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SYNTHESIS AND STUDY OF REACTIVITY OF PYRROLO[3,2-*e*][1,2,4]-TRIAZINE SYSTEM[1]

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Abstract - The corresponding 2-arylhydrazones **5** were obtained by coupling of diazonium salts with diethyl (3-methyl-1*H*-pyrrole)-2,4-dicarbamate **4**. These ones were cyclized in a alkaline medium to ethyl (2-aryl-5-methyl-3-oxo-2,3-dihydro-7*H*-pyrrolo[3,2-*e*][1,2,4]triazin-6-yl)carbamates **6** and tested for their stability. The starting carbamate **4** was prepared by a multistep synthesis from diethyl 3-methyl-1*H*-pyrrole-2,4-dicarboxylate **1**.

INTRODUCTION

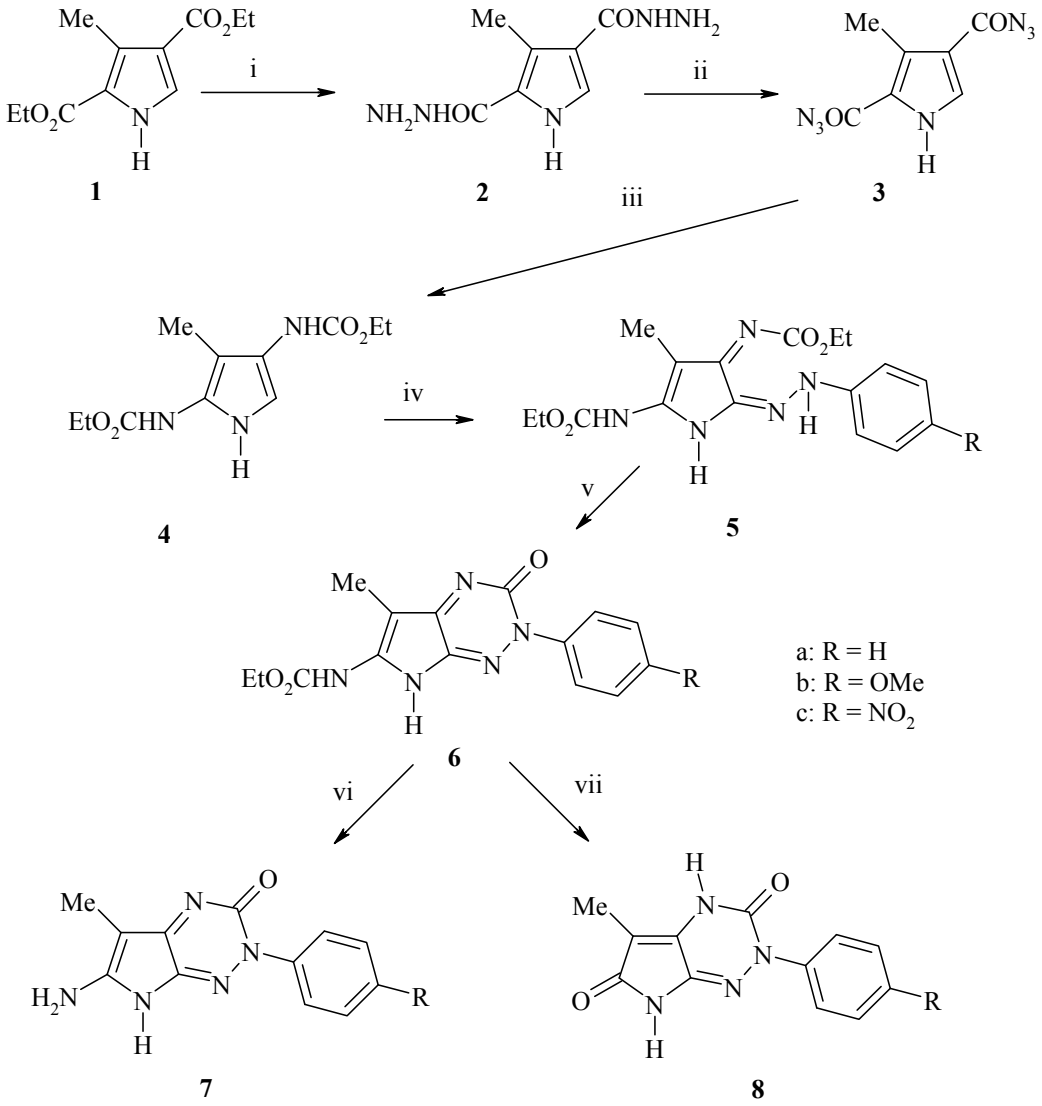
Both aza and deaza analogues of natural purine compounds are currently the object of interest mainly in connection with the research for substances with antineoplastic and virostatic activity. The pyrrolo-1,2,4-triazine systems also belong among substances of this kind. In some of them, the pyrrolo[2,1-*f*][1,2,4]triazine derivatives, an activity connected to protein kinase inhibitors[2-5] was discovered. Also some natural antibiotics (Toyocamycin, Tubercidin) containing pyrrolo[2,3-*d*]-pyrimidine (7-deazapurine) skeleton have a similar structure; they show tuberculostatic and antineoplastic activity.[6] Pyrrolo[3,2-*e*][1,2,4]triazine derivatives have been investigated only very little so far, which urged us to search for synthesis leading to this heterocyclic system.

