4.1. Chemicals and Instruments

All chemicals and solvents were purchased from commercial sources like Sigma Aldrich, TCI Chemicals, Merck, Spectrochem and SD-Fine Himedia and did not need purification prior to use. When needed, the solvents were dried over 3 Å or 4 Å molecular sieves and then distilled. Reaction progress was monitored on Merck precoated silica gel thin-layer chromatography (TLC) plates (without fluorescent indicator). Melting points were determined on a SMP 203 digital melting point apparatus from Lab Intelligence Appliances. Infrared spectra of the compounds were recorded on a JASCO FTIR 6100 spectrometer by KBr dispersion method. Mass spectra were recorded using a Water’s micro mass Q-Tofmicro spectrometer with an ESI source at Oxygen Healthcare Research Pvt. Ltd., Ahmedabad. $^1$H NMR spectra of all the intermediates and final compounds were recorded using Bruker AVANCE 400 MHz NMR spectrometers. $^{13}$C NMR spectra of the compounds were recorded using a Bruker AVANCE 100 MHz NMR spectrometer. Chemical shifts were measured in parts per million (ppm) downfield from an internal tetramethylsilane (TMS) standard. Single crystal X-ray intensity data for compound 5p was made on a Rigaku SCX mini diffractometer using graphite monochromated Mo-Kα radiation. Purity of the compounds was checked using HPLC analysis which were carried out at $\lambda_{\text{max}}$ 220 nm using column ODS C-18, 150mm×4.6 mm×4µ on AGILENT 1100 series. All NMR spectra and X-ray intensity data were procured from Saurashtra University, Rajkot.
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4.2 Synthetic Scheme

Fig. 4.1. Synthetic Scheme 1
As per the above synthetic scheme, designed series of was synthesized in three steps. In 1st step of synthesis, bromination of 1,2,4-triazole 1 was carried out successfully using two equivalent moles of bromine in an aqueous solution of equimolar KBr and KHCO₃ at 80°C with stirring. Reaction completed within 30-40 minutes with good yield of 3,5-dibromo-1H-1,2,4-triazole 2 [1a]. The compound 2 was characterized by melting point, ¹H-NMR and ESI-MS. Specific [M+2] peak pattern was observed due to presence of two bromine in the structure. Molecular ion peak matches with the mass. Further, it was confirmed by ¹H-NMR. The only one hydrogen present on N-1 showed the singlet peak at 7.26 ppm. In second step, simple nucleophilic substitution reaction occurred between 3,5-dibromo-1H-1,2,4-triazole 2 and seventeen differently substituted phenacyl bromides 3a-o or related chemicals 3p and 3q to give intermediate derivatives 4a-q. The reaction proceeds with nucleophilic attack of N-1 of intermediate 2 on α carbon of 3a-q with removal of HBr. We tried different protic solvents like methanol, ethanol, acetone etc. and aprotic solvent like DMF at different basic conditions viz. stirring, heating and reflux. The best yield and purity of compounds 4a-q were obtained with stirring in aceton using K₂CO₃ as base at 0°C initially and then at R.T. Reaction completed in around 4-6 h with good yield of compounds 4a-q [1b]. Many new substitutions were introduced in phenyl ring. Apart from these, two other related chemicals, 3p and 3q were also reacted with 3,5-dibromo-1H-1,2,4-triazole 2 to give intermediate 4p and 4q. Structures of all these intermediates 4a-q were confirmed by different spectroscopic analysis. In FT-IR spectra, the confirmatory peak of –C=O stretch was observed in the range of 1680-1710 cm⁻¹ in all the compounds which indicated the presence of carbonyl group. Next structural confirmation was made through ESI-MS spectra. Here, characteristic molecular ion peak pattern was observed due to presence of two bromine in the structure. All intermediates showed [M+1]⁺, [M+3]⁺, [M+5]⁺ peaks. Final confirmation was made by ¹H-NMR spectroscopy. The peak of –NH-proton at 1.25 ppm disappeared which indicated that reaction occurred at this N1 position of dibromotriazole ring. The aromatic protons of compound 4a-q appeared in the range of 7-8 ppm (δ value) with characteristic splitting pattern and value of coupling constants (J value) in each para, meta and ortho substituted phenyl ring. The CH₂ proton of substituted phenacyl bromide chain appeared at around 4.5 as singlet.

