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A Mild and Efficient Copper-Mediated N-Arylation of 6-Azauracil with Corresponding Boronic Acids and their Antibacterial Activity

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A convenient synthetic strategy is reported for the synthesis of N-arylated 6-azauracil through Chan-Lam cross-coupling. A variety of N-arylated 6-azauracil derivatives were synthesized in moderate to good yields by cross-coupling of 6-azauracil with arylboronic acids in the presence of $Cu(OAc)_2$ and pyridine. However the arylation position is confirmed in the case of 2-(3,5-dichlorophenyl)-1,2,4-triazine-3,5(2*H*,4*H*)-dione (**3a**), by preparing the compound in a traditional method and compared the chemical shift of the NH proton at N-3 position. Further, the synthesized compounds were screened for their antibacterial activity using agar well diffusion method. The results revealed that most of the synthesized moieties possessing promising therapeutic nature.

Keywords: 6-Azauracil, N-arylation, Chan-Lam cross-coupling, Copper acetate, Antibacterial activity.

INTRODUCTION

Azauracils have been extensively studied for their clinical applications. For example, 6-azauracil and its derivatives have been extensively used in chemotherapy of cancer [1,2], inhibit viruses [3-5]. They are also used as a fungicide [6], chemotherapeutic agents for psoriasis [7], for polyarthritis [8] and for polycythemia vera [9]. It has been reported that 1-N position of 6-azauracil is served as the best location to place a substituent to mimic the size and shape of the natural nucleoside [10]. Attachment of substituted phenyl side chain at N-1 of 6-azauracil causes increasing its potency, which was related in part to the acidity of the imide hydrogen. Therefore, substituents attached at N-1 position will affect the pharmacokinetics and the binding properties of the drug. In addition, the polarizability of N-aryl-6-azauracil extends through out the molecule and leads adaptation to the active site [11], therefore N-aryl 6-azauracil derivatives are biologically active molecules [12-15] (Fig. 1).

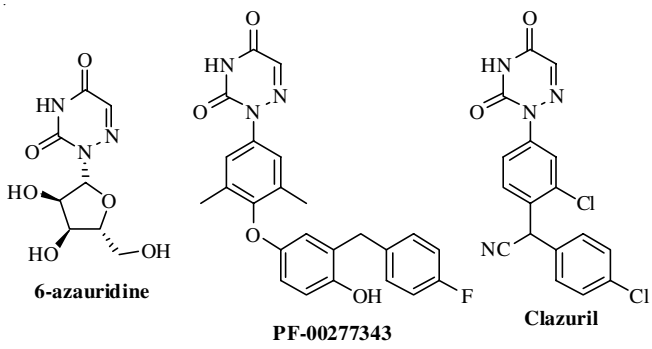
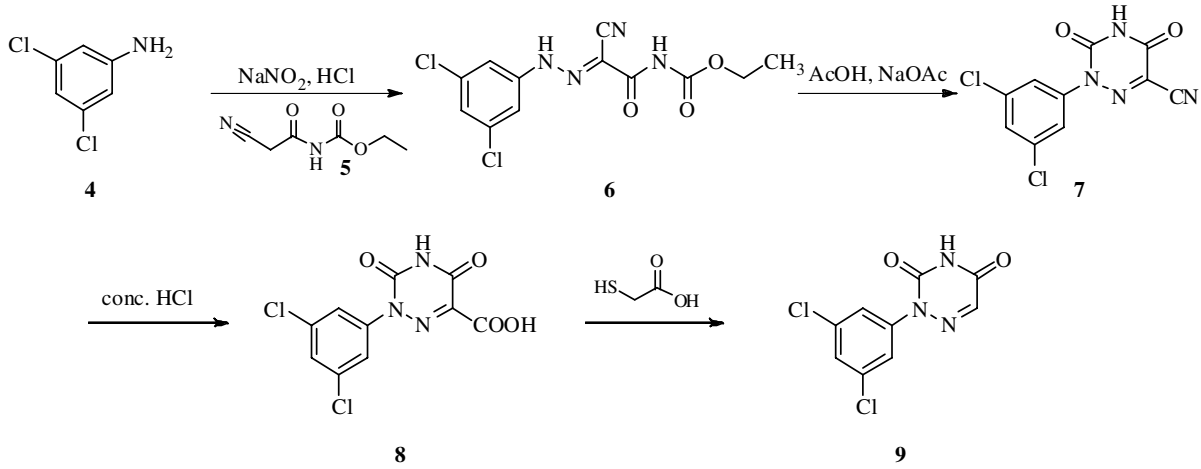


Fig. 1. Some biologically active N-aryl 6-azauracil derivatives

Traditionally N-aryl 6-azauracil derivatives were synthesized [16] from corresponding anilines in 4 steps as shown in **Scheme-I**. However, used harsh reaction conditions and laborious workup process to get the pure material. There are reports [17-19] about Chan-Lam cross-coupling of N-3 protected 6-azauracil and arylboronic acid in order to get the specific arylation at N-1 position. Recently copper catalyzed Unman coupling for N-arylation has been reported [14], the method uses expensive diamine catalyst systems, high reaction temperatures (170 °C) and inert conditions. In order to overcome all the synthetic issues, we here report a simple, feasible method to synthesize a range of N-aryl 6-azauracil derivatives



Scheme-I: Traditional scheme for N-aryl 6-azauracil. Reagents and conditions: (a) 6 N HCl (10v), $NaNO_2$ (1.0 equiv), 5 (1.0 equiv), Py (10v), 5 °C to room temp., 1 h; 86 % (b) NaOAc (1.0 equiv), AcOH (10v), 120 °C, 2 h; 85 % (c) 6N HCl in 1,4-dioxane (10v), 110 °C, 16 h; (71 %) (d) 9 (1.0 equiv), tolune, 120 °C, 2 h; 83 %

using copper-mediated Chan-Lam coupling from N-3 unprotected 6-azauracil and corresponding arylboronic acid.

