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Efficient synthesis and biological evaluation of some 2,4-diamino-furo[2,3-*d*]pyrimidine derivatives

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ABSTRACT

The carbodiimides **2**, obtained from aza-Wittig reactions of iminophosphorane **1** with aromatic isocyanates, reacted with ammonia to give ethyl 3,4-dihydro-6-methyl-4-oxo-2-arylamino-furo[2,3-*d*]pyrimidine-5-carboxylate **3**. Further reaction of **3** with $POCl_3$ and various amines generated ethyl 4-alkylamino-2-arylamino-6-methyl-furo[2,3-*d*]pyrimidine-5-carboxylate **5** in good yields. Their structures were confirmed by ^1H NMR, EI-Ms, IR and elemental analysis. Compound **5b** was further analyzed by single crystal X-ray diffraction. Compound **5** exhibited cytotoxicity against two lung cancer cell lines. For example, compound **5a** showed the best inhibition activities against A459 with IC_{50} 0.8 μM.

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The derivatives of fused pyrimidines have shown remarkable biological properties such as antitumor and antibacterial activities.[1–3] Among them, furo[2,3-*d*]pyrimidines have been considered as templates for drug discovery for many years with the inhibition of dihydrofolate reductase (DHFR) as the primary target.[4,5] More recently, furopyrimidines have been found to be active as kinase inhibitors.[6,7] For example, Gangjee et al. recently reported a series of compounds containing the backbone of 2,4-diaminofuropyrimidines which have potent dual thymidylate synthase and dihydrofolate reductase inhibitors (Fig. 1), However, there are only few reports on the synthesis of 4-alkylamino-2-arylamino-furopyrimidines, which are of considerable interest as potential biologically active compounds or pharmaceuticals.

Over past twenty years, the aza-Wittig reactions of iminophosphoranes have received increased attention in view of their utility in the synthesis of nitrogen-containing heterocyclic compounds.[8–10] Annelation of ring systems with N-heterocycles by means an aza-Wittig reaction has been widely utilized because of the availability of functionalized iminophosphoranes. Recently we have been interested in the synthesis of fused pyrimidinones and imidazolones via aza-Wittig reaction, with the aim of evaluating their biological activities.[11–13] Here, we wish to reported a new efficient synthesis and antitumor activities of ethyl 4-alkylamino-2-arylamino-6-methyl-furo[2,3-*d*]pyrimidine-5-carboxylate **5** from easily accessible iminophosphorane **1**.[14]

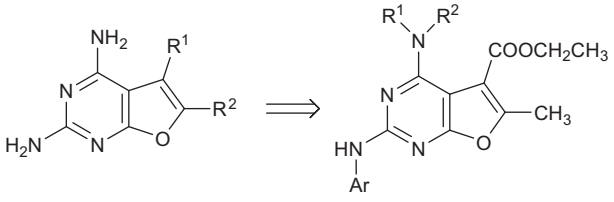

Figure 1. 2,4-Diaminofuropyrimidines have potent dual thymidylate synthase and dihydrofolate reductase inhibitors.

Iminophosphorane **1** reacted with aromatic isocyanates to give carbodiimides **2**, which were allowed to react with NH_3 to give selectively ethyl 3,4-dihydro-6-methyl-4-oxo-2-arylamino-furo[2,3-*d*]pyrimidine-5-carboxylates **3**[15] in satisfactory yields at room temperature. Compounds **3** were further converted to functionalized ethyl 4-chloro-6-methyl-2-arylamino-furo[2,3-*d*]-pyrimidine-5-carboxylates **4**[16] via reaction with $POCl_3$ at 80–85 °C (88–95%, Scheme 1).

Ethyl 4-chloro-6-methyl-2-arylamino-furo[2,3-*d*]pyrimidine-5-carboxylates **4** reacted with primary amines or secondary amines in acetonitrile at 70–80 °C to give ethyl 4-alkylamino-2-arylamino-6-methyl-furo[2,3-*d*]pyrimidine-5-carboxylate **5** in good yields[17] (80–90%, Table 1, Scheme 2).

