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Synthesis of Acyl Derivatives of Prolyl-leucinamides

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Abstract—Synthetic routes to prolyl-leucinamide, *N*-benzyl(prolyl)leucinamide, and *N*-(prolyl-leucyl)morpholine were proposed. Treatment of these compounds with benzoyl chloride or acetyl chloride in the presence of triethylamine afforded the corresponding (*N*-acylprolyl)leucinamides. Acyl derivatives of *N*-benzyl(prolyl)-leucinamide, and *N*-(prolyl-leucyl)morpholine were also synthesized by condensation of (*N*-benzoylprolyl)-leucine or (*N*-acetylprolyl)leucine with benzylamine or morpholine in the presence of *N*,*N*′-dicyclohexylcarbodiimide.

Keywords: *N*-*tert*-butyloxycarbonylprolyl-leucine methyl ester, prolyl-leucinamide, benzylamine, morpholine, acyl derivatives of prolyl-leucinamide.

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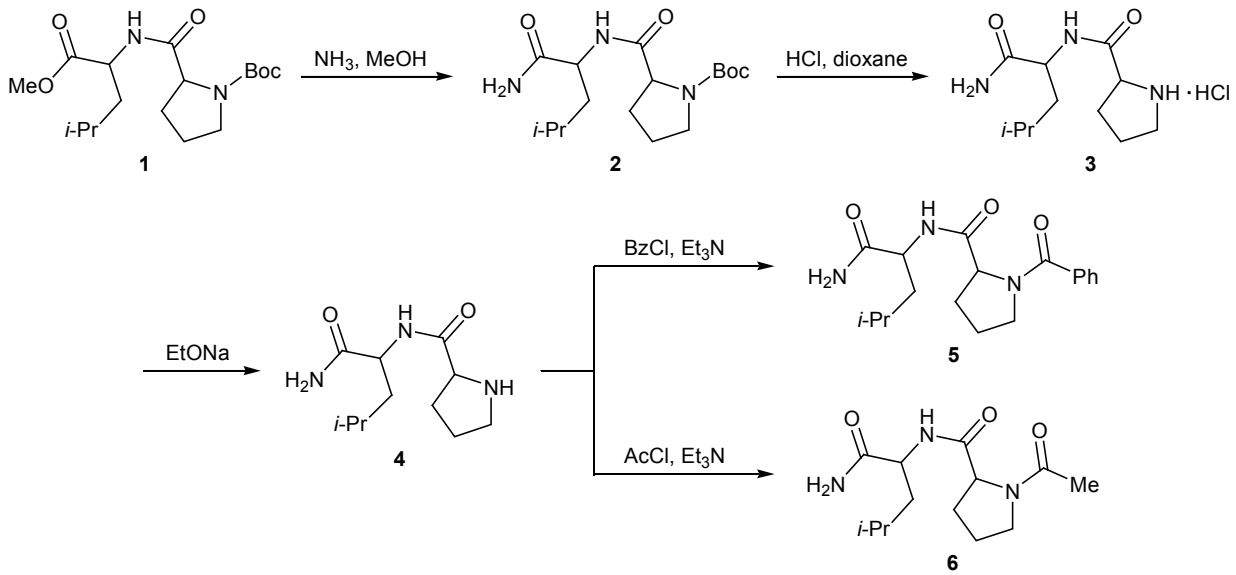
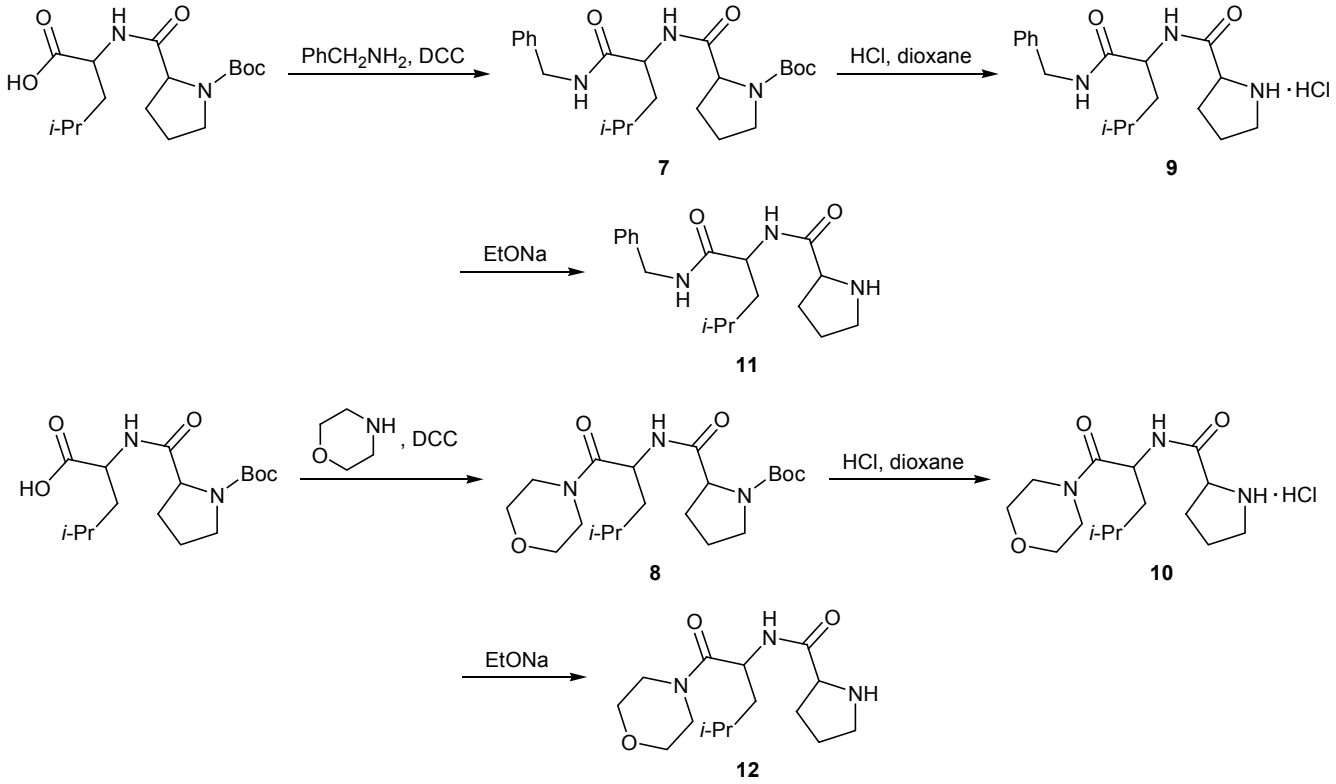
Peptides are biologically active compounds. Medicines with various therapeutic effects have been designed on the basis of peptides and are widely used in medical practice. There are published data [1–5] according to which some peptide derivatives such as amides, cyclic peptides, and acylpeptides also exhibit high physiological activity and selectivity, so that they can be used as drug substances and administered at minimum doses to avoid side effects. However, only a few examples of synthesis of acyl and amide derivatives of peptides have been reported so far. The goal of the present work was to develop methods of synthesis of prolyl-leucinamides and their acyl derivatives.