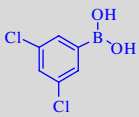
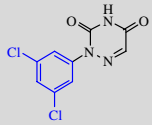
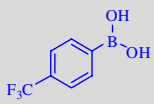
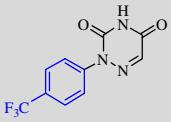
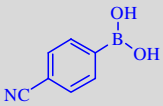
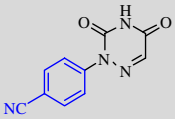
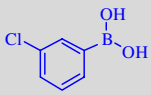
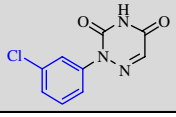
Copper-mediated Chan-Lam coupling reactions play an important role in heteroatom arylation and hetero arylation [20,21]. The development of this new methodology has attracted much attention as the reactions proceeds at lower temperature in open air and works for many hetero arenes and aryl boronic acids provides good yield of N-arylated heteroarenes. Therefore, by adopting Chan-Lam cross-coupling protocol, a variety of N-aryl 6-azauracil derivatives were synthesized and presenting for the first time.

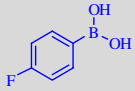
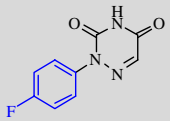
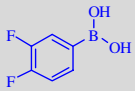
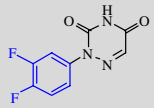
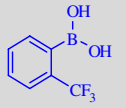
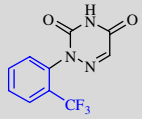
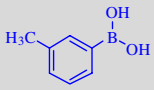
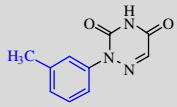
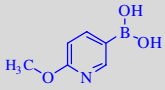
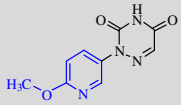
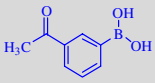
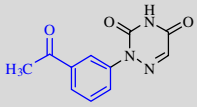
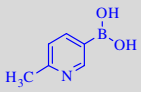
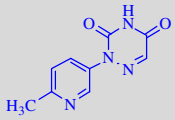
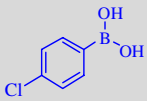
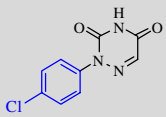
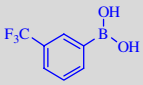
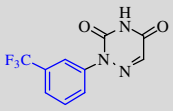
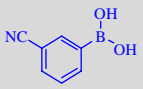
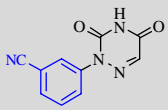
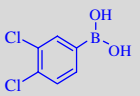
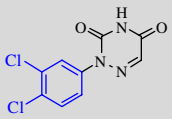
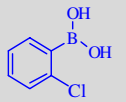
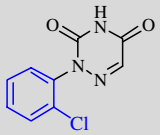
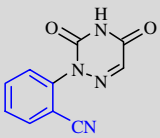
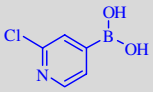
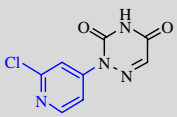
EXPERIMENTAL

All chemicals were purchased from Sigma-Aldrich; Merck and Combi blocks were used without further purification. All melting points were uncorrected and determined in one end open capillary tubes using Guna Digital Melting Point apparatus. ^{1}H NMR and ^{13}C NMR spectra were recorded on a Bruker AMX 300 spectrometer operating at 400/300 MHz for ^{1}H and 100 MHz for ^{13}C NMR. The ^{1}H NMR and ^{13}C NMR chemical shifts were expressed in ppm with reference to tetramethylsilane. High resolution mass spectroscopy (HRMS) data were recorded on high resolution micr OTOF instrument with electrospray ionization (ESI). Elemental analyses were performed on a Perkin-Elmer 240 CHN elemental analyzer.

General procedure for the synthesis of title compounds (3a-r): To a solution of 6-azauracil (100 mg, 0.88 mmol) in DMF (10.0 mL) was added base (1.76 mmol) and $Cu(OAc)_2$ (159 mg, 0.88 mmol) at room temperature. The resulting reation mixture was degassed with oxygen for 10 min and then added arylboronic acids (0.96 mmol) at room temperature and stirred at appropriate temperature (Table-1) under oxygen atmosphere. The reaction mixture was diluted with water (15 mL) and extracted with dichloromethane (3 × 15 mL). The organic layer washed with H_2O (15 mL), brine solution (15 mL), dried over Na_2SO_4 and concentrated. The obtained crude product was purified by column chromatography (0 to 10 % CH_3OH/CH_2Cl_2) to afford the title compounds.

