Analogs of biologically active compounds VIII.[1] Synthesis of some derivatives of 6-azalumazine

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Abstract
In this paper the preparations of some N-aryl derivatives of 6-azalumazine are described, by ammonolysis of the corresponding ethyl 1-aryl-N-ethoxycarbonyl-6-azacytosine-5-carboxylates. The latter were obtained in two different ways by multistep reactions from amidinoacetate.

Keywords: 1,2,4-Triazine, 1-aryl-6-azacytosine, 6-azalumazine

Introduction

Attention has been paid previously to derivatives of 6-azapteridine, especially in view of their potential biological activity,[2, 3] such as the antiviral activity [4] which was already found before. 6-Azalumazine and its derivatives are interesting from this point of view. Syntheses of the above described heterocyclic system are well known,[5–14] started from convenient derivatives of pyrimidine or 1,2,4-triazine, where the second condensed ring is closed through the synthesis. Up to now all the derivatives of 6-azalumazines[15,16] were prepared by the above-mentioned protocols, but N-aryl derivatives of the 1,2,4-triazine ring had not yet been described; this communication deals with these derivatives.

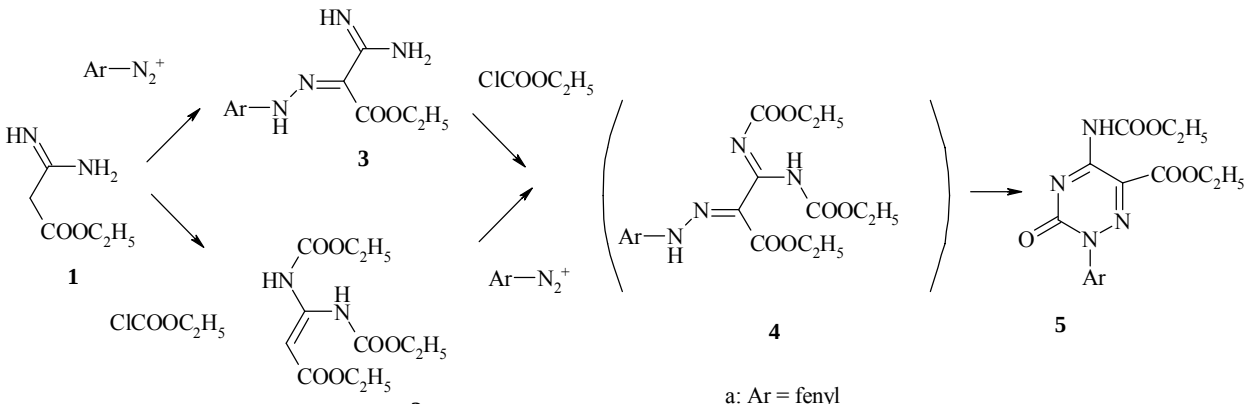
Results and Discussion

The synthesis is based on the preparation of convenient derivatives of 1-aryl-6-azacytosine-5-carboxylic acid on which the condensed pyrimidine ring is subsequently formed. A similar procedure was used to that which used in the synthesis of 1-aryl-6-azauracils[17,18] for the preparation of derivatives of 6-azacytosines.

Ethyl amidinoacetate, **1**,[19] was used as a starting material, which was treated with diazonium salts to afford the corresponding hydrazones **3a–d** in high yield. The optimal course of

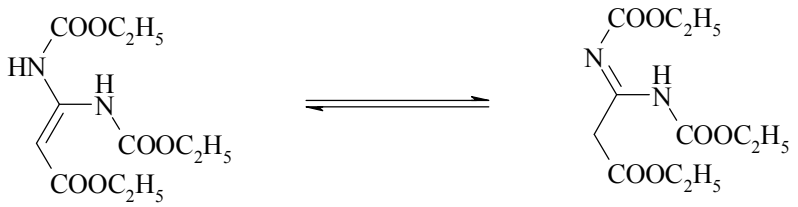
these azo-coupling reactions was found in an aqueous solution of sodium acetate and sodium carbonate. These derivatives are sensitive to hydrolysis of the ester group, so the prolonged boiling of the reaction mixture in water solution is accompanied by the formation of the poorly soluble free acid. This fact was considered when the derivatives were crystallized.