The starting compound was (*N*-*tert*-butoxycarbonylprolyl)leucine methyl ester (**1**) which was prepared by adding triethylamine to a cold (–15°C) mixture of leucine methyl ester hydrochloride and mixed anhydride obtained from *N*-*tert*-butoxycarbonylproline and isobutyl chloroformate and subjected to further transformations without isolation in the pure state. Treatment of **1** with a 6 N solution of ammonia in ethanol

Scheme 1.



Scheme 2.



gave (*N*-*tert*-butoxycarbonylprolyl)leucinamide **2**. To complete the ammonolysis process, the reaction mixture was kept for 5 days at room temperature. The Boc protecting group was removed from **2** by the action of hydrogen chloride in dioxane, and the subsequent treatment of hydrochloride **3** thus formed with an equimolar amount of sodium ethoxide afforded prolyl-leucinamide **4** (Scheme 1). Amide **4** reacted with benzoyl chloride and acetyl chloride in the presence of triethylamine to produce *N*-benzoylprolyl and *N*-acetylprolyl derivatives **5** and **6**, respectively.

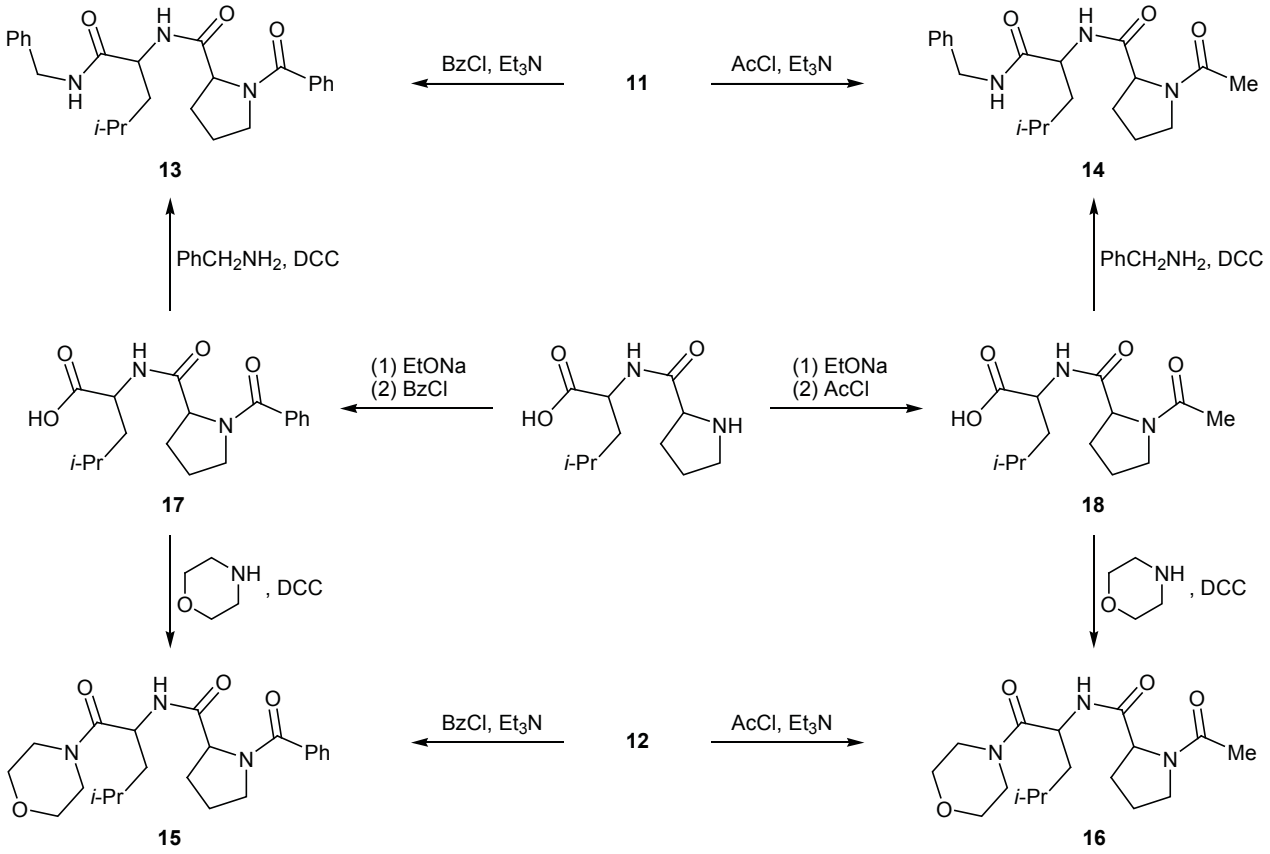
It should be noted that compound **1** failed to react with morpholine or benzylamine at room temperature, and the initial compound was recovered even after three weeks. Therefore, as starting material for the synthesis of N-substituted prolyl-leucinamides we used (*N*-*tert*-butoxycarbonylprolyl)leucine which was prepared according to the procedure developed by us previously [6].