TABLE-1
PROTOCOL FOR THE SYNTHESIS OF THE TITLE COMPOUNDS (**3a-r**)

Entry	Substrate (**2**)	Product	Temp. (°C)	Base	Time (h)	Yield (%)
3a			RT	Pyridine	2	81
3b			65	Et_3N	4	62
3c			65	Et_3N	4	68
3d			RT	Pyridine	2	79

3e			RT	Pyridine	2	77
3f			RT	Pyridine	2	90
3g			65	Et_3N	4	70
3h			RT	Pyridine	2	80
3i			65	Et_3N	4	72
3j			65	Et_3N	4	64
3k			65	Et_3N	4	60
3l			RT	Pyridine	2	70
3m			65	Et_3N	4	69
3n			65	Et_3N	4	73
3o			RT	Pyridine	2	75
3p			65	Et_3N	4	75
3q			65	Et_3N	4	60
3r			80	Et_3N	6	65

Spectral data for compounds 3a-r

2-(3,5-Dichlorophenyl)-1,2,4-triazine-3,5(2*H*,4*H*)-dione (3a) [22]**:** Off white solid; m.p.: 142-147 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 12.75 (s, 1H), 7.76-7.75 (m, 1H), 7.65 (s, 1H), 7.557 (s, 1H), 7.551 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 155.7, 148.4, 135.0, 134.1, 132.6, 130.2, 128.50, 128.25, 127.12; HRMS (ESI) *m/z*: calcd. for $C_9H_5N_3O_2Cl_2$ $[M+H]^+$ 256.9754, found 256.9748; Anal. Calcd. $C_9H_5N_3O_2Cl_2$: C, 41.89; H, 1.95; N, 16.28; Found: C, 41.86; H, 1.98; N, 16.25.

2-(4-(Trifluoromethyl)phenyl)-1,2,4-triazine-3,5(2*H*,4*H*)-dione (3b): Off white solid; m.p.: 131-135 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.72 (s, 1H), 7.89 (d, 2H, *J* = 8.4 Hz), 7.64 (s, 1H), 7.61 (d, 2H, *J* = 8.0 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 156.0, 148.7, 136.9, 135.4, 129.0, 126.0; HRMS (ESI) *m/z:* calcd. for $C_{10}H_6N_3O_2F_3$ $[M+H]^+$ 257.0408, found 257.0422; Anal. Calcd. $C_{10}H_6N_3O_2F_3$: C, 46.70; H, 2.35; N, 16.34; Found: C, 46.74; H, 2.37; N, 16.37.

4-(3,5-Dioxo-4,5-dihydro-1,2,4-triazin-2(3*H*)-yl)benzonitrile (3c) [23]**:** Off white solid; m.p.: 293-297 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 12.73 (s, 1H), 8.00 (d, 2H, *J* = 8.4 Hz), 7.65 (s, 1H), 7.59 (d, 2H, *J* = 8.7 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 155.9, 148.5, 137.5, 135.4, 133.0, 129.8, 118.2, 111.6; HRMS (ESI) *m/z:* calcd. for $C_{10}H_6N_4O_2$ $[M+H]^+$ 214.0486, found 214.0481; Anal. Calcd. $C_{10}H_6N_4O_2$: C, 56.08; H, 2.82; N, 26.16; Found: C, 56.11; H, 2.87; N, 26.12.

2-(3-Chlorophenyl)-1,2,4-triazine-3,5(2*H*,4*H*)-dione (3d) [11]**:** Off white solid; m.p.: 212-215 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.69 (s, 1H), 7.62 (s, 1H), 7.53-7.37 (m, 3H), 7.36-7.31 (m, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 156.1, 148.7, 135.4, 134.5, 132.9, 130.5, 128.8, 128.5, 127.4; HRMS (ESI) *m/z:* calcd. for $C_9H_6N_3O_2Cl$ $[M+H]^+$ 223.0143, found 223.0134; Anal. Calcd. $C_9H_6N_3O_2Cl$: C, 48.34; H, 2.70; N, 18.79; Found: C, 48.38; H, 2.76; N, 18.74.

2-(4-Fluorophenyl)-1,2,4-triazine-3,5(2*H*,4*H*)-dione (3e) [11]**:** Off white solid; m.p.: 271-276 °C: ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.67 (s, 1H), 7.60 (s, 1H), 7.39-7.37 (m, 2H), 7.33-7.31 (m, 2H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 163.0, 160.5, 156.3, 148.9, 135.3, 130.6, 130.5, 115.9, 115.6; HRMS (ESI) *m/z:* calcd. for $C_9H_6N_3O_2F$ $[M+H]^+$ 207.0438, found 207.0439; Anal. Calcd. $C_9H_6N_3O_2F$: C, 52.18; H, 2.92; N, 20.28; Found: C, 52.15; H, 2.96; N, 20.25.

2-(3,4-Difluorophenyl)-1,2,4-triazine-3,5(2*H*,4*H*)-dione (3f): Off white solid; m.p.: 234-237 °C: ^{1}H NMR (300 MHz, DMSO-d_6) δ 12.71 (s, 1H), 7.63 (s, 1H), 7.60-7.56 (m, 1H), 7.28-7.24 (m, 2H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 156.1, 148.7, 136.4, 135.3, 126.07, 126.04, 126.01, 125.9, 118.3, 118.1, 117.7, 117.5; HRMS (ESI) *m/z:* calcd. for $C_9H_5N_3O_2F_2$ $[M+H]^+$ 225.0345, found 225.0341; Anal. Calcd. $C_9H_5N_3O_2F_2$: C, 48.01; H, 2.24; N, 18.66; Found: C, 48.05; H, 2.26; N, 18.61.