The final 3rd step of ring closing reaction also proceeds via nucleophilic substitution reaction. Reaction between intermediate 4a-q and hydrazine hydrate NH₂NH₂.H₂O ended up into the designed series of triazine derivatives 5a-q with removal of HBr and H₂O [1b]. Maximum yield was obtained with around 8-9 h reflux time in methanol and then allowed to cool.
Hydrazine hydrate is sufficiently basic on its own for the nucleophilic substitution reaction so we did not add any base in the reaction mixture. Again, structures of all the final compounds 5a-q were confirmed by different spectroscopic analysis. In FT-IR spectra of compounds 5a-q, the peak of -C=O stretch disappeared which was observed in all intermediates 4a-q. This indicated that ring formation might have occurred at that position. A broad –NH stretch of newly introduced secondary cyclic amine was observed in the range of 3400-2900 cm\(^{-1}\) in all the compounds. Strong –NH bending was also observed around 1588-1570 cm\(^{-1}\). These all data supported the product formation. Further structural confirmation was made through ESI-MS spectra. Here due to removal of one bromine as HBr out of two present in intermediates 4, only [M+1]\(^+\) and [M+3]\(^+\) peaks were observed which were of almost equal or near to equal in intensity. Final confirmation was made by NMR spectroscopy. Both \(^1\)H NMR and \(^13\)C NMR spectral analysis were done. In \(^1\)H-NMR, the change was observed by the appearance of the –NH proton at around 11 ppm. In proton decoupled \(^13\)C-NMR, all the carbon of different environment appeared at different δ value as singlet. C-7 of the triazolotriazine scaffold which is attached with bromine appeared at around 147 ppm. C-9 appeared at around 137 ppm. C-4 present as cyclic –CH\(_2\)– appeared at around 44 ppm. C-3 which is substituted with aromatic ring, appeared at around 133 ppm. All the aromatic carbons of phenyl ring appeared in the range of 125-150 ppm depending upon their substitutions. Structure was also confirmed by single crystal XRD analysis of compound 5p.

Although the designed scheme 1 was up to synthesis of compounds 5a-q, we also thought of possible diversification or extension of the scheme. Few variations of the same synthetic scheme were thought of which could end up into three more different scaffolds.

Initially before the synthetic trials, these scaffolds were studied \textit{in-silico} for the DPP-4 inhibition. For that, all molecules with different R substitutions were mapped on developed pharmacophore A and screened in docking protocol. Although values are not shown here for each substitution variation we thought of, but overall all these scaffolds were also giving
acceptable pharmacophore fit and docking scores. So, such attractive in-silico results motivated us to expand the series further. We tried to synthesize each of them but unfortunately could not end up into the desired products. The various synthetic efforts which were given based on the literature, their results and probable explanation are briefly described in section 4.3.4, 4.3.5 and 4.3.6.

4.3 Experimental Procedure and Spectral Characterization

4.3.1 Synthesis of 3,5-dibromo-1H-1,2,4-triazole 2

Compound 2 was recrystallized from hot water as white solids. Yield 71.06%; m.p. 190-195°C; FT-IR (KBr, cm⁻¹): 3099, 2903, 2242.8, 2116.5, 1516.7, 1427, 1342.2, 1269.9, 1130, 1014.3, 986.4, 832, 718.3, 701.9. ¹H NMR (400 MHz, CDCl₃): δ 1.25 (s, 1H). ESI-MS calculated for (C₂HBr₂N₃) [M+1]^+ 225.85, found 223.8 [M-1]^+, 225.8 [M+1]^+, 227.9 [M+3]^+. 

4.3.2 General Procedure for Synthesis of 2-(3,5-dibromo-1H-1,2,4-triazol-1-yl)-1-substituted-ethanone 4a-q

Various substituted phenacyl bromides (3a-o) or equivalent reactants (3p, 3q) were added slowly in portions at 0°C with stirring. After complete addition, reaction mixture was stirred at room temperature for 4-6 h. The progress of reaction was monitored by TLC. After completion of chemical reaction, the reaction mixture was filtered off. The residue was washed with acetone and combined filtrate was concentrated under reduced pressure to get the solids which were then recrystallized by co-solvent recrystallization method using ethyl acetate-petroleum ether to obtain the pure solids (4a-q).