The condensations of (*N*-*tert*-butoxycarbonylprolyl)leucine with benzylamine and morpholine in the presence of *N*,*N*′-dicyclohexylcarbodiimide (DCC) afforded *N*-benzyl(*N*-*tert*-butoxycarbonylprolyl)leucinamide **7** and 4-[(*N*-*tert*-butoxycarbonylprolyl)leucyl]-morpholine **8**, respectively. Compounds **7** and **8** were deprotected by treatment with a solution of hydrogen chloride in dioxane, and hydrochlorides **9** and **10** were converted to *N*-benzyl(prolyl)leucinamide **11** and 4-(prolyl-leucyl)morpholine **12** by the action of an equimolar amount of sodium ethoxide (Scheme 2).

The acylation of **11** and **12** with benzoyl chloride or acetyl chloride in the presence of triethylamine gave *N*-benzyl(*N*-benzoylprolyl)leucinamide **13**, (*N*-acetylprolyl)-*N*-benzylleucinamide **14**, 4-(*N*-benzoylprolyl)-leucyl)morpholine **15**, and 4-[(*N*-acetylprolyl)leucyl]-morpholine **16** (Scheme 3). Compounds **13**–**16** were also synthesized independently starting from prolyl-leucine which was obtained as described in [7]. Treatment of a solution of prolyl-leucine sodium salt (prepared by reaction of prolyl-leucine with sodium ethoxide) with benzoyl chloride or acetyl chloride afforded *N*-acyl derivatives **17** and **18**, respectively, and the condensations of **17** and **18** with benzylamine and morpholine in the presence of DCC produced target N-substituted prolyl-leucinamides **13**–**16**.

The structure of the synthesized compounds was confirmed by IR and NMR spectra and elemental analyses. It should be noted that some signals in the ^{1}H and ^{13}C NMR spectra of compounds containing a *tert*-butoxycarbonyl or acyl group were doubled,

Scheme 3.



presumably due to the presence of stereoisomers. Furthermore, hydrochlorides **3**, **9**, and **10** are extremely hygroscopic, which made it difficult to obtain their accurate elemental analyses.

EXPERIMENTAL

All reactions were carried out using anhydrous organic solvents. Leucine methyl ester hydrochloride [8] and *N*-*tert*-butoxycarbonylproline [9] were synthesized by known methods. The IR spectra were recorded in KBr on a Nicolet Protégé-460 spectrometer with Fourier transform. The ^{1}H and ^{13}C NMR spectra were measured with a Bruker Avance-500 spectrometer; the chemical shifts were determined relative to the residual proton and carbon signals of the deuterated solvent. The optical rotations were measured on an ATAGO AP-300 polarimeter.

***tert*-Butyl (2*S*)-2-{[(2*S*)-1-methoxy-4-methyl-1-oxopentan-2-yl]carbamoyl}pyrrolidine-1-carboxylate (1).** Triethylamine, 10.12 g (100 mmol), was added to a solution of 21.53 g (100 mmol) of *N*-*tert*-butoxycarbonylproline in 250 mL of methylene chloride, the mixture was cooled to –15°C, and 13.66 g (100 mmol) of isobutyl chloroformate was added dropwise. The mixture was stirred for 20 min at –15°C, a suspension of 19.07 g (105 mmol) of leucine methyl ester hydrochloride in 100 mL of methylene chloride, cooled to –15°C, was added, and 10.63 g (105 mmol) of cold triethylamine was added dropwise, maintaining the temperature at –15±1°C. The mixture was then stirred at –12±2°C for 3 h, allowed to slowly warm up to room temperature, and left overnight. The mixture was washed in succession with 0.8 N aqueous HCl (3×100 mL), water (2×100 mL), a saturated solution of sodium hydrogen carbonate (3×100 mL), and water again (3×100 mL) and dried over sodium sulfate. The resulting solution was filtered, the solvent was removed under reduced pressure, and the residue was extracted with diethyl ether. The extract was filtered and concentrated to a volume of 50 mL under reduced pressure. The product was precipitated with 200 mL of cold hexane, filtered off, washed with hexane, and dried under reduced pressure. Yield 23.97 g (70%), mp 77–78°C, $[\alpha]_D^{20}$ = –75.3° (*c* = 3, MeOH); published data [7]: mp 78–80°C, $[\alpha]_D^{20}$ = –75.9° (*c* = 3, MeOH) [7]. IR spectrum, ν, cm^{-1}: 1741, 1698, 1652 (C=O), 1560

(δNH). Found, %: C 59.81; H 8.98; N 8.05. $C_{17}H_{30}N_2O_5$. Calculated, %: C 59.63; H 8.83; N 8.18.

***tert*-Butyl (2*S*)-2-{[(2*S*)-1-amino-4-methyl-1-oxopentan-2-yl]carbamoyl}pyrrolidine-1-carboxylate (2).** A solution of 22.65 g (65 mmol) of **1** in 100 mL of a 6 N solution of ammonia in methanol was kept in a hermetically closed flask for 5 days at room temperature. The mixture was filtered, the solvent was removed under reduced pressure, and the residue was extracted with THF. The extract was filtered and concentrated until crystals began to separate, 100 mL of diethyl ether–hexane (1:1) was added, the mixture was kept for 12 h at 5°C, and the precipitate was filtered off, washed with hexane, and dried under reduced pressure. Yield 18.30 g (86%), mp 157–159°C, $[\alpha]_D^{20}$ = –79.2° (*c* = 2, MeOH). IR spectrum, ν, cm^{-1}: 1701, 1677, 1600 (C=O), 1549 (δNH). ^1H NMR spectrum (CD_3OD), δ, ppm: 0.87–0.98 m (6H), 1.41 s and 1.49 s (9H), 1.52–1.74 m (3H), 1.82–2.0 m (3H), 2.14–2.21 m (1H), 3.36–3.44 m (1H), 3.46–3.54 m (1H), 4.18–4.24 m (1H), 4.38–4.47 m (1H). ^{13}C NMR spectrum, δ_C, ppm: 21.75, 22.09, 23.50, 23.64, 24.47, 25.47, 25.86, 25.91, 28.67, 28.73, 31.17, 32.45, 41.43, 42.29, 47.89, 48.21, 52.61, 61.39, 61.74, 81.35, 155.92, 156.73, 175.23, 175.37, 177.12, 177.52. Found, %: C 58.92; H 8.73; N 12.66. $C_{16}H_{29}N_3O_4$. Calculated, %: C 58.69; H 8.93; N 12.83.