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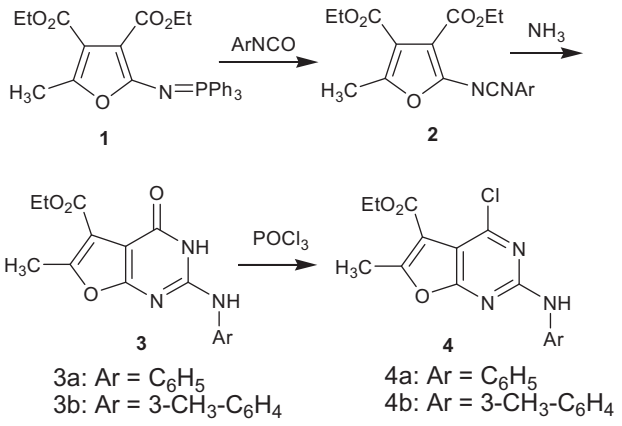


Scheme 1.

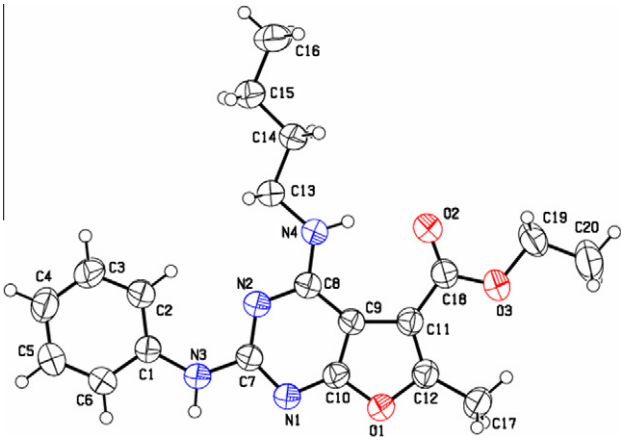
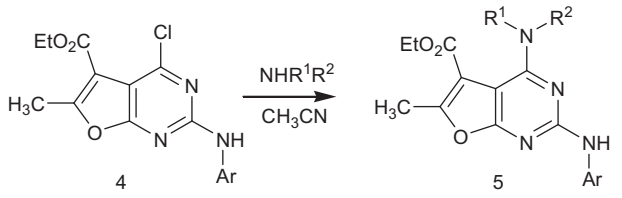


Figure 2. Ortep diagram of the crystal structure of compound **5b** (drawn at the 50% thermal ellipsoids).

Table 1
Preparation of ethyl-4-alkylamino-2-arylamino-furo[2,3-*d*]pyrimidine-5-carboxylates **5**

Compound	Ar	NR^1R^2	Conditions (°C/h)	Yields[a] (%)
5a	Ph	4-Methylpiperazin-1-yl	70–80/5	81
5b	Ph	*n*-Butylamino	70–80/6	85
5c	Ph	*iso*-Propylamino	70–80/6	87
5d	Ph	2-Hydroxyethylamino	70–80/5	80
5e	Ph	Morpholino	70–80/6	90
5f	3-CH_3–C_6H_4	*n*-Butylamino	70–80/5	84
5g	3-CH_3–C_6H_4	Morpholino	70–80/5	86
5h	3-CH_3–C_6H_4	4-Methylpiperazin-1-yl	70–80/6	80
5i	3-CH_3–C_6H_4	Diethylamino	70–80/6	82
5j	3-CH_3–C_6H_4	*iso*-Propylamino	70–80/6	85

[a] Isolated yields based on ethyl 4-chloro-6-methyl-2-arylamino-furo[2,3-*d*]pyrimidine-5-carboxylates 4.



Scheme 2.

The structure of ethyl 4-alkylamino-2-arylamino-6-methyl-furo[2,3-*d*]pyrimidine-5-carboxylate **5** was confirmed by their spectrum data. For example, the ^{1}H NMR spectrum of **5a** shows the signals of $-OCH_2$ at 4.32 ppm as quartets, signals of CH_3 at 2.29, 2.57, 1.36 ppm as singlet or triplets. The signals attributable to the NCH_2 are found at 2.47 and 3.57 ppm as broad singlet. The phenyl and ArN-H signals appeared at 6.96–7.60 ppm as multiplets. The IR spectra of **5a** revealed C=O absorption bands at 1694 cm^{-1}. The MS spectrum of **5a** shows strong molecular ion peak at *m*/*z* 395 with 100% abundance. Furthermore a single crystal of **5b** was obtained from a CH_2Cl_2 solution of **5b**.[18] X-ray structure analysis verified again the structure, and showed that all ring atoms in the furo[2,3-*d*]pyrimidine moiety are nearly coplanar and the phenyl ring is twisted with respect to the pyrimidine ring system by 8.00(7)° (Fig. 2).