The prepared hydrazones **3** were converted into the corresponding ethyl 2-aryl-3-oxo-5-ethoxycarbonylamino-2,3-dihydro-1,2,4-triazine-6-carboxylates **5a–d** by double acylation with ethyl chloroformate in pyridine. The bis-ethoxycarbonylated hydrazones **4a–d**, formed as intermediates by the acylation with ethyl chloroformate have not been isolated yet, because in an alkaline solution of pyridine the spontaneous cyclization takes place to provide the furnished compounds **5**. The most easily formed were the derivatives **5a** and **5b**. In the case of derivatives **5c** and **5d**, the reaction time was prolonged, and the non-cyclized hydrazones **4c** and **4d** were detected at shorter reaction time. These, the most probable intermediates, were separated by TLC from the reaction mixture and evidenced by mass spectroscopy (**4c**: MS (ESI) *m/z*) 413.1 $[M+H]^+$; **4d** *m/z*: 424.1 $[M+H]^+$).



Scheme 1

Compounds **5a–d** were also prepared by another method, the coupling reaction of diazonium salts with ethyl 3,3-bis[(ethoxycarbonyl)amino]acrylate **(2)**. This compound was provided by the double acylation of ethyl amidinoacetate **(1)** with ethyl chloroformate. It was supposed that there is formed ethyl 3-[(ethoxycarbonyl)amino]-3-[(ethoxycarbonyl)imino]propanoate **(2')**, but from NMR spectroscopy it was apparent that the prepared compound is tautomeric form **2**. In the ^{1}H- NMR spectra there are two protons (δ: 10.17 ppm and 11.0 ppm) belonging to an N-H carbamate group and one proton (δ: 5.77 ppm) belongs to methine hydrogen. The presence of the methine hydrogen was also proved by ^{13}C-NMR spectroscopy by the APT method, which unambiguously confirmed structure **2**.

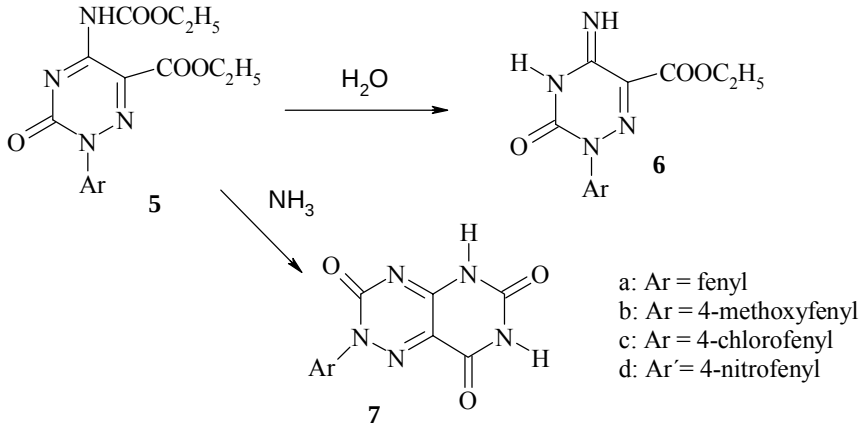


2 **2′**

Scheme 2

Compound **2** is likely in an alkaline medium to be in tautomeric equilibrium with tautomer **2'**, which depends on the basicity of the medium. The coupling reaction does not take place in pyridine solution, but it does take place in aqueous pyridine, sodium acetate, or sodium carbonate. However, the yields of these coupling reactions were relatively low. Neither a longer reaction time nor increased pH (from the presence of sodium hydroxide — where the starting material is hydrolyzed) was accompanied by a higher yield.