(2*S*)-*N*-[(2*S*)-1-Amino-4-methyl-1-oxopentan-2-yl]pyrrolidine-2-carboxamide hydrochloride (3). Amide **2**, 16.37 g (50 mmol), was dissolved in 70 mL of dioxane, 50 mL of a 6.2 N solution of hydrogen chloride in dioxane was added, and the mixture was stirred for 2 h and concentrated to a volume of 50 mL under reduced pressure. The product was precipitated with 200 mL of diethyl ether, filtered off, washed twice with diethyl ether, and dried under reduced pressure at 45°C until constant weight. Yield 11.60 g (88%), mp 145–148°C, $[\alpha]_D^{20}$ = –74.7° (*c* = 2.6, MeOH). IR spectrum, ν, cm^{-1}: 1691, 1677 (C=O), 1549 (δNH). ^1H NMR spectrum (D_2O), δ, ppm: 0.78 d (3H, *J* = 6.5 Hz), 0.82 d (3H, *J* = 6.5 Hz), 1.42–1.50 m (1H), 1.52–1.63 m (2H), 1.87–2.02 m (3H), 2.33–2.45 m (1H), 3.25–3.36 m (2H), 4.16–4.23 m (1H), 4.28–4.35 m (1H). ^{13}C NMR spectrum, δ_C, ppm: 20.78, 22.18, 23.74, 24.34, 29.82, 39.73, 46.56, 52.66, 59.48, 169.50, 176.89. Found, %: C 49.77; H 8.25; Cl 13.68; N 15.69. $C_{11}H_{21}N_3O_2 \cdot HCl$. Calculated, %: C 50.09; H 8.41; Cl 13.44; N 15.93.

(2*S*)-*N*-[(2*S*)-1-Amino-4-methyl-1-oxopentan-2-yl]pyrrolidine-2-carboxamide (4). A solution of 10.55 g (40 mmol) of hydrochloride **3** in 75 mL of ethanol was added with vigorous stirring to a solution of sodium ethoxide prepared from 0.92 g (40 mmol) of sodium and 20 mL of ethanol. The mixture was stirred for 2 h and filtered, the solvent was removed from the filtrate under reduced pressure, and the residue was extracted with THF. The extract was filtered, and the filtrate was evaporated under reduced pressure until a solid began to precipitate. The product was precipitated with 150 mL of diethyl ether, filtered off, washed with diethyl ether, and dried under reduced pressure. Yield 8 g (88%), mp 148–150°C, $[\alpha]_D^{20}$ = –58.7° (*c* = 2, MeOH). IR spectrum, ν, cm^{-1}: 1676, 1652 (C=O), 1516 (δNH). ^1H NMR spectrum (CD_3OD), δ, ppm: 0.92 d (3H, *J* = 6.5 Hz), 0.95 d (3H, *J* = 6.5 Hz), 1.54–1.67 m (3H), 1.68–1.80 m (3H), 2.08–2.16 m (1H), 2.90–3.01 m (2H), 3.66–3.72 m (1H), 4.38–4.43 m (1H). ^{13}C NMR spectrum, δ_C, ppm: 21.98, 23.52, 26.04, 26.98, 31.95, 42.44, 48.01, 52.45, 61.49, 176.95, 177.27. Found, %: C 58.27; H 9.43; N 18.61. $C_{11}H_{21}N_3O_2$. Calculated, %: C 58.12; H 9.31; N 18.49.

(2*S*)-*N*-[(2*S*)-1-Amino-4-methyl-1-oxopentan-2-yl]-1-benzoylpyrrolidine-2-carboxamide (5). Triethylamine, 1.62 g (16 mmol), was added to a solution of 3.41 g (15 mmol) of amide **4** in 50 mL of THF, and 2.25 g (16 mmol) of benzoyl chloride was added dropwise with vigorous stirring. The mixture was stirred for 2 h and concentrated to a volume of 20 mL, 100 mL of hexane was added, and the precipitate was filtered off, washed on a filter in succession with hexane (2×15 mL) and water (2×20 mL), and dried under reduced pressure at 45°C. The product was purified by reprecipitation from THF with diethyl ether–hexane (1:1). Yield 3.63 g (73%), mp 132–134°C, $[\alpha]_D^{20}$ = –113.7° (*c* = 1.5, MeOH). IR spectrum, ν, cm^{-1}: 1680, 1656, 1623 (C=O), 1543 (δNH). ^1H NMR spectrum (CD_3OD), δ, ppm: 0.8–1.0 m (6H), 1.17–1.30 m and 1.33–1.40 m (1H); 1.56–1.69 m, 1.70–1.91 m, and 1.92–2.07 m (5H); 2.25–2.36 m (1H); 3.49–3.58 m, 2.60–3.69 m, and 3.70–3.82 m (2H); 4.18–4.24 m, 4.44–4.52 m, and 4.57–4.63 m (2H); 7.38–7.82 m (5H). ^{13}C NMR spectrum, δ_C, ppm: 21.96, 23.65, 23.76, 25.46, 25.90, 26.36, 30.94, 33.23, 41.85, 42.07, 48.54, 51.72, 51.85, 52.91, 62.05, 63.37, 127.83, 128.33, 129.44, 131.07, 131.54, 137.32, 138.05, 172.14, 172.85, 174.48, 174.66, 177.10, 177.52. Found, %: C 65.42; H 7.71; N 12.54. $C_{18}H_{25}N_3O_3$. Calculated, %: C 65.23; H 7.60; N 12.68.