4.3.2.1. 2-(3,5-dibromo-1H-1,2,4-triazol-1-yl)-1-(4-methoxyphenyl)ethanone 4a

Yield: 80%; white solid; m.p. 147-151°C; FT-IR (KBr, cm⁻¹): 1683.55 (-C=O stretch), 1067.41, 1125.26 (-C-O-C stretch); ¹H NMR (400 MHz, CDCl₃): δ 3.86 (s, 3H), 7.13 (d, J = 8.92 Hz, 2H), 8.04 (d, J = 8.92 Hz, 2H), 5.99 (s, 2H). ESI-MS calculated for (C₁₁H₉Br₂N₃O₂) [M+1]^+ 373.91, found 374.1 [M+1]^+, 376 [M+3]^+, 378 [M+5]^+

4.3.2.2. 2-(3,5-dibromo-1H-1,2,4-triazol-1-yl)-1-p-tolylethanone 4b

Yield: 78%; white solid; m.p. 152-156°C; FT-IR (KBr, cm⁻¹): 1687.41 (-C=O stretch); ¹H NMR (400 MHz, CDCl₃): δ 2.46 (s, 3H), 7.36 (d, J = 8.00 Hz, 2H), 7.88 (d, J = 8.28 Hz, 2H), 4.59 (s,
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2H). ESI-MS calculated for (C_{11}H_{9}Br_{2}N_{3}O) [M+1]^+ 357.91, found 358 [M+1]^+, 360 [M+3]^+, 362 [M+5]^+

4.3.2.4. 1-(4-chlorophenyl)-2-(3,5-dibromo-1H-1,2,4-triazol-1-yl)ethanone 4d

Yield: 83%; pale yellow solid; m.p. 158-163°C; FT-IR (KBr, cm\(^{-1}\)): 1702.84 (-C=O stretch); \(^1\)H NMR (400 MHz, CDCl\(_3\)): \(\delta\) 7.94 (d, \(J = 9.0\) Hz, 2H), 7.56 (d, \(J = 9.1\) Hz, 2H), 5.62 (s, 2H). ESI-MS calculated for (C\(_{10}\)H\(_8\)Br\(_2\)C\(_3\)N\(_3\)O) [M+1]^+ 377.86, found 378 [M+1]^+, 380 [M+3]^+, 381.9 [M+5]^+

4.3.2.5. 2-(3,5-dibromo-1H-1,2,4-triazol-1-yl)-1-(4-phenylphenyl)ethanone 4e

Yield: 84%; pale yellow solid; m.p. 155-160°C; FT-IR (KBr, cm\(^{-1}\)): 1692.23 (-C=O stretch); \(^1\)H NMR (400 MHz, CDCl\(_3\)): \(\delta\) 7.98 (d, \(J = 8.32\) Hz, 2H), 7.71 (d, \(J = 8.28\) Hz, 2H), 7.58 (d, \(J = 7.24\) Hz, 2H), 7.45-7.35 (m, 3H), 4.58 (s, 2H). ESI-MS calculated for (C\(_{16}\)H\(_{11}\)Br\(_2\)N\(_3\)O) [M+1]^+ 419.93, found 420.3 [M+1]^+, 422.1 [M+3]^+, 424.1 [M+5]^+

4.3.2.6. 2-(3,5-dibromo-1H-1,2,4-triazol-1-yl)-1-(3-fluorophenyl)ethanone 4f

Yield: 75%; pale yellow solid; m.p. 94-98°C; FT-IR (KBr, cm\(^{-1}\)): 1697.05 (-C=O stretch); \(^1\)H NMR (400 MHz, CDCl\(_3\)): \(\delta\) 7.68 (d, 1H), 7.62-7.58 (m, 1H), 7.52-7.47 (m, 1H), 7.36-7.31 (m, 1H), 4.52 (s, 2H). ESI-MS calculated for (C\(_{10}\)H\(_8\)Br\(_2\)F\(_2\)N\(_3\)O) [M+1]^+ 361.89, found 362.1 [M+1]^+, 364.1 [M+3]^+, 366.1 [M+5]^+

4.3.2.7. 2-(3,5-dibromo-1H-1,2,4-triazol-1-yl)-1-(4-hydroxyphenyl)ethanone 4g

Yield: 68%; white solid; m.p.: 199-204°C; FT-IR (KBr, cm\(^{-1}\)): 3399.89 (-OH stretch), 1671.98 (-C=O stretch); \(^1\)H NMR (400 MHz, CDCl\(_3\)): \(\delta\) 7.86 (d, \(J = 7.88\) Hz, 2H), 6.90 (d, \(J = 7.92\) Hz, 2H), \(\delta\) 7.98 (d, \(J = 8.32\) Hz, 2H), 4.49 (s, 2H), 1.97 (s, 1H). ESI-MS calculated for (C\(_{16}\)H\(_7\)Br\(_2\)N\(_2\)O\(_2\)) [M+1]^+ 359.89, found 360.1 [M+1]^+, 362 [M+3]^+, 364.1 [M+5]^+