RESULTS AND DISCUSSION

The syntheses of fused 1,2,4-triazines based on the cyclization of hydrazono-carbamates are advantageous in the cases when the starting compounds can be obtained by the azo-coupling reactions with five membered aromatic heterocyclic compounds, e.g. the synthesis of the pyrazolo[3,4-*e*][1,2,4]-triazines,[7] 1,2,4-triazinoindoles[8,9] or 1,2,4-triazines with fused furan[10,11] or thiophene ring.[12]

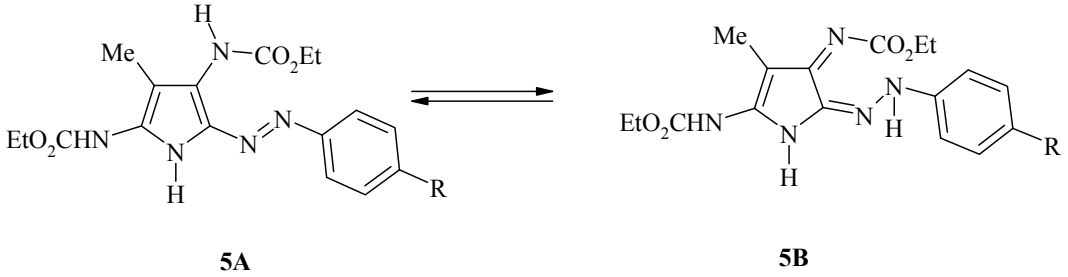
In this paper, we describe an application of this method to a pyrrole system in order to investigate an alternative route for the synthesis of little explored pyrrolo[3,2-*e*][1,2,4]triazine system.[13-15]

The starting compound for new syntheses was diethyl 3-methyl-1*H*-pyrrole-2,4-dicarboxylate[16] **1** which was converted by hydrazinolysis to the corresponding hydrazide **2**. The nitrosation in hydrochloric acid solution was the most efficient method for the transformation of hydrazide **2** to the azide **3**. The azide **3** (light sensitive compound, storage on day light causes dark colouration of product and its decomposition) was converted by Curtius rearrangement to diethyl (3-methyl-1*H*-pyrrole)-2,4-dicarbamate **4** in boiling anhydrous ethanol. This compound is highly unstable. It decomposes under wet conditions and when exposed to oxygen, resulting dark red polymer products. Slow decomposition occurs even when it is stored at –20 °C. The first signs of decompositions are visible within a few months.



Scheme 1. Reaction conditions. i) hydrazine hydrate, reflux. ii) $NaNO_2$, aqueous, HCl, 2-5 °C. iii) EtOH, reflux. iv) arene diazonium salt and pyridine. v) aqueous ethanolic Na_2CO_3, reflux; vi) aqueous NaOH, reflux. vii) aqueous HCl, reflux.

The corresponding 3-arylazocompounds **5A**, which are tautomeric with the hydrazones **5B** (Scheme 2), were prepared by a coupling reaction of diazonium salts with carbamate **4** in pyridine under inert conditions. The azo-hydrazone tautomerism with phenyl derivative **5a** was studied by IR spectroscopy in tetrachloromethane solution. For this purpose, compound **5a** containing labelled nitrogen was prepared from aniline-^{15}N. It was found that unlabelled derivative shows in the region N-H stretching vibrations an absorption maximum at 3130.4 cm^{-1} while the compound labelled with isotope ^{15}N has shifted the absorption maximum to 3123.3cm^{-1}. This shift is in accordance with the formula $\tilde{\upsilon} = 1/2\pi(K/\mu)^{1/2}$. In view of these facts we have concluded that the products of coupling reaction exists in tetrachloromethane solution in the hydrazono form **5B**. This hydrazono form was confirmed unambiguously by ^{15}N-NMR spectroscopy as well. The signal at –239.5 ppm of N-C_6H_5 fragment in the ^{15}N-NMR spectrum of ^{15}N selectively labelled compound shows that compound **5a** exists predominantly in hydrazono form (compared with ^{15}N chemical shifts of model hydrazo compounds[17]).



