyl-2-ethoxy-1,2-dihydroquinoline (5.0 g, 0.020 mole), and dichloromethane (300 ml) was added triethylamine (2.8 ml, 0.020 mole). After 17 hours, the reaction mixture was transferred to a separatory funnel where it was washed with 0.5 molar hydrochloric acid, saturated aqueous sodium bicarbonate, and saturated aqueous sodium chloride. After drying over anhydrous sodium sulfate, the dichloromethane was evaporated at reduced pressure leaving a foam which upon trituration with ether afforded a solid. Crystallization from isopropanol gave 7.5 g (62%) of **12** as colorless irregular prisms, mp 210-213°; ¹H nmr (dimethylsulfoxide-d₆): δ 0.82 (d, 3, leucine CH₃, J = 6.3 Hz), 0.87 (d, 3, leucine CH₃, J = 6.6 Hz), 1.00 (t, 3, ethyl CH₃, J = 6.8 Hz), 1.27 (s, 6, geminal CH₃s), 1.4-1.6 (m, 3, leucine CH, CH₂), 2.79 (dd, 1, phenylalanine CH₂, J_{vic} = 10.1 Hz, J_{gem} = 14.0 Hz), 3.0-3.1 (m, 3, phenylalanine CH₂, ethyl CH₂), 3.45 (s, 2, ring CH₂), 3.64 (dd, 1, 3-glycine CH₂, J_{vic} = 5.4 Hz, J_{gem} = 16.4 Hz), 3.81 (dd, 1, 3-glycine CH₂, J_{vic} = 5.4 Hz, J_{gem} = 16.4 Hz), 4.13 (s, 2, 2-glycine CH₂), 4.23 (m, 1, leucine CH), 4.54 (m, 1, phenylalanine CH), 7.1-7.5 (m, 10, aromatic), 7.71 (t, 1, ethyl amide NH, J = 4.8 Hz), 8.04 (d, 1, leucine NH, J = 8.7 Hz), 8.12 (d, 1, phenylalanine NH, J = 7.8 Hz), 8.40 (t, 1, 3-glycine NH, J = 5.4 Hz); ms: 604 (m⁺, 1).

Anal. Calcd. for C₃₃H₄₄N₆O₅: C, 65.54; H, 7.33; N, 13.90. Found: C, 65.46; H, 7.20; N, 13.58.

N-(t-Butoxycarbonyl)glycyl-L-phenylalanyl-N-ethyl-L-leucinamide (**13**).

N-(t-Butoxycarbonyl)glycyl-L-phenylalanyl-L-leucine, methyl ester [19] (20.6 g, 0.046 mole) and anhydrous ethylamine (60.0 g, 1.33 mole) were stirred at room temperature in a sealed flask. After 15 hours, the excess ethylamine was evaporated at reduced pressure leaving a foam which solidified when triturated with ether. Crystallization from ethyl acetate afforded 12.6 g (59%) of **13** as colorless irregular prisms, mp 183-185°; ¹H nmr (dimethylsulfoxide-d₆): δ 0.83 (d, 3, leucine CH₃, J = 5.4 Hz), 0.88 (d, 3, leucine CH₃, J = 6.9 Hz), 1.00 (t, 3, ethyl CH₃, J = 7.3 Hz), 1.36 (s, 9, t-boc CH₃s), 1.4-1.6 (m, 3, leucine CH, CH₂), 2.81 (dd, 1, phenylalanine CH₂, J_{vic} = 9.3 Hz, J_{gem} = 14.1 Hz), 2.9-3.1 (m, 3, phenylalanine CH₂, ethyl CH₂), 3.42 (dd, 1, glycine CH₂, J_{vic} = 5.7 Hz, J_{gem} = 16.7 Hz), 3.57 (dd, 1, glycine CH₂, J_{vic} = 5.7 Hz, J_{gem} = 16.7 Hz), 4.23 (dt, 1, leucine CH, J = 7.2 and 8.3 Hz), 4.52 (m, 1, phenylalanine CH), 7.01 (t, 1, glycine NH, J = 5.7 Hz), 7.1-7.3 (m, 5, aromatic), 7.71 (t, 1, ethyl amide NH, J = 5.4 Hz), 7.92 (d, 1, phenylalanine NH, J = 7.8 Hz), 8.03 (d, 1, leucine NH, J = 8.1 Hz).

Anal. Calcd. for C₂₄H₃₈N₄O₅: C, 62.32; H, 8.28; N, 12.11. Found: C, 62.00; H, 8.17; N, 12.13.

Glycyl-L-phenylalanyl-N-ethyl-L-leucinamide, Hydrochloride, Hydrate (**14**).

A stirred solution of protected tripeptide **13** (1.5 g, 0.0032 mole) and methanol (20 ml) was cooled to 0° in an ice bath. Gaseous hydrogen chloride was then bubbled through the solution for 30 minutes. The reaction was stirred an additional 30 minutes before being filtered. Evaporation of the filtrate and drying at high vacuum gave 1.2 g (92%) of **14** as a colorless powder, mp >210° dec; ¹H nmr (dimethylsulfoxide-d₆): δ 0.84 (d, 3, leucine CH₃, J = 5.7 Hz), 0.89 (d, 3, leucine CH₃, J = 5.4 Hz), 1.01 (t, 3, ethyl CH₃, J = 7.4 Hz), 1.3-1.7 (m, 3, leucine CH, CH₂), 2.78 (dd, 1, phenylalanine CH₂, J_{vic} = 8.7 Hz, J_{gem} = 13.7 Hz), 3.1 (m, 3, phenylalanine CH₂, ethyl CH₂), 3.4-3.6 (m, 2, glycine CH₂), 4.26 (m, 1, leucine CH), 4.62 (m, 1, phenylalanine CH), 7.1-7.4 (m, 5, aromatic), 7.98 (t, 1, ethyl amide NH, J = 5.1 Hz), 8.21 (broad s, 2, glycine NH₂), 8.43 (d, 1, leucine NH, J = 8.4 Hz), 8.74 (d, 1, phenylalanine NH, J = 8.4 Hz).

Anal. Calcd. for C₁₉H₃₀N₄O₄·HCl·0.75 H₂O: C, 55.33; H, 7.94; N, 13.58. Found: C, 55.39; H, 7.61; N, 13.64.

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