The biological activities of **5** were investigated, and the results showed that these compounds exhibited cytotoxicity against two lung cancer cell lines A459 and SPC-A-1.[19] As indicated in Table 2, compound **5a** showed the best inhibition activities against A459 with IC_{50} 0.8 μM (Table 2).

Table 2
In vitro cytotoxicity (IC_{50}[a], μM) of furo[2,3-*d*]pyrimidine **5**

Compound	A459	SPC-A-1
5a	0.8	26.1
5b	2.7	50.2
5c	21.5	–
5d	13.0	24.2
5e	15.2	69.0
5f	9.0	–
5g	14.0	70.8
5h	5.5	18.2
5i	8.8	–
5j	8.6	46.3

[a] IC_{50} is the concentration of compound required to inhibit the cell growth by 50% compared to an untreated control.

In conclusion, we have developed an efficient iminophosphorane-mediated synthesis of previously unreported ethyl 4-alkylamino-2-arylamino-6-methyl-furo[2,3-*d*]pyrimidine-5-carboxylate **5** via aza-Wittig reactions. Bioassay of the compounds indicated that 4-alkylamino-2-arylamino-6-methyl-furo[2,3-*d*]pyrimidines can be used as lead structure for developing novel antitumor drugs. Further bioassay, optimization and structure–activity relationships of the title compounds are underway.