5A **5B**

Scheme 2. Azo-hydrazone tautomerism of compound **5**.

Cyclization reactions of hydrazones **5a**-**5c** leading to ethyl (2-aryl-5-methyl-3-oxo-2,3-dihydro-7*H*-pyrrolo[3,2-*e*][1,2,4]triazin-6-yl)carbamates (**6a**-**6c**) have been carrried out by heating in the alkaline medium of sodium carbonate. Under these conditions the cyclization reaction occurs within 5 minutes. However, the cyclization of the derivate **5c** occurred only after 30 min, due to the electron withdrawing the nitro group. Although it was detected that the compounds **5** are in the hydrazone form, which is favourable for the thermal cyclization (heating above the melting point), a deep decomposition occurred (note: under acid catalysis the cyclization has not been observed).

The question of stability of the created cycle of prepared compounds **6** has been paid to its hydrolytic cleavage. It was detected that these compounds are very stable both in the mineral acids medium and in the basic hydroxide solutions as well. In contrast to similar derivatives of pyrazolo[3,4-*e*][1,2,4]triazines,[18] 1,2,4-triazine[5,6-*b*]quinoline[19] or 1,2,4-triazine[4,5-*a*]benzimidazole,[20] where the cleavage of the 1,2,4-triazine ring occurs the compounds **6** undergo the hydrolysis of the ethoxycarbonyl group. Under alkaline hydrolysis the derivatives **7** containing the basic amino group, which is capable to form salts with mineral acids are produced. If acid hydrolysis is performed the

6-oxoderivatives **8** are yielded.

The prepared compounds were tested for biological activity. Human breast adenocarcinoma cell line *MCF7* was used for cytotoxicity determination by the MTT assay.[20,21] The tested compounds showed poor cytostatic activity (IC_{50} = 50–180 μmol/l), with the exception of the moderately active hydrazones **5a** (IC_{50} = 10.3 μmol/l), **5b** (IC_{50} = 14.6 μmol/l) and amino compounds **7a** (IC_{50} = 20.6 μmol/l), **7b** (IC_{50} = 26.8 μmol/l).

EXPERIMENTAL

Melting points were determined on a Boetius stage and are uncorrected. The IR spectra were recorded in KBr wafers on an ATI Unicam Genesis FTIR instrument. The NMR spectra were registered on a Bruker Avance 300 MHz DRX spectrometer; chemical shifts are reported in ppm, the coupling constants *J* in Hz. Elemental analyses were performed with an EA 1108 Elemental Analyser (Fison Instruments). Mass spectrometric experiments were performed using an LCQ ion trap mass spectrometer (Finnigan MAT, San Jose, CA, USA).

3-Methyl-1*H*-pyrrole-2,4-dicarbohydrazide (2): A mixture of ester **1**[16] (23.0 g, 0.102 mol) in 99% hydrazine monohydrate (80 mL) was refluxed under good stirring on oil bath for 3 h. During this time, from firstly formed solution start to precipitate white solid. To this suspension was added water (100 mL), hydrazide **2** was filtered off and washed with water. The sample for analysis was prepared by crystallization from EtOH. Yield: 19.4 g (96%), mp 285-290 °C (decomp). IR (cm^{-1}): 3304, 1632, 1598, 1501, 1388, 1295. ^{1}H-NMR, (DMSO-d_6): 2.41 (s, 3H, CH_3), 4.19-4.35 (bs, 4H, 2x NH_2), 7.29 (s, 1H, 5-H), 8.68 (s, 1H, NH), 8.93 (s, 1H, NH), 11.30 (s, 1H, NH). ^{13}C-NMR, (DMSO-d_6): 11.5, 117.5, 121.7, 122.7, 125.6, 162.3, 165.5. MS (APCI, *m/z*): 198.1 $[M+H]^+$. Anal. Calcd for $C_7H_{11}N_5O_2$ (197.2): C, 42.64; H, 5.62; N, 35.51. Found: C, 42.54; H, 5.39; N, 35.67.

