The Rearrangements of 2-Amino-*N*-1*H*-pyrazolo[3,4-*d*]pyrimidin-4-ylacetamide and 2-Amino-*N*-4-pyrimidinylacetamide †

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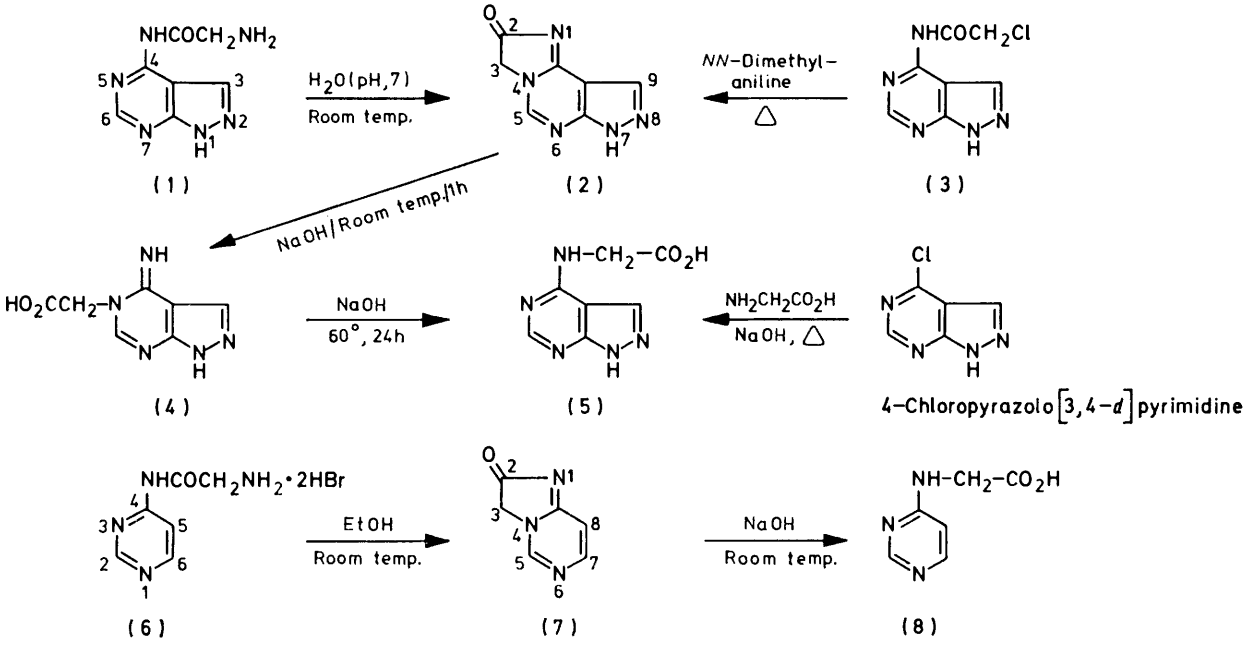
2-Amino-*N*-1*H*-pyrazolo[3,4-*d*]pyrimidin-4-ylacetamide (1) and 2-amino-*N*-4-pyrimidinylacetamide (6) undergo rearrangement to 3*H*-imidazo[1,2-*c*]pyrazolo[4,3-*e*]pyrimidin-2(7*H*)-one (2) and imidazo[1,2-*c*]pyrimidin-2(3*H*)-one (7), respectively, which in turn rearrange readily to *N*-1*H*-pyrazolo[3,4-*d*]pyrimidin-4-ylglycine (5) and *N*-4-pyrimidinylglycine (8), respectively.

THE rearrangement of 2-amino-*N*-purin-6-ylacetamide to *N*-purin-6-ylglycine has been reported.[1,2] This rearrangement occurs through the ring-opening of the pyrimidine portion of the purine followed by incorporation of the nitrogen of the amino acid as N^1 of purine in the resulting tricyclic intermediate. In this process the original N^1 of adenine is lost. The cyclic intermediate undergoes a further ring-opening followed by a Dimroth rearrangement [3] leading to the final *N*-purin-6-ylglycine. The present paper describes the chemistry and rearrangement of 2-amino-*N*-1*H*-pyrazolo[3,4-*d*]pyrimidin-4-ylacetamide and 2-amino-*N*-4-pyrimidinylacetamide into *N*-1*H*-pyrazolo[3,4-*d*]pyrimidin-4-ylglycine and *N*-4-pyrimidinylglycine respectively.