The compounds **5** are highly reactive. The ethoxycarbonylamine group is readily hydrolyzed to ethyl 5-imino-3-oxo-2-aryl-2,3,4,5-tetrahydro-1,2,4-triazine-6-carboxylates **(6a–d)**; in this, the 5-imino tautomeric form was proved by ^{1}H-NMR spectroscopy. In aqueous ammonia the diesters **5** were transformed into the desired derivatives of 6-azalumazine — the 2-aryl-2,3,4,5,6,7,8-hexahydropyrimido[4,5-*e*][1,2,4]triazin-3,6,8-triones **(7a–d)**.



5 **6** **7**

Scheme 3

The prepared compounds were tested for biological activity. Human breast adenocarcinoma cell line *MCF7* was used for cytotoxicity determination by the calcein AM assay.[20] The tested azacytosines **6a–d** and azalumazines **7a–d** showed poor cytostatic activity (IC_{50} = 70–126 μmol/l), with the exception of the moderately active hydrazones **3b** (IC_{50} = 14 μmol/l) and **3c** (IC_{50} = 29 μmol/l).

Experimental Section

General Procedures. 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).

Ethyl 3,3-bis[(ethoxycarbonyl)amino]acrylate (2). To a solution of the amidine **1** [19] (6.72 g, 51.7 mmol) in anhydrous pyridine (170 ml), ethyl chloroformate (17.4 ml, 0.18 mol) was added dropwise with stirring at 2–5°C. After 1 h of stirring, the reaction mixture was left to stand at 2°C for 18 h. Then, it was diluted slowly with water under stirring to a total volume of 2000 ml. The precipitated solid was collected on a filter, washed with water and crystallized from ethanol–water (3:1 v/v) mixture. Yield 8.68 g (61.3%, colorless crystals), mp 62–63 °C. IR (cm^{-1}): 3356, 3309, 3083, 1764, 1742, 1669, 1557, 1207, 1165, 1121; ^{1}H- NMR (DMSO-d_6): δ 1.17–1.27 (m, 9H, CH_3); 4.05–4.22 (m, 6H, CH_2); 5.77 (s, 1H, CH=); 10.17 (s, 1H, HN); 11.02 (s, 1H, NH); ^{13}C NMR (DMSO-d_6): δ 14.4, 14.5, 14.6 (all CH_3), 60.0, 62.3, 62.8 (all CH_2O), 79.3 (CH=), 148.0, 151.6, 153.9, 170.3; MS (ESI, *m/z*): 275.2 $[M+H]^+$; negative mode: 272.9 $[M-H]^-$. Anal. Calcd. for $C_{11}H_{18}N_2O_6$ (274.3); C, 48.17; H, 6.62; N, 10.21. Found C, 48.33; H, 6.64; N, 10.38.

Ethyl 3-amino-3-imino-2-(phenylhydrazono)propanoate (3a). A solution of aniline (1.71 g, 18.4 mmol) in a mixture of ice water (30 ml) and 35% hydrochloric acid (6.5 ml) was diazotized with a solution of sodium nitrite (1.27 g, 18.4 mmol) in ice water (30 ml). The mixture was stirred in ice bath for 15 min and then added portionwise to a solution of amidine hydrochloride **1.HCl** (3.24 g, 19.4 mmol), sodium acetate (15 g) in water (250 ml), which was pre-cooled to 0–5°C. After 10 min, to the mixture was added solution of sodium carbonate (20 g) in water (100 ml) and it was left to stand at 2°C for 1 h. The precipitated hydrazone was collected by filtration and washed with water. Crystallization: the crude hydrazone is dissolved in ethanol at 50–55°C and to this solution was added water to give gentle turbidity and left to stand at 2°C for 18 h. Yield 3.32 g (77.1%, relate to crude product, yellow crystals), mp 113–115 °C. IR (cm^{-1}): 3445, 1663, 1612, 1351, 1079, 764; ^{1}H NMR (DMSO-d_6): δ 1.27 (t, 3H, CH_3, J = 7.4 Hz); 4.14 (q, 2H, CH_2, J = 7.4 Hz); 7.12 (t, 1H, arom., J = 7.2 Hz); 7.2–7.3 (bs, 2H, NH); 7.35–7.49 (m, 4H, arom.); 9.90 (bs, NH); ^{13}C NMR (DMSO-d_6): δ 15.0, 59.2, 107.3, 120.2, 125.8, 129.2, 153.5, 157.3, 168.5; MS (ESI, *m/z*): 235.2 $[M+H]^+$. Anal. Calcd. for $C_{11}H_{14}N_4O_2$ (234.3); C, 56.40; H, 6.02; N, 23.92. Found C, 56.49; H, 6.06; N, 23.71.