2-(2-(Trifluoromethyl)phenyl)-1,2,4-triazine-3,5(2*H*,4*H*)-dione (3g): Off white solid; m.p.: 127-132 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.76 (s, 1H), 7.71-7.58 (m, 3H), 7.46-7.29 (m, 2H); ^{13}C NMR (100 MHz, DMSO-d_6) δ; 155.2, 147.8, 135.4, 134.4, 128.78, 128.74, 128.66, 125.15, 125.12, 125.09, 125.05, 121.76, 121.73, 121.70, 117.41, 117.22, 116.84, 116.66; HRMS (ESI) *m/z:* calcd. for $C_{10}H_6N_3O_2F_3$ $[M+H]^+$ 257.0407, found 257.0412; Anal. Calcd. $C_{10}H_6N_3O_2F_3$: C, 46.70; H, 2.35; N, 16.34; Found: C, 46.74; H, 2.37; N, 16.37.

2-*m*-Tolyl-1,2,4-triazine-3,5(2*H*,4*H*)-dione (3h) [24]**:** Off white solid; m.p.: 134-136 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 12.63 (s, 1H), 7.59 (s, 1H), 7.38-7.33 (m, 1H), 7.24 (d, 1H, *J* = 7.8 Hz), 7.12-7.09 (m, 2H), 2.33(s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 156.3, 148.9, 138.4, 135.4, 133.0, 129.3, 128.7, 128.6, 125.3, 20.72; HRMS (ESI) *m/z:* calcd. for $C_{10}H_9N_3O_2$ $[M+H]^+$ 203.0690, found 203.0678; Anal. Calcd. $C_{10}H_9N_3O_2$: C, 59.11; H, 4.46; N, 20.68; Found: C, 59.15; H, 4.48; N, 20.64.

2-(6-Methoxypyridin-3-yl)-1,2,4-triazine-3,5(2*H*,4*H*)-dione (3i): Off white solid; m.p.: 230-234 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.69 (s, 1H), 8.13 (d, 1H, *J* = 2.4 Hz), 7.68 (dd, 1H, *J* = 11.2 Hz, 2.4 Hz), 7.63 (s, 1H), 6.95 (d, 1H, *J* = 8.8 Hz), 3.88 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 163.1, 156.3, 149.0, 146.2, 139.3, 135.2, 123.7, 110.6, 53.55; HRMS (ESI) *m/z:* calcd. for $C_9H_8N_4O_3$ $[M+H]^+$ 220.0592, found 220.0572; Anal. Calcd. $C_9H_8N_4O_3$: C, 49.09; H, 3.66; N, 25.45; Found: C, 49.13; H, 3.63; N, 25.49.

2-(3-Acetylphenyl)-1,2,4-triazine-3,5(2*H*,4*H*)-dione (3j): Off white solid; m.p.: 174-177 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.69 (s, 1H), 8.05-8.03 (m, 1H), 8.02 (d, 1H, *J* = 1.6 Hz), 7.95 (t, 1H, *J* = 4.0 Hz), 7.68-7.62 (m, 2H), 2.59 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 155.2, 147.9, 137.4, 134.4, 132.0, 128.3, 127.7, 127.6, 124.3, 19.71; HRMS (ESI) *m/z:* calcd. for $C_{11}H_9N_3O_3$ $[M+H]^+$ 231.0638, found 231.0619; Anal. Calcd. $C_{11}H_9N_3O_3$: C, 57.14; H, 3.92; N, 18.17; Found: C, 57.17; H, 3.96; N, 18.12.

2-(6-Methylpyridin-3-yl)-1,2,4-triazine-3,5(2*H*,4*H*)-dione (3k): Off white solid; m.p.: 173-177 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.68 (s, 1H), 8.11 (d, 1H, *J* = 2.4 Hz), 7.67 (dd, 1H, *J* = 11.2 Hz, 2.4 Hz), 7.61 (s, 1H), 6.93 (d, 1H, *J* = 8.8 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 162.7, 155.9, 148.6, 145.8, 138.9, 134.8, 123.2, 110.2, 53.15; HRMS (ESI) *m/z:* calcd. for $C_9H_8N_4O_2$ $[M+H]^+$ 204.0643, found 204.0637; Anal. Calcd. $C_9H_8N_4O_2$: C, 52.94; H, 3.95; N, 27.44; Found: C, 52.98; H, 3.93; N, 27.48.

2-(4-Chlorophenyl)-1,2,4-triazine-3,5(2*H*,4*H*)-dione (3l) [11]**:** Off white solid; m.p.: 231-235 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 12.56 (s, 1H), 7.82 (d, 2H, *J* = 8.4 Hz), 7.48 (s, 1H), 7.42 (d, 2H, *J* = 8.0 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 155.3, 147.9, 136.1, 134.6, 128.89, 128.59, 128.27, 125.32, 125.28, 124.5, 121.8; HRMS (ESI) *m/z:* calcd. for $C_9H_6N_3O_2Cl$ $[M+H]^+$ 223.0144, found 223.0124; Anal. Calcd. $C_9H_6N_3O_2Cl$: C, 48.34; H, 2.70; N, 18.79; Found: C, 48.38; H, 2.76; N, 18.74.