In the pyrazolo[3,4-*d*]pyrimidine series, the desired compound, 2-amino-*N*-1*H*-pyrazolo[3,4-*d*]pyrimidin-4-ylacetamide (1) was prepared by condensation of 4-aminopyrazolo[3,4-*d*]pyrimidine with excess of *N*-benzyloxycarbonylglycine *p*-nitrophenyl ester to give benzyl [(1*H*-pyrazolo[3,4-*d*]pyrimidin-4-ylcarbamoyl)methyl]-carbamate. Treatment of the latter compound with a 30% solution of hydrogen bromide in acetic acid gave 2-amino-*N*-1*H*-pyrazolo[3,4-*d*]pyrimidin-4-ylacetamide dihydrobromide (1·HBr).

In neutral aqueous solution, compound (1) eliminated the elements of ammonia and rearranged to the cyclic intermediate (2). The structure of (2) was confirmed by an independent synthesis. Condensation of 4-aminopyrazolo[3,4-*d*]pyrimidine with chloroacetic anhydride in toluene afforded 2-chloro-*N*[1(2)-(chloroacetyl)-1(2)*H*-pyrazolo[3,4-*d*]pyrimidin-4-yl]acetamide, which when refluxed in ethanol gave the monochloro compound (3). Compound (3) when refluxed in acetonitrile in the presence of *NN*-dimethylaniline for 24 h gave the cyclic intermediate (2). The criteria for establishing the identity were melting points, u.v., i.r., and mass spectra, and t.l.c. When a solution of (2) in 0.15N-NaOH was kept at room temperature, an intermediate, 1,4-dihydro-4-imino-5*H*-pyrazolo[3,4-*d*]pyrimidine-5-acetic acid (4) was obtained with only a trace of *N*-1*H*-pyrazolo[3,4-*d*]pyrimidin-4-ylglycine (5). However, when the above solution was heated at 60 °C for 25 h, the intermediate acid (4) rearranged smoothly to the final product (5).



(1) (2) (3) (4) (5) (6) (7) (8)

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The structure of (5) was confirmed by its synthesis from the reaction of 4-chloropyrazolo[3,4-*d*]pyrimidine and glycine.

In the pyrimidine series, 2-amino-*N*-pyrimidinylacetamide dihydrobromide (6) was prepared by the reaction of benzyl [(4-pyrimidinylcarbamoyl)methyl]-carbamate with 30% HBr in acetic acid. The latter compound was prepared by reaction of 4-aminopyrimidine with *N*-benzyloxycarbonylglycine *p*-nitrophenyl ester in dimethylformamide–dimethyl sulphoxide. Compound (6) on stirring in ethanol at room temperature gave the fluorescent cyclic intermediate (7) which in water underwent ring-opening and rearrangement to give the *N*-4-pyrimidinylglycine (8). This product appears to be formed through the intermediacy of 6-imino-1(6*H*)-pyrimidineacetic acid. Unlike the cyclic intermediate (2), compound (7) was rather unstable and gave 4-aminopyrimidine along with the rearrangement products.

The reactions in the rearrangement of compounds (1) and (6) to the glycine derivatives (5) and (8), respectively, appear to be similar to those involved in the rearrangement of N^6-glycyladenine and N^6-glycyl-9-methyladenine.[1,2]

EXPERIMENTAL

Melting points were taken on a Mel-Temp apparatus. I.r. spectra were determined on Perkin-Elmer 457 and 137 B spectrophotometers. N.m.r. spectra were measured for solutions in [$^2H_6$]DMSO on Varian XL-100 and A-60 spectrometers, with tetramethylsilane as internal standard. U.v. spectra were determined on Cary 14 and Beckman Acta V spectrophotometers. Mass spectra were recorded on a Dupont/CEC 21-491 double focusing spectrometer. T.l.c. was carried out on silica gel PF_{254} plates; solvents (v/v) were: (A) propan-2-ol–water–concentrated ammonium hydroxide (7 : 2 : 1); (B) ethyl acetate–1-propanol–water (4 : 1 : 2) (upper phase).