3-Methyl-1*H*-pyrrole-2,4-dicarbonyl diazide (3): A stirred solution of hydrazide **2** (18.12 g, 91.88 mmol) in water (1200 mL) and 35% hydrochloric acid (36.2 mL) was nitrosated with a solution of sodium nitrite (12.68 g, 183.9 mmol) in water (75 mL) at 0-5 °C. The reaction mixture was stirred for next 3 h and allowed to stand overnight at 2-5 °C. Precipitated azide was collected on a filter, washed thoroughly with water and dried over phosphorus pentoxide in dark place. Yield: 19.3 g (96%), mp 145-150 ° (decomp). IR (cm^{-1}): 2145, 2140, 1670, 1663, 1478, 1234. ^{1}H-NMR, (DMSO-d_6): 2.53 (s, 3H, CH_3), 7.60 (s, 1H, 5-H), 11.00 (bs, 1H, NH). ^{13}C-NMR, (DMSO-d_6): 11.7, 117.4, 123.1, 131.3, 132.2, 164.3, 168.2. MS (APCI, *m/z*): 220.1 $[M+H]^+$. Anal. Calcd for $C_7H_5N_7O_2$ (219.2): C, 38.36; H, 2.30; N, 44.74. Found: C, 38.29; H, 2.44; N, 44.62.

Diethyl (3-methyl-1*H*-pyrrole)-2,4-dicarbamate (4): A solution of azide **3** (9.5 g, 43.3 mmol) in anhydrous EtOH (300 mL) was refluxed under nitrogen for 2.5 h. The formed dark red solution was concentrated in vacuo to oily product. This one was dissolved in hot mixture of EtOH (50 mL) and water (100 mL) and left to stand at 5 °C for 2 h. The precipitated dark-red gummy product was discarded, clear solution was diluted with water (100 mL) and allow to stand for 24 h at 2-5 °C. The solid product was filtered off and immediately dried in vacuo over phosphorus pentoxide. Yield: 5.32 g (48%), mp 68-72 °C. IR (cm^{-1}): 3323, 3313, 1750, 1745, 1264, 1207. ^{1}H-NMR, (DMSO-d_6): 1.18-1.28 (m, 6H, 2x CH_3), 1.77 (s, 3H, CH_3), 4.04-4.18 (m, 4H, 2x CH_2), 6.47 (s, 1H, 5-H), 8.41 (s, 1H, NH), 8.73 (s, 1H, NH), 10.26 (s, 1H, NH). ^{13}C-NMR, (DMSO-d_6): 10.5, 14.2, 14.8, 62.3, 62.9, 110.4, 116.3, 135.3, 140.1, 164.3, 168.2. MS (APCI, *m/z*): 256.2 $[M+H]^+$. Anal. Calcd for $C_{11}H_{17}N_3O_4$ (255.3): C, 51.76; H, 6.71; N, 16.46. Found: C, 51.86; H, 6.59; N, 16.40.

General procedure for ethyl (5-arylhydrazono-4-ethoxycarbonylimino-3-methyl-4,5-dihydro-1*H*-pyrrol-2-yl)carbamates (5a-5c): A solution of corresponding aromatic amine (2.0 mmol) in a mixture of ice water (8 mL) and 35% hydrochloric acid (1.2 mL) was diazotized with a solution of sodium nitrite (2.0 mmol) in ice water (4 mL). The mixture was stirred in ice bath for 15 min and then added portionwise to a solution of carbamate **4** in pyridine (16 mL) which was pre-cooled to 0–5 °C and stirred with gentle stream of nitrogen. After 1 h the reaction mixture was left to stand for 12 h at 0-5 °C and evaporated to dryness. The semi-solid product was mixed with water (20 mL) and this mixture was extracted with $CHCl_3$ (3 x 10 mL). The $CHCl_3$ extract was washed with water, dried ($MgSO_4$), evaporated to dryness and residue crystallized from aq. EtOH.