2-(3-(Trifluoromethyl)phenyl)-1,2,4-triazine-3,5(2*H*,4*H*)-dione (3m) [11]**:** Off white solid; m.p.: 123-127 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 12.50 (s, 1H), 7.93-7.85 (m, 2H), 7.77-7.72 (m, 2H), 7.67 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 157.1, 149.7, 137.9, 136.4, 130.7, 130.4, 130.0, 127.1, 127.0,126.7, 126.3, 123.6; HRMS (ESI) *m/z:* calcd. for $C_{10}H_6N_3O_2F_3$ $[M+H]^+$ 257.0407, found 257.0412; Anal. Calcd. $C_{10}H_6N_3O_2F_3$: C, 46.70; H, 2.35; N, 16.34; Found: C, 46.74; H, 2.37; N, 16.37.

3-(3,5-Dioxo-4,5-dihydro-1,2,4-triazin-2(3*H*)-yl)-benzonitrile (3n) [11]**:** Off white solid; m.p.: 196-199 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.78 (s, 1H), 7.71 (s, 1H), 7.61-7.60 (m, 3H), 7.43-7.41 (m, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 156.4, 149.1, 138.0, 136.0, 135.1, 133.6, 131.5, 130.4, 118.8, 112.2; HRMS (ESI) *m/z:* calcd. for $C_{10}H_6N_4O_2$ $[M+H]^+$ 214.0486, found 214.0474; Anal. Calcd. $C_{10}H_6N_4O_2$: C, 56.08; H, 2.82; N, 26.16; Found: C, 56.11; H, 2.87; N, 26.12.

2-(3,4-Dichlorophenyl)-1,2,4-triazine-3,5(2*H*,4*H*)-dione (3o) [11]**:** Off white solid; m.p.: 224-227 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 12.78 (s, 1H), 7.79-7.78 (m, 1H), 7.68 (s, 1H), 7.59-7.58 (m, 2H); ^{13}C NMR (100 MHz, DMSO-

d_6) δ 155.4, 148.0, 134.7, 133.8, 132.2, 129.8, 128.1, 127.8, 126.7; HRMS (ESI) *m/z:* calcd. for $C_9H_5N_3O_2Cl_2$ $[M+H]^+$ 256.9754, found 256.9743; Anal. Calcd. $C_9H_5N_3O_2Cl_2$: C, 41.89; H, 1.95; N, 16.28; Found: C, 41.86; H, 1.98; N, 16.25.

2-(2-Chlorophenyl)-1,2,4-triazine-3,5(2*H*,4*H*)-dione (3p): Off white solid; m.p.: 213-216 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 12.39 (s, 1H), 7.81-7.73 (m, 2H), 7.66-7.61 (m, 2H), 7.56 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 155.2, 147.8, 134.4, 133.6, 132.0, 129.6, 127.9, 127.6, 126.5; HRMS (ESI) *m/z:* calcd. for $C_9H_6N_3O_2Cl$ $[M+H]^+$ 223.0144, found 223.0149; Anal. Calcd. $C_9H_6N_3O_2Cl$: C, 48.34; H, 2.70; N, 18.79; Found: C, 48.38; H, 2.76; N, 18.74.

2-(3,5-Dioxo-4,5-dihydro-1,2,4-triazine-2(3*H*)-yl)-benzonitrile (3q): Off white solid; m.p.: 196-199 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 12.47 (s, 1H), 7.90-7.82 (m, 2H), 7.74-7.64 (m, 2H), 7.63 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 154.1, 146.8, 135.7, 133.7, 132.8, 131.3, 128.1, 116.5, 109.9; HRMS (ESI) *m/z:* calcd. for $C_{10}H_6N_4O_2$ $[M+H]^+$ 214.0486, found 214.0481; Anal. Calcd. $C_{10}H_6N_4O_2$: C, 56.08; H, 2.82; N, 26.16; Found: C, 56.11; H, 2.87; N, 26.12.

2-(2-Chloropyridin-4-yl)-1,2,4-triazine-3,5(2*H*,4*H*)-dione (3r): Off white solid; m.p.: 202-205 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.66 (s, 1H), 8.10 (d, 1H, J = 2.4 Hz), 7.66 (dd, 1H, J = 11.2 Hz, 2.4 Hz), 7.59 (s, 1H), 6.91 (d, 1H, J = 8.8 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 163.3, 156.6, 149.2, 146.4, 139.5, 123.9, 110.8, 53.77; HRMS (ESI) *m/z:* calcd. for $C_8H_5N_4O_2Cl$ $[M+H]^+$ 224.0096, found 224.0072; Anal. Calcd. $C_8H_5N_4O_2Cl$: C, 42.78; H, 2.24; N, 24.94; Found: C, 42.75; H, 2.27; N, 24.91.

Experimental procedure for the compound **9** in a traditional method.

(E)-Ethyl 2-cyano-2-(2-(3,5-dichlorophenyl)hydrazono)-acetylcarbamate (6): 6 N Hydrochloric acid (10 mL) was added to 3,5-dichloroaniline (**4**) (1 g, 6.21 mmol) at a 10 °C. The resulting slurry was cooled to 5 °C in an ice-bath and a solution of sodium nitrite (0.42 g, 6.21 mmol) in water (2 mL) was added at such a rate that the temperature does not rise above 5 °C. Ethyl 2-cyanoacetylcarbamate (**5**) (0.96 g, 6.21 mmol) and pyridine (10.0 mL) was added portion wise to the resulting solution of the diazonium salt with vigorous stirring at 5 °C and reaction mixture was raised to 5 °C and stirred at same temperature for 1 h. The solids were filtered and washed thoroughly with water to remove alkaline impurities. The crude product was recrystallized from toluene to afford (E)-ethyl 2-cyano-2-(2-(3,5-dichlorophenyl)hydrazono)acetylcarbamate (**6**) as an off white solid. (1.75 g, 86 %). m.p.: 194-196 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 12.16 (bs, 1H), 10.94 (s, 1H), 7.96-7.72 (m, 3H), 4.20 (q, 2H), 1.26 (t, 3H, J = 7.2 Hz); MS (MM) *m/z* 326.9 $[M–H]^–$; Anal. Calcd. $C_{12}H_{10}N_4O_3Cl_2$: C, 43.79; H, 3.06; N, 17.02; Found: C, 43.76; H, 3.09; N, 17.05.