(2*S*)-1-Acetyl-*N*-[(2*S*)-1-amino-4-methyl-1-oxopentan-2-yl]pyrrolidine-2-carboxamide (6) was synthesized in a similar way from 3.41 g (15 mmol) of **4** and 1.26 g (16 mmol) of acetyl chloride using 1.62 g

(16 mmol) of triethylamine. Yield 2.87 g (71%), mp 163–166°C, $[\alpha]_D^{20}$ = –98.3° (*c* = 1.5, MeOH). IR spectrum, ν, cm^{-1}: 1683, 1652, 1636 (C=O), 1544 (δNH). ^1H NMR spectrum (CD_3OD), δ, ppm: 0.90–1.01 m (6H), 1.60–1.75 m (3H), 1.85–1.93 m and 1.95–2.06 m (3H), 2.12 s (3H), 2.17–2.28 m and 2.31–2.39 m (1H); 3.45–3.53 m, 3.56–3.63 m, and 3.66–3.72 m (2H); 4.36–4.43 m and 4.47–4.53 m (2H). ^{13}C NMR spectrum, δ_C, ppm: 21.67, 22.16, 22.39, 23.53, 23.63, 23.85, 25.76, 25.94, 26.08, 31.03, 33.13, 41.45, 42.02, 48.09, 49.48, 52.67, 52.77, 61.59, 62.34, 172.32, 172.87, 174.22, 174.66, 177.08, 177.56. Found, %: C 58.14; H 8.75; N 15.46. $C_{13}H_{23}N_3O_3$. Calculated, %: C 57.97; H 8.61; N 15.60.

***tert*-Butyl (2*S*)-2-{[(2*S*)-1-(benzylamino)-4-methyl-1-oxopentan-2-yl]carbamoyl}pyrrolidine-1-carboxylate (7).** A solution of 21.63 g (105 mmol) of DCC in 200 mL of THF and 11.25 g (105 mmol) of benzylamine were added in succession to a solution of 32.84 g (100 mmol) of (2*S*)-2-{[(2*S*)-1-(*tert*-butoxycarbonyl)pyrrolidine-2-carbonyl]amino}-4-methylpentanoic acid. The mixture was stirred for 20 h, the precipitate was filtered off, the solvent was removed under reduced pressure, and the residue was washed with cold hexane. The product was extracted with diethyl ether, the extract was filtered and concentrated to a volume of 50 mL under reduced pressure, 200 mL of hexane was added, and the mixture was kept for 24 h at 3°C. The precipitate was filtered off, washed with cold hexane, dried under reduced pressure, and reprecipitated from diethyl ether with hexane. Yield 29.23 g (70%), mp 102–104°C, $[\alpha]_D^{20}$ = –69.8° (*c* = 2, MeOH). IR spectrum, ν, cm^{-1}: 1700, 1643 (C=O), 1555 (δNH). ^1H NMR spectrum ($CDCl_3$), δ, ppm: 0.83–0.94 m (6H), 1.23 s and 1.37 s (9H), 1.48–1.70 m (3H), 1.71–1.86 m (3H), 1.92–2.12 m (1H), 3.28–3.45 m (2H), 4.06–4.27 m (1H), 4.29–4.58 m (3H), 7.17–7.30 m (5H). ^{13}C NMR spectrum, δ_C, ppm: 21.68, 22.16, 23.02, 23.13, 24.67, 24.82, 25.14, 28.26, 31.14, 31.59, 40.64, 41.29, 43.32, 46.90, 47.25, 51.60, 51.91, 60.35, 60.86, 80.69, 127.15, 127.30, 127.59, 128.51, 138.03, 138.35, 154.53, 155.81, 172.07, 172.32, 173.0; 173.06. Found, %: C 66.34; H 8.65; N 9.87. $C_{23}H_{35}N_3O_4$. Calculated, %: C 66.16; H 8.45; N 10.06.

***tert*-Butyl (2*S*)-2-{[(2*S*)-4-methyl-1-(morpholin-4-yl)-1-oxopentan-2-yl]carbamoyl}pyrrolidine-1-carboxylate (8)** was synthesized in a similar way from 32.84 g (100 mmol) of (2*S*)-2-{[(2*S*)-1-(*tert*-butoxycarbonyl)pyrrolidine-2-carbonyl]amino}-4-methylpentanoic acid and 9.15 g (105 mmol) of morpholine using 21.63 g (105 mmol) of DCC. Yield 28.62 g (72%), mp 92–94°C, $[\alpha]_D^{20}$ = –78.7° (*c* = 2, MeOH). IR spectrum, ν, cm^{-1}: 1694, 1659, 1642 (C=O), 1543 (δNH). ^1H NMR spectrum (CD_3OD), δ, ppm: 0.90–0.99 m (6H), 1.43 s and 1.45 s (9H), 1.47–1.53 m and 1.55–1.64 m (2H), 1.67–1.78 m (1H), 1.80–1.99 m (3H), 2.09–2.28 m (1H), 3.34–3.42 m (1H), 3.44–3.59 m (3H), 3.60–3.75 m (6H), 4.20–4.28 m (1H), 4.83–4.91 m (1H). ^{13}C NMR spectrum, δ_C, ppm: 21.99, 22.19, 23.58, 24.42, 25.37, 25.74, 25.83, 28.65, 28,72, 31.09, 32.42, 41.88, 42.02, 43.67, 47.31, 47.85, 48.16, 48.43, 61.06, 61.21, 67.67, 81.04, 81.24, 155.85, 156.20, 172.49, 174.75, 175.21. Found, %: C 60.55; H 8.71; N 10.38. $C_{20}H_{35}N_3O_5$. Calculated, %: C 60.43; H 8.87; N 10.57.