Acknowledgments

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15. Preparation of ethyl 3,4-dihydro-6-methyl-4-oxo-2-arylamino-furo[2,3-*d*]pyrimidine-5-carboxylate **3a**–**3b**. To a solution of ammonia hydrate (~20 mmol) in EtOH (20 mL) was added carbodiimide (5 mmol)[14] **2** in methylene chloride. After stirring for 1–2 h, the solution was concentrated under reduced pressure and the residue was recrystallized from methylene chloride/petroleum ether to give ethyl 3,4-dihydro-6-methyl-4-oxo-2-arylamino-furo[2,3-*d*]pyrimidine-5-carboxylate **3a**–**3b**. Ethyl 3,4-dihydro-6-methyl-4-oxo-2-phenylamino-furo[2,3-*d*]pyrimidine-5-carboxylate (**3a**). White crystals (yield, 90%) mp: 281–282 °C. IR (KBr): 3236 (N–H), 1706 (C=O), 1568, 1379, 1048. ^{1}H NMR (400 MHz, DMSO-d_6) δ: 1.37(t, J = 7.2, 3H, CH_3), 2.88 (s, 3H, CH_3), 4.34 (q, J = 7.2, 2H, CH_2), 5.22 (s, H, NH), 7.33–7.63 (m, 6H, Ar-H and ArN-H). MS *m*/*z*: 313 (M^+, 100), 198 (43), 145 (50), 77 (12). Anal. Calcd for $C_{16}H_{15}N_3O_4$ (313.31), C, 61.34; H, 4.83; N, 13.41. Found: C, 61.01; H, 4.58; N, 13.29. Ethyl 2-(*m*-tolylamino)-3,4-dihydro-6-methyl-4-oxofuro[2,3-*d*]pyrimidine-5-carboxylate (**3b**). White crystals (yield, 88%), mp: 262–284 °C. IR (KBr): 3337 (N–H), 1700 (C=O), 1545, 1168. ^{1}H NMR (400 MHz, DMSO-d_6) δ: 1.37 (t, J = 7.2, 3H, CH_3), 2.43 (s, 3H, CH_3), 2.85 (s, 3H, CH_3), 4.34 (q, J = 7.2, 2H, CH_2), 5.24 (s, H, NH), 7.06–7.54 (m, 5H, Ar-H and ArN-H). MS *m*/*z*: 327 (M^+, 100), 203 (37), 160 (29), 91 (46). Anal. Calcd for $C_{17}H_{17}N_3O_4$ (327.33): C, 62.38; H, 5.23; N, 12.84. Found: C, 62..21; H, 5.17; N, 12.75.
16. Preparation of ethyl 4-chloro-6-methyl-2-arylamino-furo[2,3-*d*]pyrimidine-5-carboxylates **4**: To a mixture of **3** (5 mmol) prepared above and phosphorous oxychloride (5 mL) were stirred at 80–90 °C for 6–8 h. The solution was concentrated under reduced pressure to remove excessive phosphorous oxychloride. The residue was poured into ice water and recrystallized from methylene chloride/petroleum ether to give ethyl 2-arylamino-4-chloro-6-methyl-furo[2,3-*d*]pyrimidine-5-carboxylate **4a**–**4b**. Ethyl 4-chloro-6-methyl-2-phenylamino-furo [2,3-*d*] pyrimidine-5-carboxylate (**4a**). White crystals (yield, 91%), mp: 165–167 °C. IR (KBr): 3361 (N–H), 1707 (C=O), 1523, 1450, 1165, 754. ^{1}H NMR (400 MHz, $CDCl_3$) δ: 1.42 (t, J = 7.2, 3H, CH_3), 2.68 (s, 3H, CH_3), 4.41 (q, J = 7.2, 2H, CH_2), 7.07–7.63 (m, 6H, Ar-H and ArN-H). MS *m*/*z*: 331 (M^+, 100), 302 (25), 250 (20), 222 (9), 180 (5), 118 (6), 77 (10). Anal. Calcd for $C_{16}H_{14}ClN_3O_3$ (331.75): C, 57.93; H, 4.25; N, 12.67. Found: C, 57.65; H, 4.00; N, 12.52. Ethyl 4-chloro-6-methyl-2-(*m*-tolylamino)-furo[2,3-*d*]pyrimidine-5-carboxylate (**4b**). White crystals (yield, 93%), mp: 140–142 °C. IR (KBr): 3361 (N–H), 1702 (C=O), 1530, 1438, 748. ^{1}H NMR (400 MHz, $CDCl_3$) δ: 1.40 (t, J = 7.2, 3H, CH_3), 2.30 (s, 3H, CH_3), 2.62 (s, 3H, CH_3), 4.40 (q, J = 7.2, 2H, CH_2), 6.99–7.58 (m, 5H, Ar-H, ArN-H). MS *m*/*z*: 345 (M^+, 100), 315 (45), 250 (28), 77 (8). Anal. Calcd for $C_{17}H_{16}ClN_3O_3$ (345.78): C, 59.05; H, 4.66; N, 12.15. Found: C, 58.96; H, 4.60; N, 12.03.