2-(3,5-Dichlorophenyl)-3,5-dioxo-2,3,4,5-tetrahydro-1,2,4-triazine-6-carbonitrile (7): To a solution of (E)-ethyl 2-cyano-2-(2-(3,5-dichlorophenyl)hydrazono)acetylcarbamate (**6**) (1.50 g, 4.57 mmol) in acetic acid (15 mL) was added sodium acetate (0.374 g, 4.57 mmol). The resulting reaction mixture was stirred at 120 °C for 2 h. After completion of starting material by TLC analysis (90:10 dichloromethane/methanol) the reaction mixture was poured into water. The solids were filtered and washed with water. The crude product was recrystallized from ethanol to afford the 2-(3,5-dichlorophenyl)-3,5-dioxo-2,3,4,5-tetrahydro-1,2,4-triazine-6-carbonitrile (**7**) as an off white solid. (1.10 g, 85 %). m.p.: 234-236 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 13.32 (bs, 1H), 7.78-7.74 (m, 3H); MS (MM) *m/z* 280.9 $[M–H]^–$; Anal. Calcd. $C_{10}H_4N_4O_2Cl_2$: C, 42.43; H, 1.42; N, 19.79; Found: C, 42.40; H, 1.45; N, 19.74.

2-(3,5-Dichlorophenyl)-3,5-dioxo-2,3,4,5-tetrahydro-1,2,4-triazine-6-carboxylic acid (8): 6 N HCl in 1,4-dioxane (10 mL) was added to the 2-(3,5-dichlorophenyl)-3,5-dioxo-2,3,4,5-tetrahydro-1,2,4-triazine-6-carbonitrile (**7**) (1.00 g, 3.54 mmol) at room temperature. The resulting suspension was stirred for 110 °C for 16 h. After completion of starting material by TLC analysis (90:10 dichloromethane/methanol) the reaction mixture was poured into water. The solids were filtered and washed with water. The crude product was recrystallized from ethanol to afford the 2-(3,5-dichlorophenyl)-3,5-dioxo-2,3,4,5-tetrahydro-1,2,4-triazine-6-carboxylic acid (**8**) as an off white solid. (0.76 g, 71 %). m.p.: 257-259 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 7.89-7.85 (m, 2H), 7.74-7.72 (m, 1H); MS (MM) *m/z* 299.9 $[M–H]^–$; Anal. Calcd. $C_{10}H_5N_3O_4Cl_2$: C, 39.76; H, 1.67; N, 13.91; Found: C, 39.72; H, 1.63; N, 13.95.

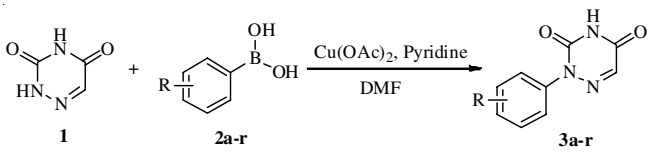
2-(3,5-Dichlorophenyl)-1,2,4-triazine-3,5(2*H*,4*H*)-dione (9): To a solution of 2-(3,5-dichlorophenyl)-3,5-dioxo-2,3,4,5-tetrahydro-1,2,4-triazine-6-carboxylic acid (**8**) (0.50 g, 1.66 mmol) in toluene (5.0 mL) was added 2-mercaptoacetic acid (0.10 mL) at room temperature. The resulting solution was stirred for 120 °C for 2 h. After completion of starting material by TLC analysis (80:20 hexanes/ethyl acetate) then the reaction mixture was poured into water. The solids were filtered and washed with water. The crude product was recrystallized from toluene to afford to afford the 2-(3,5-dichlorophenyl)-1,2,4-triazine-3,5(2*H*,4*H*)-dione (**9**) as an off white solid. (0.35 g, 83 %). m.p.: 142-146 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 12.75 (s, 1H), 7.76-7.75 (m, 1H), 7.65 (s, 1H), 7.557 (s, 1H), 7.551 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 155.7, 148.4, 135.0, 134.1, 132.6, 130.2, 128.50, 128.25, 127.12; HRMS (ESI) *m/z:* calcd. for $C_9H_5N_3O_2Cl_2$ $[M+H]^+$ 256.9754, found 256.9742; Anal. Calcd. $C_9H_5N_3O_2Cl_2$: C, 41.89; H, 1.95; N, 16.28; Found: C, 41.86; H, 1.98; N, 16.25

Antibacterial activity: Antibacterial activity of all the synthesized compounds was screened by Agar well diffusion method, as recommended by the National Committee for Clinical Laboratory standards against Gram-positive bacteria such as *Staphylococcus aureus* (NCIM 2079) and Gram-negative bacteria *Escherichia coli* (NCIM 2068) at two concentrations 100 and 200 μg/mL. The obtained results were compared with standard drug chloramphenicol. The fresh culture of bacteria is obtained by inoculating bacteria into nutrient broth media and incubated at 37 °C for 24 h. This culture mixed with nutrient agar media was poured into petri dishes under aseptic conditions. After solidification of media, bores are made by using sterile cork borer (8 mm diameter). Into these cups, standard drug and synthesized drugs are introduced; the plates were placed in the refrigerator at 100 °C for proper diffusion of drugs into the media. After 2 h, the petri plates were transferred to an incubator and maintained at 37 ± 2 °C for 24 h. After the

incubation period, the petri plates were observed for the zone of inhibition. The results are evaluated by comparing the zone of inhibition shown by the synthesized compounds with standard drug.