Benzyl [(1H-*Pyrazolo*[3,4-d]*pyrimidin*-4-*ylcarbamoyl*)-*methyl*]*carbamate*.—To a hot solution of 4-aminopyrazolo-[3,4-*d*]pyrimidine [4] (1.21 g, 9 mmol) in a mixture of dimethylformamide (20 ml) and dimethyl sulphoxide (20 ml) was added a solution of *N*-benzyloxycarbonylglycine *p*-nitrophenyl ester (5.94 g, 18 mmol) in dimethylformamide (13 ml) over 15 min. The solution was stirred at 90 °C for 4 h and then at room temperature for another 15 h. The solvent was removed *in vacuo*. The oily residue was triturated with ether (50 ml) and then filtered to isolate the solidified material. The crude product was crystallized three times from ethanol to give the *carbamate* as pale yellow crystals (804 mg, 27%); m.p. 216—218°; $\nu_{max.}$ (KBr) 3 316 (NH), 1 738 (C=O), and 1 700 cm^{-1} (NH–CO); $\lambda_{max.}$ (0.1NHCl, EtOH) 263 (ε 10 488), $\lambda_{max.}$ (EtOH) 264 (ε 10 420) (in 0.1N-NaOH the compound degrades to 4-aminopyrazolo[3,4-*d*]-pyrimidine); δ 3.25 (2 H, s, $NHCH_2$), 5.05 (2 H, s, OCH_2), 7.35 (5 H, s, aromatic), 8.4 (1 H, s, 3-H or 6-H), 8.45 (1 H, s, 6-H or 3-H); *m/e* 218 (M^+ — 108), 190, 161, 135, 119, 108, and 91 (Found: C, 55.05; H, 4.3; N, 25.75. $C_{15}H_{14}N_6O_3$ requires C, 55.2; H, 4.3; N, 25.75%).

2-*Amino*-N-1H-*pyrazolo*[3,4-d]*pyrimidin*-4-*ylacetamide Dihydrobromide* (1-HBr).—A solution of benzyl [(1*H*-pyrazolo[3,4-*d*]pyrimidin-4-ylcarbamoyl)methyl]carbamate (57 mg, 0.17 mmol) in HBr–acetic acid (30%; 1 ml) was stirred for 1 h at room temperature. To this solution was added ether (5 ml), and the mixture stirred for 10 min. The precipitate was filtered off and dried *in vacuo* to afford the *dihydrobromide* (1-HBr) as a yellow amorphous powder (57 mg, 95%); m.p. 210° (decomp.); $\nu_{max.}$ (KBr) 3 430 (NH_2), 3 000 (salt), and 1 722 cm^{-1} (C=O) (Found: C, 23.45; H, 3.1; N, 23.6. $C_7H_8N_6O{\cdot}2HBr$ requires C, 23.7; H, 2.8; N, 23.7%).

2-*Amino*-N-1H-*pyrazolo*[3,4-d]*pyrimidin*-4-*ylacetamide* (1).—To a slurry of (1-HBr) (100 mg, 0.28 mmol) in $CHCl_3$ (1.5 ml) was added purified triethylamine (0.14 ml). The mixture was stirred at 0 °C for 10 min and filtered. The residue was washed with ether (5 ml) and cold water (2 ml), and dried *in vacuo* to give the desired *product* (1) (37 mg, 62%); m.p. 180° (decomp.); $\nu_{max.}$ (KBr) 3 400—2 500 (NH_2, NH) and 1 700 cm^{-1} (C=O) (Found: C, 40.4; H, 4.5; N, 40.2. $C_7H_8N_6O{\cdot}H_2O$ requires C, 40.0; H, 4.75; 40.0%).