Ethyl [4-ethoxycarbonylimino-3-methyl-5-(phenylhydrazono)-4,5-dihydro-1*H*-pyrrol-2-yl]carbamate (5a): Yield: 60%, mp 97-99 °C. IR (cm^{-1}): labelled with ^{15}N (CCl_4): 3299, 3123, 2983, 1753, 1598, 1518, 1256, 1053, unlabelled (CCl_4): 3299, 3130, 2983, 1753, 1598, 1518, 1256, 1053. ^{1}H-NMR, (DMSO-d_6): 1.27-1.31 (m, 6H, 2x CH_3), 1.87 (s, 3H, CH_3), 4.16-4.21 (m, 4H, 2x CH_2), 6.88-6.95 (m, 1H, arom.), 7.25-7.33 (m, 4H, arom.), 9.46 (s, 1H, NH), 10.79 (s, 1H, NH), 10.92 (s, 1H, NH). ^{13}C-NMR, (DMSO-d_6): 9.6, 14.3, 14.5, 60.7, 61.0, 112.9, 120.4, 120.7, 129.0, 132.2, 136.4, 152.9, 162.3, 163.0, 163.2. ^{15}N-NMR, (DMSO-d_6): -239.5 (chemical shift referred to CH_3NO_2). MS (APCI, *m/z*): 360.2 $[M+H]^+$. Anal. Calcd for $C_{17}H_{21}N_5O_4$ (359.4): C, 56.82; H, 5.89; N, 19.49. Found: C, 56.86; H, 5.71; N, 19.30.

Ethyl [4-ethoxycarbonylimino-3-methyl-5-(4-methoxyphenylhydrazono)-4,5-dihydro-1*H*-pyrrol-2-yl]carbamate (5b): Yield: 55%, mp 81-83 °C. IR (cm^{-1}): 3293, 2990, 1740, 1515, 1247. ^{1}H-NMR, (DMSO-d_6): 1.25-1.32 (m, 6H, 2x CH_3), 1.82 (s, 3H, CH_3), 3.71 (s, 3H, CH_3O), 4.15-4.22 (m, 4H, 2x CH_2), 6.88 (d, 2H, J = 9.0 Hz, arom.), 7.18 (d, 2H, J = 9.0 Hz, arom.), 9.35 (s, 1H, NH), 10.66 (s, 1H,

NH), 10.85 (s, 1H, NH). MS (APCI, *m/z*): 390.2 $[M+H]^+$. Anal. Calcd for $C_{18}H_{23}N_5O_5$ (389.4): C, 55.52; H, 5.95; N, 17.98. Found: C, 55.47; H, 6.12; N, 17.88.

Ethyl [4-ethoxycarbonylimino-3-methyl-5-(4-nitrophenylhydrazono)-4,5-dihydro-1*H*-pyrrol-2-yl]-carbamate (5c): Yield: 68%, mp 107-109 °C, IR (cm^{-1}): 3328, 2997, 1721, 1637, 1525, 1500, 1249. ^{1}H-NMR, (DMSO-d_6): 1.28-1.32 (m, 6H, 2x CH_3), 1.85 (s, 3H, CH_3), 4.15-4.24 (m, 4H, 2x CH_2), 7.85 (d, 2H, J = 9.0 Hz, arom.), 8.20 (d, 2H, J = 9.0 Hz, arom.), 9.46 (s, 1H, NH), 10.77 (s, 1H, NH), 10.92 (s, 1H, NH). MS (APCI, *m/z*): 405.2 $[M+H]^+$. Anal. Calcd for $C_{17}H_{20}N_6O_6$ (404.4): C, 50.49; H, 4.99; N, 20.78. Found: C, 50.37; H, 4.89; N, 20.85.

General procedure for ethyl (2-aryl-5-methyl-3-oxo-2,3-dihydro-7*H*-pyrrolo[3,2-*e*][1,2,4]triazin-6-yl)carbamates (6a-6c): A mixture of compound **5** (1.0 mmol) and sodium carbonate (1.3 mmol) in solution of EtOH (15 mL) and water (10 mL) was refluxed for 30 min. After cooling, the formed solution was neutralized with 10% aqueous AcOH, precipitated solid filtered off and washed with water. The sample for analysis was prepared by crystallization from EtOH.