Ethyl 3-amino-3-imino-2-(4-methoxyphenylhydrazono)propanoate (3b). This compound was prepared in a similar way to compound **3a**, using 4-methoxyaniline. Yield 85.7 %, light orange solid, mp 151–152 °C. IR (cm^{-1}): 3448, 3305, 1652, 1606, 1371, 1091, 839; ^{1}H NMR (DMSO-d_6): δ 1.26 (t, 3H, CH_3, J = 7.4 Hz), 3.75 (s, 3H, CH_3), 4.18 (q, 2H, CH_2, J = 7.4 Hz), 6.92 (d, 2H, arom., J = 9.0 Hz), 7.10–7.29 (bs, 2H, NH), 7.43 (d, 2H, arom., J = 9.0 Hz); 9.71 (bs, NH); MS (ESI, *m/z*): 265.2 $[M+H]^+$. Anal. Calcd. for $C_{12}H_{16}N_4O_3$ (264.3); C, 54.53; H, 6.10; N, 21.20. Found C, 54.36; H, 6.19; N, 21.24.

Ethyl 3-amino-3-imino-2-(4-chlorophenylhydrazono)propanoate (3c). Prepared in a similar way to compound **3a**, using 4-chloroaniline. Yield 93.3 %, orange solid, mp 153–154 °C. IR (cm^{-1}): 3451, 3308, 1651, 1623, 1363, 1087, 833; ^{1}H NMR (DMSO-d_6): δ 1.26 (t, 3H, CH_3, J = 7.4 Hz); 4.16 (q, 2H, CH_2, J = 7.4 Hz); 7.37 (d, 2H, arom., J = 9.0 Hz); 7.41–7.46 (bs, 2H, NH); 7.47 (d, 2H, arom., J = 9.0 Hz); 9.80 (bs, NH); MS (ESI, *m/z*): 269.3 $[M+H]^+$. Anal. Calcd. for $C_{11}H_{13}ClN_4O_2$ (268.7); C, 49.17; H, 4.87; N, 20.85. Found C, 49.52; H, 5.04; N, 20.38.

Ethyl 3-amino-3-imino-2-(4-nitrophenylhydrazono)propanoate (3d). This compound was prepared in a similar way to compound **3a**, using 4-nitroaniline. Yield 90.6 %, red solid, mp 198–200 °C. IR (cm^{-1}): 3448, 3310, 1659, 1627, 1353, 1080, 846; ^{1}H NMR (DMSO-d_6): δ 1.28 (t, 3H, CH_3, J = 7.4 Hz); 4.16 (q, 2H, CH_2, J = 7.4 Hz); 7.58 (d, 2H, arom., J = 9.0 Hz); 7.60–7.70 (bs, 2H, NH); 8.19 (d, 2H, arom., J = 9.0 Hz); 10.11 (bs, NH); MS (ESI, *m/z*): 280.1 $[M+H]^+$. Anal. Calcd. for $C_{11}H_{13}N_5O_4$ (279.3); C, 47.31; H, 4.69; N, 25.08. Found C, 47.31; H, 4.61; N, 25.38.