(2*S*)-*N*-[(2*S*)-1-(Benzylamino)-4-methyl-1-oxopentan-2-yl]pyrrolidine-2-carboxamide hydrochloride (9). Compound **7**, 27.14 g (65 mmol), was dispersed in 30 mL of dioxane, 50 mL of a 6.2 N solution of hydrogen chloride in dioxane was added, and the mixture was stirred for 2 h. The product was precipitated by adding 250 mL of hexane, and the precipitate was filtered off, washed twice with diethyl ether, and dried under reduced pressure at 45°C until constant weight. Yield 18.21 g (83%), $[\alpha]_D^{20}$ = –42.4° (*c* = 2, MeOH). IR spectrum, ν, cm^{-1}: 1680, 1656 (C=O), 1549 (δNH). ^1H NMR spectrum (CD_3OD), δ, ppm: 0.92 d (3H, *J* = 6.5 Hz), 0.96 d (3H, *J* = 6.5 Hz), 1.54–1.78 m (3H), 1.87–2.05 m (3H), 2.38–2.49 m (1H), 3.31–3.43 m (2H), 4.30–4.48 m (4H). ^{13}C NMR spectrum, δ_C, ppm: 22.01, 23.32, 24.88, 25.79, 31.03, 41.75, 43.88, 48.41, 53.91, 60.78, 128.12, 128.36, 129.42, 139.77, 169.64, 174.18. Found, %: C 60.88; H 8.17; Cl 10.26; N 11.69. $C_{18}H_{27}N_3O_2 \cdot HCl$. Calculated, %: C 61.09; H 7.97; Cl 10.02; N 11.87.

(2*S*)-*N*-[(2*S*)-4-Methyl-1-(morpholin-4-yl)-1-oxopentan-2-yl]pyrrolidine-2-carboxamide hydrochloride (10) was synthesized in a similar way from 28.84 g (65 mmol) of **8**. Yield 18.66 g (86%), mp 168–170°C, $[\alpha]_D^{20}$ = –43.6° (*c* = 2, MeOH). IR spectrum, ν, cm^{-1}: 1683, 1635 (C=O), 1549 (δNH). ^1H NMR spectrum (CD_3OD), δ, ppm: 0.96 d (6H, *J* = 6.5 Hz), 1.44–1.52 m (1H), 1.59–1.68 m (1H), 1.70–1.78 m (1H), 1.95–2.10 m (3H), 2.44–2.53 m (1H), 3.32–3.44 m (2H), 3.45–3.52 m (1H), 3.52–3.59 m (1H), 3.60–3.72 m (6H), 4.37–4.44 m (1H), 4.82–4.90 m (1H). ^{13}C NMR spectrum, δ_C, ppm: 21.85, 23.55, 24.95, 25.87, 31.02, 41.44, 43.71, 47.21, 47.46, 49.39, 60.78, 67.62, 169.46, 172.15. Found, %: C 53.78; H 8.58; Cl 10.83; N 12.24. $C_{15}H_{28}ClN_3O_2$. Calculated, %: C 53.96; H 8.45; Cl 10.62; N 12.59.

(2*S*)-*N*-[(2*S*)-1-(Benzylamino)-4-methyl-1-oxopentan-2-yl]pyrrolidine-2-carboxamide (11). A solution of 14.16 g (40 mmol) of hydrochloride **9** in 50 mL of ethanol was added with vigorous stirring to a solution of sodium ethoxide prepared from 0.92 g (40 mmol) of sodium and 20 mL of ethanol. The mixture was stirred for 3 h and filtered, and the solvent was removed from the filtrate under reduced pressure. The residue was extracted with THF, the extract was filtered and concentrated to a volume of 20 mL under reduced pressure, 150 mL of hexane was added, and the precipitate was filtered off, washed with hexane, and dried under reduced pressure. The product was purified by reprecipitation from THF with hexane. Yield 9.27 g (73%), mp 76–78°C, $[\alpha]_D^{20}$ = –41.6° (*c* = 2, MeOH). IR spectrum, ν, cm^{-1}: 1656, 1637 (C=O), 1547 (δNH). ^{1}H NMR spectrum (CD_3OD), δ, ppm: 0.91 d (3H, *J* = 6.0 Hz), 0.94 d (3H, *J* = 6.0 Hz), 1.54–1.79 m (6H), 2.04–2.12 m (1H), 2.86–2.96 m (2H), 3.62–2.68 m (1H), 4.35 q (2H, *J* = 14 Hz), 4.43–4.48 m (1H), 7.20–7.31 m (5H). ^{13}C NMR spectrum, δ_C, ppm: 22.10, 23.42, 26.01, 27.01, 31.98, 42.39, 43.94, 48.01, 52.87, 61.45, 128.12, 128.37, 129.45, 139.73, 174.43, 177.09. Found, %: C 68.25; H 8.74; N 13.08. $C_{18}H_{27}N_3O_2$. Calculated, %: C 68.11; H 8.57; N 13.24.

(2*S*)-*N*-[(2*S*)-4-Methyl-1-(morpholin-4-yl)-1-oxopentan-2-yl]pyrrolidine-2-carboxamide (12) was synthesized in a similar way from 13.35 g (40 mmol) of hydrochloride **10** using 0.92 g (40 mmol) of sodium. Yield 8.92 g (75%), mp 90–93°C, $[\alpha]_D^{20}$ = –62.8° (*c* = 2, MeOH). IR spectrum, ν, cm^{-1}: 1660, 1632 (C=O), 1514 (δNH). ^{1}H NMR spectrum (CD_3OD), δ, ppm: 0.94 d (3H, *J* = 6.5 Hz), 0.95 d (3H, *J* = 6.5 Hz), 1.44–1.52 m (1H), 1.55–1.68 m (2H), 1.69–1.79 m (3H), 2.08–2.16 m (1H), 2.89–2.98 m (2H), 3.43–3.52 m (1H), 3.61–3.72 m (6H), 4.70–4.80 m (1H), 4.85–4.92 m (1H). ^{13}C NMR spectrum, δ_C, ppm: 22.08, 23.61, 25.98, 27.06, 32.09, 42.20, 43.65, 47.29, 48.05, 48.18, 61.42, 67.64, 172.45, 176.81. Found, %: C 60.72; H 9.24; N 14.02. $C_{15}H_{27}N_3O_2$. Calculated, %: C 60.58; H 9.15; N 14.13.