RESULTS AND DISCUSSION

The method used to synthesize derivatives of 6-azauracil was shown in **Scheme-II**. Initially, the method was optimized by the treatment of 6-azauracil with 3,5-dichlorophenyl boronic acid in the presence of various copper sources, bases in DMF as shown in Table-2. Among all of the conditions screened, $Cu(OAc)_2$ gives mono arylated product of **3a** in good yield. In addition, yield was improved furthermore when pyridine was used as a base. With the higher equivalents of boronic acid (~ 4 equiv) and increasing reaction temperature and time (~ 12 h) resulted in large quantities of diaryl substitution. To avoid diaryl products, boronic acid equivalents reduced to 1.1, reaction temperature to room temperature and reaction time to 2 h.



Scheme-II: Reaction of 6-azauracil with phenylboronic acid

TABLE-2
OPTIMIZATION OF CHAN-LAM COUPLING TO ACCESS N-ARYL 6-AZAURACIL[a]

Entry	Copper salt	Base	Yield[b]
1	CuI	Et_3N	15 %
2	CuI	Pyridine	18 %
3	CuBr	Et_3N	N.R
4	CuBr	Pyridine	N.R
5	$CuSO_4$	Et_3N	N.R
6	$CuSO_4$	Pyridine	N.R
7	$Cu(OAc)_2$	Et_3N	80 %
8	$Cu(OAc)_2$	Pyridine	81 %

[a]Reactions were performed with 6-azauracil **1** (1.0 mmol), Copper salt (1.0 mmol), base (2.0 mmol), arylboronic acid **2** (1.1 mmol), DMF (10 mL) at room temperature for 2 h; [b]Isolated yield; N.R. = No reaction

In this method confirmation of exact position of arylation is crucial as there is a chance of N-arylation at 3-N position. In order to confirm the substituted position, we have synthesized compound **3a** using a traditional method as shown in **Scheme-I**, where 3,5-dichloroaniline was treated with sodium nitrite in presence of 6 N HCl to get the diazonium salt which was then treated with ethyl (2-cyanoacetyl)carbamate (**5**) to get the intermediate **6**. The intermediate **6** was refluxed in acetic acid in presence of sodium acetate to afford 1-(3,5-dichlorophenyl)-5-cyano-6-azauracil (**7**). The cyano group of compound **7** was hydrolyzed with 6N HCl in 1,4-dioxane to get the acid compound **8**. Decarboxylation of compound **8** was carried out in 2-mercaptoacetic acid to afford the 1-(3,5-dichlorophenyl)-6-azauracil (**3a**). The NH proton at N-3 position of 6-azouracils prepared in both routes has showed same δ ppm values in ^{1}H NMR at δ12.6 ppm, which has confirmed unambiguously the compound **3a** prepared in both the routes are same.

By utilizing the conditions that are employed to synthesize **3a**, as shown in **Scheme-II**, several N-aryl 6-azouracils were synthesized and showed in Table-1. Electron donating substituents on boronic acid afforded N-arylated products in good yields at room temperature and pyridine as base, electron-withdrawing substituents on boronic acids required 80 °C and triethyl amine as base in order to proceed the reaction and afford in relatively good yields.

The synthesized compounds were screened for their antibacterial activity using Agar well diffusion method [25] against Escherichia coli (Gram-negative) and Staphylococcus aureus (Gram-positive) strains. The screening data are shown in Table-3 indicated that the compounds **3b**, **3g** and **3m** bearing trifluromethyl group showed significant antibacterial activity. The introduction of chloro moiety on phenyl ring **3a**, **3d**, **3l** and **3o** exhibited more activity than fluorine containing compounds **3e** and **3f**. Remaining compounds were found to possess moderate activity against Gram-positive and Gram-negative strains when compared with standard drug chloramphenicol.

TABLE-3
ANTIBACTERIAL ACTIVITY OF THE TITLE COMPOUNDS (**3a-3r**)

Compound	Zone of inhibition (mm)			
	Escherichia coli Gram-negative) (conc. µg/mL)		*Staphylococcus aureus* (Gram-positive) (conc. µg/mL)	
	100	200	100	200
3a	16	19	18	23
3b	26	29	23	27
3c	9	10	11	15
3d	17	19	18	21
3e	13	15	14	15
3f	14	17	15	18
3g	25	28	22	26
3h	12	15	8	17
3i	8	12	10	16
3j	7	13	9	15
3k	12	14	14	19
3l	18	21	19	20
3m	24	28	23	27
3n	8	13	10	13
3o	15	17	18	25
3p	16	19	17	21
3q	11	15	13	15
3r	10	16	12	13
Chloramphenicol	27	30	25	29

Conclusion

An efficient and mild synthetic procedure for the synthesis of N-aryl 6-azauracil derivatives starting from corresponding boronic acids was described and the position of N-arylation for obtained N-aryl 6-azouracils were confirmed by ^{1}H NMR spectra of the same compound that was synthesized in traditional method. As it is a convenient and cost-effective method, this can be utilized in the manipulations of 6-azauracil derivatives to develop drugs based on azauracils. The synthesized compounds were screened for their antibacterial activity using agar well diffusion method against Escherichia coli and *Staphylococcus aureus* strains. Most of the tested compounds have shown moderate to good antibacterial activity.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this article.