Ethyl (5-methyl-3-oxo-2-phenyl-2,3-dihydro-7*H*-pyrrolo[3,2-*e*][1,2,4]triazin-6-yl)carbamate (6a): Yield: 84%, mp > 360 °C, IR (cm^{-1}): 3283, 3005, 2863, 1699, 1610, 1262, 1260. ^{1}H-NMR, (DMSO-d_6): 1.27 (t, 3H, J = 6.9 Hz, CH_3), 1.89 (s, 3H, CH_3), 4.13 (q, 2H, J = 6.9 Hz, CH_2), 7.37 (t, 1H, J = 7.4 Hz, arom.), 7.49 (t, 2H, J = 7.4, arom.), 7.57 (d, 2H, J = 7.4 Hz, arom.), 10.96 (s, 1H, NH), 12.01 (bs, 1H, NH). ^{13}C-NMR, (DMSO-d_6): 6.7, 14.2, 60.8, 125.6, 126.8, 128.3, 128.5. Due to low solubility other signals were not observed. MS (APCI, *m/z*): 314.1 $[M+H]^+$. Anal. Calcd for $C_{15}H_{15}N_5O_3$ (313.3): C, 57.50; H, 4.83; N, 22.35. Found: C, 57.48; H, 4.62; N, 22.37.

Ethyl [5-methyl-3-oxo-2-(4-methoxyphenyl)-2,3-dihydro-7*H*-pyrrolo[3,2-*e*][1,2,4]triazin-6-yl]carbamate (6b): Yield: 87%, mp > 360 °C. IR (cm^{-1}): 3337, 2948, 1702, 1595, 1513, 1258. ^{1}H-NMR, (DMSO-d_6): 1.25 (t, 3H, J = 6.9 Hz, CH_3), 1.87 (s, 3H, CH_3), 3.78 (s, 3H, CH_3O), 4.08 (q, 2H, J = 6.9 Hz, CH_2), 6.98 (d, 2H, J = 9.2 Hz, arom.), 7.40 (d, 2H, J = 9.2 Hz, arom.), 10.96 (s, 1H, NH), 12.03 (bs, 1H, NH). MS (APCI, *m/z*): 344.2 $[M+H]^+$. Anal. Calcd for $C_{16}H_{17}N_5O_4$ (343.3): C, 55.97; H, 4.99; N, 20.40. Found: C, 55.85; H, 5.04; N, 20.31.

Ethyl-[5-methyl-3-oxo-2-(4-nitrophenyl)-2,3-dihydro-7*H*-pyrrolo[3,2-*e*][1,2,4]triazin-6-yl]carbamate (6c): Yield: 63%, mp > 360°C. IR (cm^{-1}): 3353, 2978, 1705, 1622, 1528, 1345, 1257. ^{1}H-NMR, (DMSO-d_6): 1.25 (t, 3H, J = 6.9 Hz, CH_3), 1.85 (s, 3H, CH_3), 4.15 (q, 2H, J = 6.9 Hz, CH_2), 7.90 (d, 2H, J = 9.0 Hz, arom.), 8.29 (d, 2H, J = 9.0 Hz, arom.), 10.97 (s, 1H, NH), 12.05 (bs, 1H, NH). MS (APCI, *m/z*): 359.2 $[M+H]^+$. Anal. Calcd for $C_{15}H_{14}N_6O_5$ (358.3): C, 50.28; H, 3.94; N, 23.45. Found: C, 50.30; H, 3.99; N, 23.38.

General procedure for 6-amino-2-aryl-5-methyl-2,3-dihydro-7*H*-pyrrolo[3,2-*e*][1,2,4]triazin-3-ones (7a-7c): A solution of **6** (0.15 mmol) in 1M NaOH (5 mL) was refluxed for 30 min. The cooled solution was neutralized with 10% aqueous AcOH, precipitated solid filtered off, washed with water and dried at 120°C for 1.5 h.

6-Amino-5-methyl-2-phenyl-2,3-dihydro-7*H*-pyrrolo[3,2-*e*][1,2,4]triazin-3-one (7a): Yield: 87%, mp > 300 °C (decomp). IR (cm^{-1}): 3395, 3068, 1667, 1554, 1427. ^{1}H-NMR, (DMSO-d_6): 1.82 (s, 3H, CH_3), 7.33 (t, 1H, J = 7.6, arom.), 7.46 (t, 2H, J = 7.6, arom.), 7.56 (d, 2H, J = 7.6, arom.), 8.95 (bs, 2H, NH_2), 11.67 (bs, 1H, NH). ^{13}C-NMR, (DMSO-d_6): 6.4, 125.5, 126.2, 128.1, 128.2. Due to low solubility other signals were not observed. MS (APCI, *m/z*): 242.1 $[M+H]^+$. Anal. Calcd for $C_{12}H_{11}N_5O$ (241.2): C, 59.74; H, 4.60; N, 29.03. Found: C, 59.58; H, 4.70; N, 28.94.