Ethyl 5-[(ethoxycarbonyl)amino]-3-oxo-2-phenyl-2,3-dihydro-1,2,4-triazine-6-carboxylate (5a). Method A. To a cooled solution (2–5°C) of compound **3a** (1.24 g, 5.30 mmol) in anhydrous pyridine (80 ml), ethyl chloroformate (6.0 ml, 62.5 mmol) was added dropwise with stirring at 2–5°C. After 3 h of stirring, the mixture was left to stand at 2°C for 18 h. Then, it was diluted slowly with water under stirring to a total volume of 1000 ml. The compound starts to precipitate after standing during some hours. The precipitated solid was collected on a filter, washed with water and crystallized from ethanol–water (1:1 v/v) mixture. Yield 1.46 g of hydrate of **5a**. Anhydrous compound was obtained by drying at 115°C for 1.5 h. Yield 1.38 g (78.7 %).

Method B. A solution of aniline (230 mg, 2.48 mmol) in a mixture of ice-water (3 ml) and 35% hydrochloric acid (2 ml) was diazotized with a solution of sodium nitrite (171.1 mg, 2.48 mmol) in ice-water (5 ml). The solution was stirred in an ice bath for 15 min and then added portionwise to a mixture of compound **2** (680 mg, 2.48 mmol), sodium carbonate (1.0 g) and sodium acetate (4 g) in a mixture of pyridine (30 ml) and water (17 ml), pre-cooled to 0–5°C. The mixture was stirred for 6 h and left to stand at 0–5 °C for 48 h and slowly diluted with water to a total volume of 400 ml. The next day the precipitated solid was collected, washed with water and crystallized from ethanol–water (1:1 v/v) mixture. Yield 530 mg of hydrate of **5a**; anhydrous compound was obtained by drying at 115 °C for 1.5 h. Yield 503 mg (61.2 %, pale yellow needles), mp 153–155 °C. IR (cm^{-1}): 3244, 1764, 1698, 1536, 1184, 1105; ^{1}H NMR (DMSO-d_6): δ 1.27 (t, 3H, CH_3, J = 6.9 Hz); 1.31 (t, 3H, CH_3, J = 7.2 Hz); 4.21 (q, 2H, CH_2, J = 6.9 Hz); 4.31 (q, 2H, CH_2, J = 7.2 Hz); 7.40–7.55 (m, 3H, arom.); 7.68 (d, 2H, arom., J = 24 Hz); 10.92 (s, 1H, NH); ^{13}C NMR (DMSO-d_6): δ 14.2, 14.3, 62.8, 63.3, 126.0, 127.1, 129.2, 129.4, 138.1, 141.2, 151.6, 154.2, 162.3; MS (ESI, *m/z*): 333.2 $[M+H]^+$. Anal. Calcd. for $C_{15}H_{16}N_4O_5$ (332.3); C, 54.22; H, 4.85; N, 16.86. Found C, 54.42; H, 4.67; N, 16.59.

Ethyl 5-[(ethoxycarbonyl)amino]-3-oxo-2-(4-methoxyphenyl)-2,3-dihydro-1,2,4-triazine-6-carboxylate (5b). Method A. This compound was prepared in a similar way to compound **5a**, using compound **3b** (1.40 g, 5.30 mmol). This compound crystallized in anhydrous form. Yield 1.22 g (63.5 %).

Method B. Prepared in a similar way to compound **5a**, using 4-methoxyaniline (305 mg, 2.48 mmol). Yield 320 mg (35.6 %, yellow needles), mp 150–152°C. IR (cm^{-1}): 3260, 1777, 1693, 1538, 1172, 1103. ^{1}H NMR (DMSO-d_6): δ 1.25 (t, 3H, CH_3, J = 6.9 Hz); 1.27 (t, 3H, CH_3, J =

7.2 Hz); 3.80 (s, 3H, CH_3); 4.19 (q, 2H, CH_2, *J* = 6.9 Hz); 4.29 (q, 2H, CH_2, *J* = 7.2 Hz); 7.05 (d, 2H, arom., *J* = 9.0 Hz); 7.49 (d, 2H, arom., *J* = 9.0 Hz); 10.91 (s, 1H, NH); MS (ESI, *m/z*): 363.2 $[M+H]^+$. Anal. Calcd. for $C_{16}H_{18}N_4O_6$ (362.3); C, 53.04; H, 5.01; N, 15.47. Found C, 52.89; H, 4.88; N, 15.23.