4.3.2.8. 2-(3,5-dibromo-1H-1,2,4-triazol-1-yl)-1-(3-nitrophenyl)ethanone 4h

Yield: 65%; buff yellow solid; m.p: 139-143°C; FT-IR (KBr, cm\(^{-1}\)): 1697.05 (-C=O stretch), 1530.24 and 1350.89 (-N-O stretch); \(^1\)H NMR (400 MHz, CDCl\(_3\)): \(\delta\) 7.86 (d, \(J = 7.88\) Hz, 2H), 6.90 (d, \(J = 7.92\) Hz, 2H), \(\delta\) 8.79 (d, 1H), 8.49 (d, \(J = 8.2\) Hz, 1H), 8.32 (d, \(J = 7.72\) Hz, 1H), 7.78-7.74 (t, 1H), 5.77 (s, 2H). ESI-MS calculated for (C\(_{10}\)H\(_8\)Br\(_2\)N\(_3\)O\(_3\)) [M+1]^+ 388.88, found 388.4 [M+1]^+, 390.5 [M+3]^+, 392.5 [M+5]^+
4.3.2.9. 2-(3,5-dibromo-1H-1,2,4-triazol-1-yl)-1-(3-methoxyphenyl)ethanone 4i

Yield: 84%; white solid; m.p. 116-120°C; FT-IR (KBr, cm\(^{-1}\)): 1692.23 (-C=O stretch), 1127.19 (C-O-C Stretch); \(^1\)H NMR (400 MHz, CDCl\(_3\)): δ 8.30 (s, 3H), 7.47-7.14 (m, 4H), 4.54 (s, 2H). ESI-MS calculated for (C\(_{11}\)H\(_9\)Br\(_2\)N\(_2\)O) [M+1]\(^+\) 373.91, found 373.5 [M+1]\(^+\), 374.4 [M+3]\(^+\), 377.5 [M+5]\(^+\)

4.3.2.10. 3-(2-(3,5-dibromo-1H-1,2,4-triazol-1-yl)acetyl)benzonitrile 4j

Yield: 73%; yellow solid; m.p.: 128-132°C; FT-IR (KBr, cm\(^{-1}\)): 1698.98 (-C=O stretch), 2236.06 (-CN stretch); \(^1\)H NMR (400 MHz, CDCl\(_3\)): δ 8.19 (s, 1H), 8.15-8.13 (m, 1H), 7.92-7.89 (m, 1H), 7.69-7.65 (t, 1H), 4.56 (s, 2H). ESI-MS calculated for (C\(_{11}\)H\(_9\)Br\(_2\)N\(_2\)O) [M+1]\(^+\) 368.89, found 368.9 [M+1]\(^+\), 370.9 [M+3]\(^+\), 372.9 [M+5]\(^+\)

4.3.2.11. 2-(3,5-dibromo-1H-1,2,4-triazol-1-yl)-1-(2,4-difluorophenyl)ethanone 4k

Yield: 85%; red solid; m.p. 130-135°C; FT-IR (KBr, cm\(^{-1}\)): 1697.05 (-C=O stretch), \(^1\)H NMR (400 MHz, CDCl\(_3\)): δ 8.02 (s, 1H), 7.97 (m, 1H), 8.15 (d, J=8.16, 1H), 7.69-7.65 (t, 1H), 4.50 (s, 2H). ESI-MS calculated for (C\(_{11}\)H\(_9\)Br\(_2\)Cl\(_2\)N\(_2\)O) [M+1]\(^+\) 399.9, found 399.6 [M+1]\(^+\), 381.7 [M+3]\(^+\), 383.7 [M+5]\(^+\)

4.3.2.12. 2-(3,5-dibromo-1H-1,2,4-triazol-1-yl)-1-(3,4-dichlorophenyl)ethanone 4l

Yield: 81%; pale yellow solids; m.p. 191-196°C; FT-IR (KBr, cm\(^{-1}\)): 1707.66 (-C=O stretch), \(^1\)H NMR (400 MHz, CDCl\(_3\)): δ 7.99 (s, 1H), 7.74 (d, J=8.36, 1H), 7.60 (d, J = 8.36, 1H), 4.50 (s, 2H). ESI-MS calculated for (C\(_{11}\)H\(_9\)Br\(_2\)Cl\(_2\)N\(_2\)O) [M+1]\(^+\) 411.82, found 411.6 [M+1]\(^+\), 413.5 [M+3]\(^+\), 414.5 [M+5]\(^+\)