6-Amino-5-methyl-2-(4-methoxyphenyl)-2,3-dihydro-7*H*-pyrrolo[3,2-*e*][1,2,4]triazin-3-one (7b): Yield: 82%, mp > 300 °C (decomp). IR (cm^{-1}): 3381, 3065, 1672, 1550, 1437. ^{1}H-NMR, (DMSO-d_6): 1.83 (s, 3H, CH_3), 3.75 (s, 3H, CH_3O), 7.01 (d, 2H, J = 9.0 Hz, arom.), 7.50 (d, 2H, J = 9.0 Hz, arom.), 8.90 (bs, 2H, NH_2), 11.70 (bs, 1H, NH). MS (APCI, *m/z*): 272.2 $[M+H]^+$. Anal. Calcd for $C_{13}H_{13}N_5O_2$ (271.3): C, 57.56; H, 4.83; N, 25.82. Found: C, 57.65; H, 4.93; N, 25.74.

6-Amino-5-methyl-2-(4-nitrophenyl)-2,3-dihydro-7*H*-pyrrolo[3,2-*e*][1,2,4]triazin-3-one (7c): Yield: 92%, mp > 330 °C (decomp). IR (cm^{-1}): 3370, 3063, 1650, 1523, 1551, 1425. ^{1}H-NMR, (DMSO-d_6): 1.89 (s, 3H, CH_3), 7.86 (d, 2H, J = 9.0 Hz, arom.), 8.36 (d, 2H, J = 9.0 Hz, arom.), 8.87 (bs, 2H, NH_2), 11.71 (bs, 1H, NH). MS (APCI, *m/z*): 286.1 $[M+H]^+$. Anal. Calcd for $C_{12}H_{10}N_6O_3$ (286.2): C, 50.35; H, 3.52; N, 29.36. Found: C, 50.32; H, 3.56; N, 29.27.

General procedure for 2-aryl-5-methyl-2*H*-pyrrolo[3,2-*e*][1,2,4]triazine-3,6(4*H*,7*H*)-diones (8a-8c): A solution of compound **6** (0.15 mmol) in 2M-HCl (7 mL) was refluxed for 1 h. Precipitated crystalline yellow solid was collected on a filter by suction, washed with water and dried at 120 °C for 1.5 h.

5-Methyl-2-phenyl-2*H*-pyrrolo[3,2-*e*][1,2,4]triazine-3,6(4*H*,7*H*)-dione (8a): Yield: 81%, mp > 340 °C (decomp). IR (cm^{-1}): 3014, 2863, 1694, 1662, 1369, 1082. ^{1}H-NMR, (DMSO-d_6): 1.82 (s, 3H, CH_3), 7.35 (t, 1H, J = 7.6, arom.), 7.47 (t, 2H, J = 7.6, arom.), 7.55 (d, 2H, J = 7.6, arom.), 10.66 (bs, 1H, NH), 11.85 (bs,1H, NH). ^{13}C-NMR, (DMSO-d_6): 4.6, 96.7, 123.8, 125.0, 126.7, 126.9, 130.7, 139.8, 141.7, 170.1. MS (APCI, *m/z*): 243.1 $[M+H]^+$. Anal. Calcd for $C_{12}H_{10}N_4O_2$ (242.2) C, 59.50; H, 4.16; N, 23.13. Found: C, 59.54; H, 4.20; N, 23.07.

5-Methyl-2-(4-methoxyphenyl)-2*H*-pyrrolo[3,2-*e*][1,2,4]triazine-3,6(4*H*,7*H*)-dione (8b): Yield: 88%, mp > 310 °C (decomp). IR (cm^{-1}): 3020, 2870, 1697, 1651, 1368, 1080. ^{1}H-NMR, (DMSO-d_6): 1.82 (s, 3H, CH_3), 3.79 (s, 3H, CH_3O), 7.00 (d, 2H, J = 8.7 Hz, arom.), 7.49 (d, 2H, J = 8.7 Hz, arom.), 10.60 (bs, 1H, NH), 11.81 (bs, 1H, NH). MS (APCI, *m/z*): 273.2 $[M+H]^+$. Anal. Calcd for $C_{13}H_{12}N_4O_3$ (272.3): C,

57.35; H, 4.44; N, 20.58. Found: C, 57.32; H, 4.54; N, 20.47.