Ethyl 5-[(ethoxycarbonyl)amino]-3-oxo-2-(4-chlorophenyl)-2,3-dihydro-1,2,4-triazine-6-carboxylate (5c). Method A: This compound was prepared in a similar way to **5a**, using compound **3c** (1.43 g, 5.30 mmol), but the reaction time was prolonged to 72 h. This compound crystallized anhydrous. Yield 1.42 g (73.2 %).

Method B. This compound was prepared in a way similar to compound **5a**, using 4-chloroaniline (316 mg, 2.48 mmol), but the reaction time was prolonged to 72 h. Yield 296 mg (32.6 %, pale yellow needles), mp 154–156°C. IR (cm^{-1}): 3253, 1770, 1695, 1535, 1170, 1103; ^1H NMR (DMSO-d_6): δ 1.25 (t, 3H, CH_3, *J* = 6.9 Hz); 1.27 (t, 3H, CH_3, *J* = 7.2 Hz); 4.19 (q, 2H, CH_2, *J* = 6.9 Hz); 4.30 (q, 2H, CH_2, *J* = 7.2 Hz); 7.61 (s, 4H, arom.); 10.93 (s, 1H, NH); MS (ESI, *m/z*): 367.4 $[M+H]^+$. Anal. Calcd. for $C_{15}H_{15}ClN_4O_5$ (366.8); C, 49.12; H, 4.12; N, 15.28. Found C, 49.30; H, 4.01; N, 15.25.

Ethyl 5-[(ethoxycarbonyl)amino]-3-oxo-2-(4-chlorophenyl)-2,3-dihydro-1,2,4-triazine-6-carboxylate (5d). Method A: This compound was prepared in a similar way to compound **5a** using compound **3d** (1.47 g, 5.30 mmol), but the reaction time was prolonged to 72 h. This compound crystallized in anhydrous form. Yield 1.40 g (70.1 %).

Method B. Prepared in a similar way to compound **5a**, using 4-nitroaniline (325.5 mg, 2.48 mmol), but the reaction time was prolonged to 72h. Yield 305 mg (34.3 %, pale yellow solid), mp 179–181°C. IR (cm^{-1}): 3250, 1770, 1698, 1551, 1348, 1176; ^1H NMR (DMSO-d_6): δ 1.25 (t, 3H, CH_3, *J* = 6.9 Hz); 1.27 (t, 3H, CH_3, *J* = 7.2 Hz); 4.20 (q, 2H, CH_2, *J* = 6.9 Hz); 4.31 (q, 2H, CH_2, *J* = 7.29 Hz); 7.90 (d, 2H, arom., *J* = 9.0 Hz); 8.39 (d, 2H, arom., *J* = 9.0 Hz); 11.04 (s, 1H, NH); MS (ESI, *m/z*): 378.1 $[M+H]^+$. Anal. Calcd. for $C_{15}H_{15}N_5O_7$ (377.3); C, 47.75; H, 4.01; N, 18.56. Found C, 47.49; H, 4.23; N, 18.46.

Ethyl 5-imino-3-oxo-2-phenyl-2,3,4,5-tetrahydro-1,2,4-triazine-6-carboxylate (6a). A mixture of **5a** (177.4 mg, 0.534 mmol) in xylene (4 ml) and water (0.1 ml) was heated at reflux for 60 min. Hexane (5 ml) was added to the cooled mixture, and left to stand at 2°C for 1 h. The precipitated solid was collected on a filter and washed with hexane. The sample for analysis was prepared by crystallization from ethanol–water (3:2 v/v). Yield 110.2 mg (79.3 %, white needles), mp 229–231°C. IR (cm^{-1}): 3400, 3092, 1681, 1641, 1512, 1111. ^1H NMR (DMSO-d_6): δ 1.27 (t, 3H, CH_3, *J* = 6.9 Hz); 4.32 (q, 2H, CH_2, *J* = 6.9 Hz); 7.40–7.55 (m, 5H, arom.); 7.88 (s, 1H, NH); 8.63 (s, 1H, NH); ^{13}C NMR (DMSO-d_6): δ 14.4, 62.3, 126.1, 128.6, 129.0, 141.8, 152.1,156.2,157.8, 163.1; MS (ESI, *m/z*): 261.2 $[M+H]^+$. Anal. Calcd. for $C_{12}H_{12}N_4O_3$ (260.3); C, 55.38; H, 4.65; N, 21.53. Found C, 55.03; H, 4.33; N, 21.62.