17. Preparation of ethyl 6-methyl-4-alkylamino-2-arylamino-furo[2,3-*d*]pyrimidine-5-carboxylate **5a**–**5j**. To a solution of **4** (1 mmol) prepared above in CH_3CN (5 mL) was added K_2CO_3 and primary amines or secondary amines (3 mmol). The mixture was stirred at 70–80 °C for 6–8 h. The solution was concentrated under reduced pressure and the residue was recrystallized from methylene chloride/petroleum ether to give 6-methyl-4-alkylamino-2-arylamino-furo[2,3-*d*]pyrimidine-5-carboxylate **5a**–**5j**. Ethyl 6-methyl-4-(4-methylpiperazin-1-yl)-2-(phenylamino)furo[2,3-*d*]pyrimidine-5-carboxylate (**5a**). White crystals (yield, 81%). Mp: 126–128 °C. ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 1.36 (t, J = 7.2, 3H, CH_3), 2.29 (s, 3H, CH_3), 2.47 (br s, 4H, 2 × NCH_2), 2.57 (s, 3H, CH_3), 3.57 (br s, 4H, 2 × NCH_2), 4.32 (t, J = 7.2, 2H, CH_2), 6.96–7.60 (m, 6H, Ar-H and ArN-H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ: 13.8, 14.3, 46.0, 48.1, 54.5, 61.0, 93.9, 109.3, 118.9, 121.8, 128.6, 139.8, 155.1, 155.8, 159.6, 163.9, 168.3. IR (KBr): 3364 (N–H), 1694 (C=O), 1628, 1531, 1410, 748. MS (70 eV) *m*/*z* (%): 395 (M^+, 100), 302 (30), 260 (17), 222 (26), 77 (9). Anal. Calcd for $C_{21}H_{25}N_5O_3$ (395.45): C, 63.78; H, 6.37; N, 17.71. Found: C, 63.68; H, 6.43; N, 17.66. Ethyl 4-(butylamino)-6-methyl-2-(phenylamino) furo[2,3-*d*]pyrimidine-5-carboxylate (**5b**). White crystals (yield, 85%). Mp: 144–146 °C. ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 0.98 (t, J = 7.2, 3H, CH_3), 1.40 (t, J = 7.2, 3H, CH_3), 1.45–1.48 (m, 4H, CH_2CH_2), 2.63 (s, 3H, CH_3), 3.53–3.56 (m, 2H, CH_2), 4.37 (t, J = 7.2, 2H, CH_2), 6.97–7.68 (m, 6H, Ar-H and ArN-H), 8.20 (s, 1H, N–H); IR (KBr): 3378, 3287 (N–H), 1693 (C=O), 1628, 1532, 1415, 1138, 770 cm^{-1}. MS (70 eV) *m*/*z* (%): 368 (M^+, 100), 339 (36), 279 (41), 252 (29), 184 (5), 77 (5). Anal. Calcd for $C_{20}H_{24}N_4O_3$ (368.43): C, 65.20; H, 6.57; N, 15.21. Found: C, 65.07; H, 6.43; N, 15.10. Ethyl 4-(iso-propylamino)-6-methyl-2-(phenylamino) furo[2,3-*d*]pyrimidine-5-carboxylate (**5c**). White crystals (yield, 87%). Mp: 181–182 °C. ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 1.32 (d, J = 6.6, 6H, 2 × CH_3), 1.40 (t, J = 7.2, 3H, CH_3), 2.63 (s, 3H, CH_3), 4.36 (t, J = 7.2, 3H, CH_2 and CH), 6.97–7.67 (m, 6H, Ar-H and ArN-H), 8.10 (s, 1H, N–H); IR (KBr): 3288 (N–H), 1692 (C=O), 1618, 1533, 1417, 1127, 771. MS (70 eV) *m*/*z* (%): 354 (M^+, 100), 339 (49), 293 (42), 252 (20), 147 (9), 119 (13), 77 (5). Anal. Calcd for $C_{19}H_{22}N_4O_3$ (354.40): C, 64.39; H, 6.26; N, 15.81. Found: C, 64.24; H, 6.13; N, 15.67. Ethyl 4-(2-hydroxyethylamino)-6-methyl-2-(phenylamino) furo[2,3-*d*]pyrimidine-5-carboxylate (**5d**). White crystals (yield, 80%). Mp: 129–131 °C. ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 1.40 (t, J = 7.2, 3H, CH_3), 2.62 (s, 3H, CH_3), 3.68 (s, 1H, OH), 3.73 (t, J = 4.8, 2H, CH_2), 3.86 (t, J = 4.8, CH_2), 4.34 (t, J = 4.8, 2H, CH_2), 6.99–7.61 (m, 6H, Ar-H and ArN-H), 8.53 (s, 1H, N–H); IR (KBr): 3319 (N–H), 1691 (C=O), 1620, 1538, 1445, 1142, 770. MS (70 eV) *m*/*z* (%): 356 (M^+, 100), 325 (46), 280 (67), 252 (43), 77 (8). Anal. Calcd for $C_{18}H_{20}N_4O_4$ (356.38): C, 60.66; H, 5.66; N, 15.72. Found: C, 60.47; H, 5.78; N, 15.61. Ethyl 6-methyl-4-morpholino-2-(phenylamino) furo[2,3-*d*]pyrimidine-5-carboxylate (**5e**). White crystals (yield, 90%). Mp: 144–146 °C. ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 1.40 (t, J = 7.2, CH_3), 2.61 (s, CH_3), 3.54 (t, J = 4.8, CH_2), 3.80 (t, J = 4.8, CH_2), 4.35 (q, J = 7.2, CH_3); 7.00–7.62 (m, 6H, Ar-H and ArN-H); IR (KBr): 3326 (N–H), 1713 (C=O), 1599, 1531, 1447, 1124, 746. MS (70 eV) *m*/*z* (%): 382 (M^+, 100), 337 (28), 324 (41), 222 (14), 118 (8), 77 (8). Anal. Calcd for $C_{20}H_{22}N_4O_4$ (382.41): C, 62.82; H, 5.80; N, 14.65. Found: C, 62.58; H, 5.79; N, 14.55. Ethyl 2-(*m*-tolylamino)-4-(butylamino)-6-methylfuro[2,3-*d*]pyrimidine-5-carboxylate (**5f**). White crystals (yield, 84%). Mp: 123–125 °C. ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 0.97 (t, J = 7.2, 3H, CH_3), 1.40 (t, J = 7.2, 3H, CH_3), 1.44–1.48 (m, 4H, CH_2CH_2), 2.34 (s, 3H, CH_3), 2.62 (s, 3H, CH_3), 3.53–3.56 (m, 2H, CH_2), 4.34 (t, J = 7.2, 2H, CH_2), 6.78–7.49 (m, 5H, Ar-H and ArN-H), 8.18 (s, 1H, N–H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ: 13.8, 14.1, 14.3, 14.7, 20.3, 21.6, 31.5, 40.7, 61.2, 92.6, 108.8, 115.8, 119.4, 122.4, 128.5, 138.3, 140.1, 156.0, 157.4, 165.5, 166.4. IR (KBr): 3378, 3286 (N–H), 1692 (C=O), 1628, 1532, 1415, 770. MS (70 eV) *m*/*z* (%): 382 (M^+, 100), 322 (34), 277 (15), 222 (16), 118 (7), 91 (5). Anal. Calcd for $C_{21}H_{26}N_4O_3$ (382.46): C, 65.95; H, 6.85; N, 14.65. Found: C, 65.89; H, 6.79; N, 14.55. Ethyl 2-(*m*-tolylamino)-6-methyl-4-morpholino-furo[2,3-*d*]pyrimidine-5-carboxylate (**5g**). White crystals (yield, 86%). Mp: 177–179 °C. ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 1.36 (t, J = 7.2, 3H, CH_3), 2.33 (s, 3H, CH_3), 2.59 (s, 3H, CH_3), 3.53 (t, J = 4.8, 4H, 2 × CH_2), 3.80 (t, J = 4.8, 4H, 2 × CH_2), 4.34 (q, J = 7.2, 2H, CH_2); 6.80–7.42 (m, 5H, Ar-H and ArN-H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ: 13.9, 14.2, 21.5, 48.9, 61.0, 66.4, 94.0, 109.2, 116.1, 119.6, 122.7, 128.4, 138.2, 139.7, 155.5, 155.9, 159.8, 163.8, 168.3. IR (KBr): 3336 (N–H), 1706 (C=O), 1549, 1531, 1442, 748. MS (70 eV) *m*/*z* (%): 396 (M^+, 100), 352 (19), 291 (14), 222 (21), 118 (6), 91 (7). Anal. Calcd for $C_{21}H_{24}N_4O_4$ (396.44): C, 63.62; H, 6.10; N, 14.13. Found: C, 63.56; H, 5.99; N, 14.04. Ethyl 2-(*m*-tolylamino)-6-methyl-4-(4-methylpiperazin-1-yl) furo[2,3-*d*]pyrimidine-5-carboxylate (**5h**). White crystals (yield, 80%). Mp: 148–150 °C. ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 1.36 (t, J = 7.2, 3H, CH_3), 2.30 (s, 3H, CH_3), 2.33 (s, 3H, CH_3), 2.48 (br s, 4H, 2 × CH_2), 2.58 (s, 3H, CH_3), 3.59 (br s, 4H, 2 × CH_2), 4.31 (q, J = 7.2, 2H, CH_2), 6.80–7.45 (m, 5H, Ar-H and ArN-H); IR (KBr): 3342 (N–H), 1699 (C=O), 1545, 1528, 740 cm^{-1}. MS (70 eV) *m*/*z* (%):409 (M^+, 100), 365 (24), 260 (35), 118 (14), 91 (5). Anal. Calcd for $C_{22}H_{27}N_5O_3$ (409.48): C, 64.53; H, 6.65; N, 17.10. Found: C, 64.50; H, 6.48; N, 17.01. Ethyl 2-(*m*-tolylamino)-4-(diethylamino)-6-methylfuro [2,3-*d*]pyrimidine-5-carboxylate (**5i**). White crystals (yield, 82%). Mp: 158–160 °C. ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 1.15 (t, J = 7.2, 6H, 2 × CH_3), 1.36 (t, J = 7.2, 3H, CH_3), 2.34 (s, 3H, CH_3), 2.55 (s, 3H, CH_3), 3.51 (q, J = 7.2, 4H, 2 × CH_2), 4.31 (q, J = 7.2, 2H, CH_2), 6.79–7.50 (m, 5H, Ar-H and ArN-H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ: 12.7, 13.3, 14.1, 21.3, 43.0, 60.8, 93.0, 109.7, 115.9, 119.4, 122.2, 128.1, 137.9, 140.0, 153.5, 155.8, 159.0, 164.2, 168.1. IR (KBr): 3364 (N–H), 1702 (C=O), 1545, 1530, 748. MS (70 eV) *m*/*z* (%): 382 (M^+, 100), 322 (34), 277 (15), 118 (21), 91 (8). Anal. Calcd for $C_{21}H_{26}N_4O_3$ (382.46): C, 65.95; H, 6.85; N, 14.65. Found: C, 65.86; H, 6.79; N, 14.58. Ethyl 2-(*m*-tolylamino)-4-(isopropylamino)-6-methylfuro[2,3-*d*]pyrimidine-5-carboxylate (**5j**). White crystals (yield, 85%). Mp: 168–170 °C. ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 1.32 (d, J = 7.2, 6H, 2 × CH_3), 1.40 (t, J = 7.2, 3H, CH_3), 2.34 (s, 3H, CH_3), 2.62 (s, 3H, CH_3), 4.35 (t, J = 7.2, 3H, CH and CH_2), 6.78–7.54 (m, 5H, Ar-H and ArN-H), 8.08 (s, 1H, N–H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ: 14.1, 14.7, 21.5, 22.5, 42.3, 61.1, 92.5, 108.7, 115.8, 119.4, 122.2, 128.4, 138.2, 140.2, 155.9, 156.6, 157.4, 165.4, 166.4. IR (KBr): 3288 (N–H), 1696 (C=O), 1618, 1532, 1418, 710. MS (70 eV) *m*/*z* (%): 368 (M^+, 100), 324 (34), 296 (12), 239 (25), 118 (6), 91 (7). Anal. Calcd for $C_{20}H_{24}N_4O_3$ (368.43): C, 65.20; H, 6.57; N, 15.21. Found: C, 65.14; H, 6.49; N, 15.08.
18. *X-ray crystal structure analysis for compound* (**5b**). Formula $C_{20}H_{24}N_4O_3$, colorless crystal. The crystal is of monoclinic, space group $P21/n$ with a = 8.0852(5) Å, b = 9.3902(6) Å, c = 25.3685(17) Å, β = 97.4950(10)°, V = 1909.6(2) Å^3, Z = 4, Dc = 1.282 g/cm^3, F(0 0 0) = 784, μ = 0.088 mm^{-1}, R = 0.0495 and wR = 0.1465 for 4748 observed reflections with $I > 2\sigma(I_0)$. X-ray structure showed that all ring atoms in the furo[2,3-*d*]pyrimidine moiety are nearly coplanar and the phenyl ring is twisted with respect to the pyrimidine ring system by 8.00(7)°. Crystallographic data for **5b** have been deposited in the Cambridge Crystallographic Data Center as supplementary publication numbers CCDC 778650. Copies of the data may be obtained, free of charge, on application to CCDC, 12 Union Road, Cambridge, CB2 1EZ, UK (fax: +44 1223 336033 or e-mail: deposit@ccdc.cam.ac.uk).
19. Cytotoxic activities were evaluated by using standard MTT assay after exposure of cells to the tested compounds for 48 h. Results are presented as mean values of two independent experiments done in quadruplicates. Coefficients of variation were <10%.