REFERENCES

1. R.E. Handschumacher, P. Calabresi, A.D. Welch, V. Bono, H. Fallon and E. Frei, *Cancer Chemother. Rep.*, **21**, 1 (1962).
2. T.R. Walters, R.J.A. Aur, K. Hernandez, T. Vietti and D. Pinkel, *Cancer*, **29**, 1057 (1972); https://doi.org/10.1002/1097-0142(197204)29:4<1057::AID-CNCR2820290455>3.0.CO;2-Q.
3. B. Rada and D. Blaskovic, *Acta Virol.*, **10**, 1 (1966).
4. D. Falke and B. Rada, *Acta Virol.*, **14**, 115 (1970).
5. I. Alexeeva, L. Palchikovskaya, A. Shalamay, L. Nosach, V. Zhovnovataya, O. Povnitsa and N. Dyachenko, *Acta Biochim. Polon.*, **47**, 95 (2000).
6. Y. Kabbaj, H.B. Lazrek, J.L. Barascut and J.L. Imbach, *Nucleos. Nucl. Nucl. Acids*, **24**, 161 (2005); https://doi.org/10.1081/NCN-55695.
7. P. Calabresi and R.W. Turner, *Ann. Intern. Med.*, **64**, 352 (1966); https://doi.org/10.7326/0003-4819-64-2-352.
8. R. Gunther, *Wien. Med. Wochenschr.*, **117**, 985 (1967).
9. R.C. Deconti and P. Calabresi, *Ann. Intern. Med.*, **73**, 575 (1970); https://doi.org/10.7326/0003-4819-73-4-575.
10. B.L. Mylari, M.W. Miller, H.L. Howes, S. Figdor, J.E. Lynch and R.C. Koch, *J. Med. Chem.*, **20**, 475 (1977); https://doi.org/10.1021/jm00214a003.
11. M.W. Miller, B.L. Mylari, H.L. Howes, J.E. Lynch, M.J. Lynch and R.C. Koch, *J. Med. Chem.*, **22**, 1483 (1979); https://doi.org/10.1021/jm00198a010.
12. J.W. St Geme and H.L. Martin, *Infect. Immun.*, **11**, 915 (1975).
13. K.A. Steffenhagen, B.C. Easterday and G.J. Galasso, *J. Infect. Dis.*, **133**, 603 (1976); https://doi.org/10.1093/infdis/133.6.603.
14. J.J. Li, L.H. Mitchell and R.L. Dow, *Bioorg. Med. Chem. Lett.*, **20**, 306 (2010); https://doi.org/10.1016/j.bmcl.2009.10.109.
15. O. Vanparijs, L. Hermans, L. Van der Flaes, K. Vlaminck and R. Marsboom, *Tijdschr. Diergeneeskd.*, **113**, 190 (1988).
16. J. Slouka, *Monatsh. Chem.*, **96**, 134 (1965); https://doi.org/10.1007/BF00912302.
17. E. Riguet, B. Campo, A. Gibelin and K. Mhalla, Novel Triazinedione Derivatives as GABA-β receptor Modulators, WO2008056257 A2 (2008).
18. I. Leroy, E. Dupont-Passelaigue, K. Valeille, Y. Rival and D. Junquero, Derivatives of Triazines and Uracils, Their Preparation and Their Application in Human Therapeutics, WO2010006962 A1 (2010).
19. Z. Rankovic, J. Cai, X. Fradera, M. Dempster, A. Mistry, A. Mitchell, C. Long, E. Hamilton, A. King, S. Boucharens, C. Jamieson, J. Gillespie, I. Cumming, J. Uitdehaag and M.V. Zeeland, *Bioorg. Med. Chem. Lett.*, **20**, 1488 (2010); https://doi.org/10.1016/j.bmcl.2010.01.116.
20. P.Y.S. Lam, C.G. Clark, S. Saubern, J. Adams, M.P. Winters, D.M.T. Chan and A. Combs, *Tetrahedron Lett.*, **39**, 2941 (1998); https://doi.org/10.1016/S0040-4039(98)00504-8.
21. D.M.T. Chan, K.L. Monaco, R.P. Wang and M.P. Winters, *Tetrahedron Lett.*, **39**, 2933 (1998); https://doi.org/10.1016/S0040-4039(98)00503-6.
22. J.W. Mcfarland, C.B. Cooper and D.M. Newcomb, *J. Med. Chem.*, **34**, 1908 (1991); https://doi.org/10.1021/jm00110a023.
23. M.W. Miller, 2-Phenyl-as-triazine-3,5(2H,4H)diones, US Patent 3912723 A (1975).
24. M.W. Miller, B.L. Mylari, H.L. Howes, S.K. Figdor, M.J. Lynch, J.E. Lynch and R.C. Koch, *J. Med. Chem.*, **23**, 1083 (1980); https://doi.org/10.1021/jm00184a005.
25. E. Shivarani, R. Parameswar, V. Harinadhababu and Y.S. Ranganath, *Int. J. Pharm. Sci.*, **4**, 424 (2012).