3,7-*Dihydroimidazo*[1,2-c]*pyrazolo*[4,3-e]*pyrimidin*-2-*one* (*Cyclic Intermediate*) (2). *Method A*.—To a solution of (1-HBr) (600 mg, 1.71 mmol) in water (20 ml) was added 1N-NaOH solution to bring the pH from 1.8 to 7.0. The solution was stirred for 15 min and then evaporated to dryness *in vacuo*. The residue was crystallized from ethanol–water to give the pale brown *cyclic intermediate* (2) (65 mg, 27%); m.p. 280° (decomp.); $\nu_{max.}$ (KBr) 3 000, 1 625 (C=O), 1 500, and 775 cm^{-1}; $\lambda_{max.}$ (H_2O) 287 (ε 12 057); δ 4.52 (2 H, s, CH_2 disappeared on adding D_2O), 8.2 (1 H, s, 5-H or 9-H), and 8.4 (1 H, s, 9-H or 5-H); *m/e* 175 (M^+), 161, 147, 135, 119, 108, 93, and 91 (Found: C, 46.0; H, 3.25; N, 38.15. $C_7H_5N_5O{\cdot}0.5H_2O$ requires C, 45.6; H, 3.25; N, 38.0%).

Conversion of 2-*Chloro*-N-1H-*pyrazolo*[3,4-d]*pyrimidin*-4-*ylacetamide* (3) *into the Cyclic Intermediate* (2). *Method B*.—A mixture of (3) (550 mg, 2.6 mmol) and *NN*-dimethylaniline (330 mg, 2.5 mmol) in acetonitrile (70 ml) was refluxed at 80 °C for 24 h. The mixture was cooled, filtered to remove unchanged (3), and the filtrate evaporated to dryness. The residue was triturated with acetonitrile (25 ml) and the crystalline material collected on a filter. The precipitate was extracted with hot toluene (3 × 30 ml) to remove unchanged (3). The remaining residue was crystallized from ethanol–water to give the pale brown crystalline *cyclic intermediate* (2) (65 mg, 12.4%); m.p. 280° (decomp.); i.r. spectrum identical with that prepared by method A; $\lambda_{max.}$ (H_2O) 287 (ε 13 128); *m/e* 175 (M^+), 161, 147, 135, 119, 93, and 91 (Found: C, 41.85; H, 4.20; N, 34.65. $C_7H_5N_5O{\cdot}1.5H_2O$ requires C, 41.5; H, 3.95: N, 34.65%.

2-*Chloro*-N-{1(2)-(*chloroacetyl*)-1(2)H-*pyrazolo*[3,4-d]-*pyrimidin*-4-*yl*}*acetamide*.—A mixture of 4-aminopyrazolo-[3,4-*d*]pyrimidine [4] (270 mg, 2 mmol) and chloroacetic anhydride (3.4 g, 20 mmol) in toluene (20 ml) was refluxed for 30 min and then allowed to cool at room temperature overnight. The pale yellow *bischloroacetyl compound* was collected on a filter (380 mg, 66%); m.p. 195—200°; $\nu_{max.}$ (KBr) 3 240, 3 180 (NH), and 1 740 cm^{-1} (C=O); $\lambda_{max.}$ (0.1N-HCl, EtOH) 260 (ε 9 360); $\lambda_{max.}$ (EtOH) 268 (ε 10 062); δ 4.35 (2 H, s, 4-$NCOCH_2$), 5.1 (2 H, s, 1- or 2-$NCOCH_2$), 8.4 (1 H, s, 3-H or 6-H), and 8.64 (1 H, s, 6-H or 3-H); *m/e* 289 (M + 1), 287 (M^+ — 1), 251, 211, 176, 175, 162, 147, 135, 119, 108, and 91 (Found: C, 36.0; H, 2.6; N, 24.55. $C_9H_7Cl_2N_5O_2$ requires C, 36.1; H, 2.45; N, 24.3%.