General procedure for the synthesis of acyl derivatives of N-substituted prolyl-leucinamides 13–16. *a*. Amide **11** or **12**, 10 mmol, was dissolved in 100 mL of THF, 1.11 g (11 mmol) of triethylamine was added, and 11 mmol of benzoyl chloride or acetyl chloride was then added. The mixture was stirred for 3 h and concentrated to a volume of 15 mL, 100 mL of hexane was added, and the precipitate was filtered off, washed in succession with hexane (2×20 mL) and water (2×20 mL), and dried under reduced pressure at room temperature. The products were purified by reprecipitation from THF with hexane.

(2*S*)-1-Benzoyl-*N*-[(2*S*)-1-(benzylamino)-4-methyl-1-oxopentan-2-yl]pyrrolidine-2-carboxamide (13) was synthesized from 3.17 g of amide **11** and 1.55 g of benzoyl chloride in the presence of 1.11 g of triethylamine. Yield 2.87 g (68%), mp 58–61°C, $[\alpha]_D^{20}$ = –137.1° (*c* = 2, MeOH). IR spectrum, ν, cm^{-1}: 1703, 1654, 1620 (C=O), 1545 (δNH). ^{1}H NMR spectrum (CD_3OD), δ, ppm: 0.80 d (*J* = 6.5 Hz), 0.85 d (*J* = 6.5 Hz), 0.93 d (*J* = 6.5 Hz), and 0.96 d (*J* = 6.5 Hz) (6H); 1.25–1.40 m (1H), 1.61–1.86 m (3H), 1.88–1.99 m (2H), 2.17–2.32 m (1H), 3.46–3.53 m and 3.59–3.79 m (2H), 4.19–4.59 m (4H); 7.18–7.29 m, 7.34–7.49 m, and 7.55 d (*J* = 8.2 Hz) (10H). ^{13}C NMR spectrum, δ_C, ppm: 22.01, 22.09, 23.40, 23.49, 23.67, 25.43, 25.89, 26.31, 30.91, 33.16, 41.65, 41.85, 43.88, 43.95, 48.47, 51.65, 52.97, 53.37, 62.02, 63.29, 127.75, 128.10, 128.31, 128.43, 129.38, 129.43, 130.59, 130.97, 131.49, 133.55, 137.25, 138.03, 139.79, 139.82, 172.01, 172.77, 174.12, 174.28, 174.46, 174.51. Found, %: C 71.41; H 7.58; N 9.74. $C_{25}H_{31}N_3O_3$. Calculated, %: C 71.23; H 7.41; N 9.97.

(2*S*)-1-Acetyl-*N*-[(2*S*)-1-(benzylamino)-4-methyl-1-oxopentan-2-yl]pyrrolidine-2-carboxamide (14) was synthesized from 3.17 g of amide **11** and 0.86 g of acetyl chloride in the presence of 1.11 g of triethylamine. Yield 2.52 g (70%), mp 69–72°C, $[\alpha]_D^{20}$ = –87.6° (*c* = 2, MeOH). IR spectrum, ν, cm^{-1}: 1687, 1659, 1621 (C=O), 1549 (δNH). ^{1}H NMR spectrum (CD_3OD), δ, ppm: 0.90–0.97 m (6H); 1.53–1.78 m, 1.88–1.93 m, 1.94–2.03 m, 1.97 s, 2.06, and 2.11–2.29 m (10H); 3.42–3.66 m (2H), 4.29–4.54 m (4H), 7.18–7.32 m (5H). ^{13}C NMR spectrum, δ_C, ppm: 21.87, 22.16, 22.36, 23.41, 23.51, 23.61, 23.77, 25.69, 25.89, 25.99, 30.93, 33.08, 41.40, 41.86, 43.90, 47.97, 49.38, 53.13, 53.30, 61.42, 62.19, 128.06, 128.13, 128.32, 128.38, 129.39, 129.46, 139.87, 172.26, 172.72, 174.19, 174.28, 174.54, 174.68. Found, %: C 67.04; H 8.35; N 11.46. $C_{20}H_{29}N_3O_3$. Calculated, %: C 66.83; H 8.13; N 11.69.

(2*S*)-1-Benzoyl-*N*-[(2*S*)-4-methyl-1-(morpholin-4-yl)-1-oxopentan-2-yl]pyrrolidine-2-carboxamide (15) was synthesized from 2.97 g of amide **12** and 1.55 g of benzoyl chloride in the presence of 1.11 g of triethylamine. Yield 2.89 g (72%), mp 157–159°C, $[\alpha]_D^{20}$ = –118.8° (*c* = 2, MeOH). IR spectrum, ν, cm^{-1}: 1685, 1643, 1600 (C=O), 1553 (δNH). ^{1}H NMR spectrum (CD_3OD), δ, ppm: 0.82–0.85 m and 0.95–0.99 m (6H); 1.08–1.38 m, 1.47–1.54 m, 1.62–1.72 m, 1.77–1.88 m, 1.91–2.05 m, and 2.23–2.36 m (7H); 3.38–

3.53 m (2H), 3.54–3.75 m (8H); 4.45–4.50 m, 4.56–4.65 m, and 4.89–4.95 m (2H); 7.33–7.48 m and 7.58–7.63 m (5H). ^{13}C NMR spectrum, δ_C, ppm: 21.83, 22.14, 23.56, 23.63, 25.42, 25.72, 26.29, 30.95, 33.16, 41.33, 41.91, 43.67, 47.19, 47.32, 48.64, 51.63, 61.52, 63.22, 67.66, 127.82, 128.19, 129.41, 130.97, 131.41, 137.44, 138.07, 171.83, 172.34, 172.60, 172.76, 173.94, 174.07. Found, %: C 65.88; H 7.62; N 10.36. $C_{22}H_{31}N_3O_4$. Calculated, %: C 65.81; H 7.78; N 10.47.