Ethyl 5-imino-3-oxo-2-(4-methoxyphenyl)-2,3,4,5-tetrahydro-1,2,4-triazine-6-carboxylate (6b). Prepared in a similar way to compound **6a**, using compound **5b** (46.3 mg, 0.16 mmol). The sample for analysis was obtained by crystallization from ethanol. Yield 32.2 mg (86.8 %, pale yellow needles), mp 248–250°C. IR (cm^{-1}): 3400, 3090, 1698, 1652, 1514, 1124. ^1H NMR (DMSO-d_6): δ 1.28 (t, 3H, CH_3, *J* = 6.9 Hz); 3.81 (s, 3H, CH_3); 4.32 (q, 2H, CH_2, *J* = 6.9 Hz); 7.03 (d, 2H, arom., *J* =9.0 Hz); 7.42 (d, 2H, arom., *J* =9.0 Hz); 7.83 (s, 1H, NH); 8.54 (s, 1H,

NH); MS (ESI, *m/z*): 291.2 $[M+H]^+$. Anal. Calcd. for $C_{13}H_{14}N_4O_4$ (290.3); C, 53.79; H, 4.86; N, 19.30. Found C, 53.93; H, 4.76; N, 19.34.

Ethyl 5-imino-3-oxo-2-(4-chlorophenyl)-2,3,4,5-tetrahydro-1,2,4-triazine-6-carboxylate (6c). Prepared in a similar way to compound **6a** using compound **5c** (44.3 mg, 0.15 mmol). The sample for analysis was crystallized from ethanol. Yield 31.3 mg (87.9 %, white needles), mp 251–253°C. ^{1}H NMR (DMSO-d_6): δ 1.28 (t, 3H, CH_3, J = 6.9 Hz); 4.33 (q, 2H, CH_2, J = 6.9 Hz); 7.57 (s, 4H, arom.); 7.90 (s, 1H, NH); 8.65 (s, 1H, NH); IR (cm^{-1}): 3405, 3085, 1719, 1694, 1493, 1126; MS (ESI, *m/z*): 295.3 $[M+H]^+$. Anal. Calcd. for $C_{12}H_{11}ClN_4O_3$ (294.7); C, 48.91; H, 3.76; N, 19.01. Found C, 48.98; H, 3.89; N, 18.86.

Ethyl 5-imino-3-oxo-2-(4-nitrophenyl)-2,3,4,5-tetrahydro-1,2,4-triazine-6-carboxylate (6d). Prepared in a similar way to compound **6a** using compound **5d** (48.5 mg, 0.159 mmol). The sample for analysis was crystallized from ethanol. Yield 30.7 mg (78.3 %, light orange solid), mp 262–264°C. IR (cm^{-1}): 3400, 3075, 1719, 1698, 1500, 1348, 1094; ^{1}H NMR (DMSO-d_6): δ 1.30 (t, 3H, CH_3, J = 6.9 Hz); 4.35 (q, 2H, CH_2, J = 6.9 Hz); 7.87 (d, 2H, arom., J =9.0 Hz); 7.89 (s, 1H, NH); 8.35 (d, 2H, arom., J =9.0 Hz); 8.78 (s, 1H, NH); MS (ESI, *m/z*): 306.1 $[M+H]^+$. Anal. Calcd. for $C_{12}H_{11}N_5O_5$ (305.3); C, 47.22; H, 3.63; N, 22.94. Found C, 47.14; H, 3.81; N, 22.87.