2-*Chloro*-N-1H-*pyrazolo*[3,4-d]*pyrimidin*-4-*ylacetamide* (3).—A mixture of 4-aminopyrazolo[3,4-*d*]pyrimidine [4] (675 mg, 5 mmol) and chloroacetic anhydride (2.27 g, 10 mmol) in toluene (50 ml) was refluxed for 1 h. The solvent was removed *in vacuo*. To the residue was added 95% ethanol (25 ml); the mixture was refluxed for 1 h, and the ethanol was removed *in vacuo*. The n.m.r. spectrum of the residue showed that the peak at 5.1 due to 1- or 2-$NCOCH_2Cl$ had disappeared. The residue was crystallized twice from toluene to give the white crystalline *monochloro-compound* (3), m.p. 161—175° (decomp.): $\nu_{max.}$ (KBr) 3 250 (NH), 1 740 (C=O), and 1 530 cm^{-1}; $\lambda_{max.}$ (0.1N-HCl) 260.5 nm (ε 11 632); δ 4.5 (2 H, s, 4-N-$COCH_2$), 8.3 (1 H, s, 3-H or 6-H), 8.5 (1 H, s, 6-H or 3-H); *m/e* 211 (M^+), 176, 175, 162, 147, 135, 119, 108, 93, and 91 (Found: C, 39.5; H, 2.85; N, 33.05. $C_7H_6ClN_5O$ requires C, 39.7; H, 2.85; N, 33.3%).

N-1H-*Pyrazolo*[3,4-d]*pyrimidin*-4-*ylglycine* (5). *Method A*.—To a suspension of 4-chloropyrazolo[3,4-*d*]pyrimidine [4] (307 mg, 2 mmol) and glycine (225 mg, 3 mmol) in water (12 ml) was added 1N-sodium hydroxide solution to bring the pH of the suspension from 3.6 to 9. The mixture was refluxed for 2.5 h; during this period the pH was checked and readjusted intermittently with 1N-sodium hydroxide solution. When the pH of the mixture remained constant (*ca.* 9) the mixture was filtered while hot. To the filtrate was added 95% ethanol (60 ml) and the solution was kept at 5 °C for 18 h. The resulting crystalline precipitate was collected on a filter and dried to give the sodium salt of (5). The sodium salt was then dissolved in water (6 ml) and the pH of the solution brought to 3.2 with 88% formic acid. The free-base glycine derivative (5) separated out (225 mg, 56%); m.p. 300° (decomp.) (lit.,[5] decomp. >215°): $\nu_{max.}$ (KBr) 3 400 (NH, CO_2H), and 1 675 cm^{-1} (C=O); $\lambda_{max.}$ (0.1N-HCl) 268 (ε 10 177); $\lambda_{max.}$ (0.1N-NaOH) 267 (ε 9 447); *m/e* 193 (M^+), 175, 149, 147, 135, 120, 119, 108, 93, and 91 (Found: C, 41.6; H, 3.8; N, 34.85. Calc. for $C_7H_7N_5O_2 \cdot 0.5H_2O$: C, 41.6; H, 3.95: N, 34.65%).

Rearrangement of the Cyclic Intermediate (2) *into* N-1H-*Pyrazolo*[3,4-d]*pyrimidin*-4-*ylglycine* (5). *Method B*.—A solution of the cyclic intermediate (2) (110 mg, 0.63 mmol) in aqueous sodium hydroxide (0.15N; 20 ml) was stirred at room temperature for 1 h. During this time the starting material disappeared (t.l.c.; solvent A) and the intermediate acid (4) (R_F 0.74) along with a trace amount of the glycine derivative (5) (R_F 0.80) began to appear. The solution was then heated at 60 °C for 25 h when t.l.c. (solvent A) showed conversion of the intermediate acid to the final product (5) to be complete.* A trace amount of 4-aminopyrazolo[3,4-*d*]pyrimidine was also formed. The solution was concentrated to *ca.* 10 ml and the pH (12.0) of this solution was brought to 2.8 by addition of 88% formic acid. The precipitate obtained after acidification was dissolved in aqueous sodium hydroxide (0.1N; 10 ml) and the solution filtered. The filtrate was reacidified with 88% formic acid and quickly filtered. The filtrate, after keeping at room temperature overnight, gave the glycine derivative (5) (70 mg, 54%), identical (i.r., u.v.) with that prepared by Method A, $\lambda_{max.}$ (0.1N-HCl) 267 (ε 9 990); $\lambda_{max.}$ (0.1N-NaOH) 267 (ε 10 015) (Found: C, 43.5: H, 3.6; N, 35.95. Calc. for $C_7H_7N_5O_2$: C, 43.35; H, 3.6; N, 36.2%).