(2*S*)-1-Acetyl-*N*-[(2*S*)-4-methyl-1-(morpholin-4-yl)-1-oxopentan-2-yl]pyrrolidine-2-carboxamide (16) was synthesized from 2.97 g of amide **12** and 0.86 g of acetyl chloride in the presence of 1.11 g of triethylamine. Yield 2.41 g (71%), mp 115–117°C, $[\alpha]_D^{20}$ = –77.3° (*c* = 2, MeOH). IR spectrum, ν, cm^{-1}: 1644 br (C=O), 1557 (δ NH). ^{1}H NMR spectrum (CD_3OD), δ, ppm: 0.89–0.98 m (6H), 1.40–1.51 m (1H), 1.55–1.76 m (2H), 1.81–1.90 m (1H), 1.92 s and 2.08 s (3H), 1.94–2.05 m (2H), 2.14–2.22 m and 2.29–2.37 m (1H), 3.43–3.69 m (10H), 4.39–4.48 m (1H), 4.85–4.95 m (1H). ^{13}C NMR spectrum, δ_C, ppm: 22.33, 22.42, 23.76, 23.81, 24.03, 25.89, 25.92, 26.38, 31.16, 33.30, 42.16, 43.90, 47.53, 48.77, 49.03, 49.56, 60.97, 62.34, 67.88, 172.28, 172.45, 172.61, 172.74, 174.21, 174.38. Found, %: C 60.29; H 8.68; N 12.16. $C_{17}H_{29}N_3O_4$. Calculated, %: C 60.15; H 8.61; N 12.38.

b. Compound **17** or **18**, 20 mmol, was dissolved in 100 mL of THF, a solution of 4.12 g (20 mmol) of DCC in 50 mL of THF was added, the mixture was stirred for 15 min, and 22 mmol of benzylamine or morpholine was added. The mixture was stirred for 24 h and filtered, the filtrate was concentrated to a volume of 30 mL under reduced pressure, 150 mL of hexane was added, and precipitate was filtered off, washed first with hexane and then with water, and dried under reduced pressure. The product was purified by reprecipitation from THF with hexane. The IR and NMR spectra of the isolated compounds were identical to those given above. Yield: **13**, 6.15 g (73%); **14**, 5.18 g (72%); **15**, 6.02 g (75%); **16**, 4.75 g (70%).

(2*S*)-2-{[(2*S*)-1-Benzoylpyrrolidine-2-carbonyl]-amino}-4-methylpentanoic acid (17). Prolyl-leucine, 11.41 g (50 mmol), was added to a solution of sodium ethoxide prepared from 1.15 g (50 mmol) of sodium and 150 mL of ethanol. The mixture was stirred until the mixture became homogeneous, and 7.03 g (50 mmol) of benzoyl chloride was added dropwise. The mixture was stirred for 48 h and filtered, the solvent was removed from the filtrate under reduced pressure, and the residue was extracted with THF. The extract was filtered and concentrated to a volume of 30 mL under reduced pressure, 100 mL of hexane was added, and the precipitate was filtered off, washed with hexane, and dried under reduced pressure. The product was reprecipitated from THF with diethyl ether–hexane (1:3). Yield 13.62 g (82%), mp 176–178°C, $[\alpha]_D^{20}$ = –109.3° (*c* = 2, MeOH). IR spectrum, ν, cm^{-1}: 1744, 1671, 1619 (C=O), 1557 (δ NH). ^{1}H NMR spectrum (CD_3OD), δ, ppm: 0.85–0.98 m (6H), 1.20–1.29 m and 1.37–1.50 m (1H); 1.62–1.71 m, 1.79–1.90 m, and 1.93–2.10 m (5H); 2.24–2.35 m (1H); 3.45–3.51 m, 3.58–3.64 m, 3.67–3.73 m, and 3.74–3.80 m (2H); 4.20–4.24 m, 4.43–4.50 m, and 4.60–4.68 m (2H); 7.36–7.49 m (4H), 7.58 d (1H, *J* = 9.5 Hz). ^{13}C NMR spectrum, δ_C, ppm: 21.62, 21.92, 23.48, 23.60, 25.49, 25.84, 26.22, 30.85, 33.02, 41.08, 41.62, 48.47, 51.64, 51.75, 52.13, 61.47, 63.46, 127.79, 128.16, 129.42, 129.47, 130.99, 131.41, 137.41, 137.47, 171.87, 172.89, 174.38, 174.63, 175.47, 175.83. Found, %: C 65.25; H 7.41; N 8.26. $C_{18}H_{24}N_2O_4$. Calculated, %: C 65.04; H 7.28; N 8.43.

(2*S*)-2-{[(2*S*)-1-Acetylpyrrolidine-2-carbonyl]-amino}-4-methylpentanoic acid (18) was synthesized in a similar way from 11.41 g (50 mmol) of prolyl-leucine and 3.93 g (50 mmol) of acetyl chloride using 1.15 g (50 mmol) of sodium. Yield 10.81 g (80%), mp 150–152°C, $[\alpha]_D^{20}$ = –77.9° (*c* = 2, MeOH). IR spectrum, ν, cm^{-1}: 1737, 1663, 1599 (C=O), 1558 (δ NH). ^{1}H NMR spectrum (CD_3OD), δ, ppm: 0.91–0.97 m (6H); 1.58–1.70 m, 1.72–1.82 m, 1.85–1.97 m, 1.98 s, 2.0–2.07 m, 2.08 s, 2.10–2.21 m, and 2.30–2.38 m (10H); 3.44–3.51 m and 3.52–3.70 m (2H), 4.37–4.50 m (2H). ^{13}C NMR spectrum, δ_C, ppm: 21.55, 21.91, 22.17, 22.21, 23.51, 23.81, 25.61, 25.84, 26.20, 30.86, 33.04, 41.28, 41.79, 48.02, 49.37, 52.29, 52.34, 60.94, 62.51, 172.13, 172.48, 174.31, 174.49, 175.99, 176.27. Found, %: C 57.92; H 8.43; N 10.14. $C_{13}H_{22}N_2O_4$. Calculated, %: C 57.76; H 8.20; N 10.36.

CONFLICT OF INTEREST

The authors declare the absence of conflict of interest.

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