2-Phenyl-2,3,5,6,7,8-hexahydropyrimido[4,5-e][1,2,4]triazin-3,6,8-trione (7a). A solution of compound **5a** (100.6 mg, 0.303 mmol) in 26% aq. ammonia (6 ml) was stirred for 30 min, with precipitation from the solution. The mixture was evaporated and the resulting yellow compound crystallized from water and dried at 110°C for 90 min. Yield 62.2 mg (83.7 %, yellow needles), mp 327–329°C. IR (cm^{-1}): 3250, 1717, 1682, 1595, 1438, 1277; ^{1}H NMR (DMSO-d_6): δ 7.47–7.65 (m, 5H, arom.); 11.83 (s, 1H, NH); 12.32 (bs, 1H, NH); ^{13}C NMR (DMSO-d_6): δ 125.1,125.9, 129.0, 129.4, 141.4, 150.1, 152.5, 155.7, 158.2; MS (ESI, *m/z*): 258.1 $[M+H]^+$. Anal. Calcd. for $C_{11}H_7N_5O_3$ (257.2); C, 51.37; H, 2.74; N, 27.23. Found C, 51.22; H, 2.70; N, 27.31.

2-(4-Methoxyphenyl)-2,3,5,6,7,8-hexahydropyrimido[4,5-e][1,2,4]triazin-3,6,8-trione (7b). Prepared in a similar way to compound **7a** using compound **5b** (125.0 mg, 0.345 mmol). Yield 85.2 mg (86.0 %, yellow needles), mp 319–321°C. IR (cm^{-1}): 3247, 1741, 1688, 1618, 1428, 1267; ^{1}H NMR (DMSO-d_6): δ 3.82 (s, 3H, CH_3); 7.07 (d, 2H, arom, J = 9.0 Hz); 7.49 (d, 2H, arom, J = 9.0 Hz); 11.80 (bs, 1H, NH); 12.20 (bs, 1H, NH); MS (ESI, *m/z*): 288.2 $[M+H]^+$. Anal. Calcd. for $C_{12}H_9N_5O_4$ (287.2); C, 50.18; H, 3.16; N, 24.38. Found C, 49.97; H, 3.19; N, 24.30.

2-(4-Chlorophenyl)-2,3,5,6,7,8-hexahydropyrimido[4,5-e][1,2,4]triazin-3,6,8-trione (7c). Prepared in a similar way to compound **7a**, using compound **5c** (83.5 mg, 0.228 mmol). Yield 52.3 mg (78.8 %, pale yellow needles), mp 345–350°C. IR (cm^{-1}): 3250, 1739, 1698, 1619, 1316; ^{1}H NMR (DMSO-d_6): δ 7.62 (s, 4H, arom.); 11.80 (bs, 1H, NH); 12.31 (bs, 1H, NH); MS (ESI, *m/z*): 292.2 $[M+H]^+$. Anal. Calcd. for $C_{11}H_6ClN_5O_3$ (291.7); C, 45.30; H, 2.07; N, 24.01. Found C, 45.24; H, 2.01; N, 24.28.

2-(4-Nitrophenyl)-2,3,5,6,7,8-hexahydropyrimido[4,5-e][1,2,4]triazin-3,6,8-trione (7d). Prepared in a similar way to compound **7a**, using compound **5d** (103.3 mg, 0.273 mmol). Yield 61.1 mg (73.8 %, light orange solid), mp 355–357°C. IR (cm^{-1}): 3245, 1733, 1707, 1611, 1313; ^{1}H NMR (DMSO-d_6): δ 7.92 (d, 2H, arom, J = 9.0 Hz); 8.37 (d, 2H, arom, J = 9.0 Hz); 11.80 (bs, 1H, NH); 12.31 (bs, 1H, NH); MS (ESI, *m/z*): 303.1 $[M+H]^+$. Anal. Calcd. for $C_{11}H_6N_6O_5$ (302.2); C, 43.72; H, 2.00; N, 27.81. Found C, 43.65; H, 1.96; N, 27.77.

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