5-Methyl-2-(4-nitrophenyl)-2*H*-pyrrolo[3,2-*e*][1,2,4]triazine-3,6(4*H*,7*H*)-dione (8c): Yield: 84%, mp > 360 °C (decomp). IR (cm^{-1}): 3021, 2877, 1690, 1527, 1343. ^{1}H-NMR, (DMSO-d_6): 1.89 (s, 3H, CH_3), 7.88 (d, 2H, J = 9.0 Hz, arom.), 8.40 (d, 2H, J = 9.0 Hz, arom.), 10.59 (bs, 1H, NH), 11.83 (bs, 1H, NH). MS (APCI, *m/z*): 288.1 $[M+H]^+$. Anal. Calcd for $C_{12}H_9N_5O_4$ (287.2): C, 50.18; H, 3.16; N, 24.38. Found: C, 50.13; H, 3.25; N, 24.31.

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REFERENCES

1. Part 31 of "Cyclization Reactions of Hydrazones" series. For the previous paper see: J. Stýskala, L. Stýskalová, J. Slouka, and M. Hajdúch, *Eur. J. Med. Chem.* (2007) doi:10.1016/j.ejmech. 2007.01.008 – in press.
2. A. V. Gavai, P. Chen, and D. J. Norris, *US Pat. Appl. Publ*. 2007004732 (*C. A.,* 2007, **146**, 121990).
3. H. Mastalerz, D. M. Vyas, G. L. Trainor, and A. V. Gavai, *US Pat. Appl. Publ*. 2007004731 (*C. A.,* 2007, **146**, 121991).
4. S. J. O'Connor, J. Dumas, W. Lee, J. Dixon, and D. Cantin, *PCT Int. Appl. WO* 2007056170 (*C. A.,* 2007, **146**, 521829).
5. R. M Borzilleri, X. Zheng, L. Qian, and Ch. Ellis, *J. Med. Chem.,* 2005, **48**, 3991.
6. K. Anzai, G. Nakamura, and S. Suzuki, *J. Antibiotics*, 1957, **10A**, 201.
7. J. Slouka and P. Peč, *Monatsh. Chem*., 1972, **103**, 1444.
8. P. Peč and J. Slouka, *Chem. Zvesti*, 1975, **29**, 418.
9. J. Slouka, V. Bekárek, and V. Štemberk, *Coll. Czech. Chem. Commun*., 1978, **43**, 960.
10. J. Stýskala, J. Slouka, M. Hejsek, and V. Bekárek, *Coll. Czech. Chem. Commun*., 1997, **62**, 1754.
11. J. Stýskala and J. Slouka, *Heterocycl. Commun*., 1999, **5**, 349.
12. J. Stýskala and J. Slouka, *Heterocycl. Commun*., 1999, **5**, 157.
13. K. Kirschke, B. Costisella, M. Ramm, and B. Schulz, *J. Prakt. Chem*., 1990, **332**, 143.
14. S. G. Alekseev, V. N. Charushin, O. N. Chupakhin, G. G. Aleksandrov, S. V. Shorshnev, and A. I. Chernyshev, *Izvestiya Akademii Nauk SSSR, Seriya Khimicheskaya,* 1989, 1637.
15. V. N. Charushin, N. N. Mochulskaya, A. A. Andreiko, V. I. Filyakova, M. I. Kodess, and O. N. Chupakhin, *Tetrahedron Lett.,* 2003, **44**, 2421.
16. R. B. Woodward, W. A. Ayer, J. M. Beaton, F. Bickelhaupt, and R. Bonnett, *Tetrahedron*, 1990, **46**, 7599.

17. A. Lyčka, *Annu. Rep. NMR Spectr*., 2000, **42**, 1.
18. J. Slouka, V. Bekárek, and P. Peč, *Acta Univ. Palacki. Olomuc., Fac. Rerum Nat*., 1976, **49**, 227 (*C. A*., 1977, **87**, 68292).
19. J. Stýskala, A. Lyčka, and J. Slouka, *J. Heterocyclic Chem*., 2002, **39**, 1305.
20. J. Slouka and M. Budíková, *Acta Univ. Palacki. Olomuc., Fac. Rerum Nat*., 1974, **45**, 113 (*C. A*., 1975, **82**, 125360).
21. V. Nosková, P. Džubák, G. Kuzmina, A. Ludková, D. Stehlík, R. Trojanec, A. Janošťáková, G. Kořínková, V. Mihál, and M. Hajdúch, *Neoplasma*, 2002, **49**, 416.
22. M. Hajdúch, V. Mihál, J. Minařík, E. Faber, and M. Šafářová, *Cytotechnology*, 1996, **19**, 243.