Benzyl [(4-*Pyrimidinylcarbamoyl*)*methyl*]*carbamate*.—This was prepared from 4-aminopyrimidine using a procedure similar to that for the preparation of benzyl [(1*H*-pyrazolo-[3,4-*d*]pyrimidin-4-ylcarbamoyl)methyl]carbamate, yield 46%, m.p. 160—162°; $\nu_{max.}$ (KBr) 3 320, 1 715 (C=O), 1 685, and 1 580 cm^{-1}; $\lambda_{max.}$ (H_2O) 259.5 (ε 9 139); $\lambda_{max.}$ (0.1N-HCl) 265 (ε 15 232); $\lambda_{max.}$ (0.1N-NaOH) 282 (ε 7 819) (Found: C, 57.5; H, 4.75; N, 20.25. $C_{13}H_{12}N_4O_3$ requires C, 57.5; H, 4.45; N, 20.6%).

2-*Amino*-N-4-*pyrimidinylacetamide Dihydrobromide* (6).—A mixture of benzyl [4-pyrimidinylcarbamoyl)methyl]carbamate (60 mg, 0.22 mmol) in a 30% solution of HBr in acetic acid (1 ml) was stirred for 2 h. Sufficient ether was added to the solution for complete precipitation of the *dihydrobromide* (6) which was collected on a filter, and washed with ether (56 mg, 90%); m.p. 145° (decomp.); $\nu_{max.}$ (KBr) 3 050, 1 730 (C=O), 1 630, 1 575, and 1 190 cm^{-1} (Found: C, 23.1; H, 3.25; N, 17.75; Br, 49.95. $C_6H_8N_4O \cdot 2HBr$ requires C, 23.0; H, 3.2; N, 17.95; Br, 50.05%).

Imidazo[1,2-c]*pyrimidin*-2(3H)-*one* (*Cyclic Intermediate*) (7).—A mixture of the dihydrobromide (6) (310 mg, 0.98 mmol) in ethanol (500 ml) was stirred for 24 h, filtered, and the filtrate evaporated to dryness *in vacuo*. The residue was triturated with acetonitrile (50 ml). This solution was treated with charcoal and concentrated to give the cyclic intermediate (7) (46 mg, 19%); $\nu_{max.}$ (KBr) 3 500 and 1 625 cm^{-1}; *m/e* 135(M^+). Satisfactory analytical data for C and N could not be obtained while those for H and Br were in the acceptable range.

N-4-*Pyrimidinylglycine* (8).—A solution of the dihydrobromide (6) (500 mg, 1.6 mmol) in water (500 ml) was stirred at room temperature for 20 h and then evaporated to dryness. The residue was subjected to preparative t.l.c. (solvent B). The component having R_F 0.12 was eluted with ethanol. The ethanolic solution was evaporated to dryness and the residue treated with 0.1N-NaOH solution (4 ml) for 2 h. The solution was brought to pH 2.8 by addition of 88% HCO_2H and subjected to preparative t.l.c. (solvent A). The component with R_F 0.70 was eluted with ethanol. The ethanolic solution was evaporated to dryness and the residue crystallized from ethanol–water to give 42 mg (15%) of the *glycine derivative* (8), m.p. 250° (decomp.); $\nu_{max.}$ (KBr) 3 500 and 1 650 cm^{-1}; $\lambda_{max.}$ (0.1N-HCl) 257 (ε 16 832); *m/e* 153 (M^+), 135, 119, 107, and 95 (Found: C, 45.95; H, 4.55; N, 26.5. $C_6H_7N_3O_2 \cdot 0.25H_2O$ requires C, 45.65; H, 4.75; N, 26.6%).

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* The alkaline solution of an analogous compound, 1,5-dimethyl-4(5*H*)-iminopyrazolo[3,4-*d*]pyrimidine upon exposure to the atmosphere for 24 h rearranges to 1-methyl-4-methylaminopyrazolo[3,4-*d*]pyrimidine.[6]

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