4.3.2.13. 2-(3,5-dibromo-1H-1,2,4-triazol-1-yl)-1-(4-(trifluoromethyl)phenyl)ethanone 4m

Yield: 78%; yellow solids; m.p. 120-123°C; FT-IR (KBr, cm\(^{-1}\)): 1673.66 (-C=O stretch), \(^1\)H NMR (400 MHz, CDCl\(_3\)): δ 8.11 (d, J=8.16, 2H), 7.85 (d, J=8.28, 2H), 5.65 (s, 2H). ESI-MS calculated for (C\(_{11}\)H\(_9\)Br\(_2\)F\(_3\)N\(_2\)O) [M+1]\(^+\) 411.88, found 411.9 [M+1]\(^+\), 414.0 [M+3]\(^+\), 415.8 [M+5]\(^+\)

4.3.2.14. 1-(3-chloro-4-fluorophenyl)-2-(3,5-dibromo-1H-1,2,4-triazol-1-yl)ethanone 4n
Yield: 76%; pale yellow solids; m.p. 190-194°C; FT-IR (KBr, cm⁻¹): 1690.06 (-C=O stretch), 1H NMR (400 MHz, CDCl₃): δ 8.09 (d, J=6.88, 1H), 7.92 (m, J=2.24, 1H), 7.36 (t, J = 8.44, 1H), 5.60 (s, 2H). ESI-MS calculated for (C₁₀H₃Br₂ClFN₃O) [M+1]+ 395.85, found 395.8 [M+1]+, 397.9 [M+3]+, 399.8 [M+5]+

4.3.2.15. 2-(3,5-dibromo-1H-1,2,4-triazol-1-yl)-1-(2-methoxyphenyl)ethanone 4o

Yield: 81%; white solids; m.p. 130-134°C; FT-IR (KBr, cm⁻¹): 1687.34 (-C=O stretch), 1135.22 (C-O-C Stretch); 1H NMR (400 MHz, CDCl₃): δ 7.97 (d, J=7.8, 1H), 7.63 (t, J=8.68, 1H), 7.11-7.05 (m, 2H), 5.60 (s, 2H), 4.03 (s, 3H). ESI-MS calculated for (C₁₁H₉Br₂N₂O₂) [M+1]+ 377.91, found 373.9 [M+1]+, 375.8 [M+3]+, 377.5 [M+5]+

4.3.2.16. 2-(3,5-dibromo-1H-1,2,4-triazol-1-yl)-1-(naphthalene-2-yl)ethanone 4p

Yield: 84%; pale yellow solid; m.p. 133-137°C; FT-IR (KBr, cm⁻¹): 1698.98 (-C=O stretch); 1H NMR (400 MHz, CDCl₃): δ 8.42 (s, 1H), 7.95-7.85 (m, 4H), 7.64-7.54 (m, 2H), 5.69 (s, 2H). ESI-MS calculated for (C₁₄H₇Br₂N₂O) [M+1]+ 393.91, found 394 [M+1]+, 396 [M+3]+, 398 [M+5]+

4.3.3 Synthesis of 7-bromo-3-substituted-1,4-dihydro-[1,2-4]triazolo[5,1-c][1,2,4]triazine 5a-q

4.3.3.1. 7-bromo-3-(4-methoxyphenyl)-1,4-dihydro-[1,2,4]triazolo[5,1-c][1,2,4]triazine 5a

Yield: 71%; white solid; m.p.: 273-277°C; FT-IR (KBr, cm⁻¹): 3231 (-NH stretch), 1588.09 (-NH bending); 1H NMR (400 MHz, DMSO-d₆): δ ppm 3.81 (s, 3H), 5.21 (s, 2H), 7.02 (d, J = 8.92 Hz, 2H), 7.71 (d, J = 8.88 Hz, 2H), 11.55 (s, 1H, NH). 13C NMR (100 MHz, DMSO-d₆): δ ppm 160.40, 147.44, 137.87, 131.15, 128. 21, 126.54, 113.92, 55.26, 44.69. ESI-MS calculated for (C₁₁H₁₀BrN₃O) [M+1]+ 308.0, found 308.0 [M+1]+, 310.0 [M+3]+

4.3.3.2. 7-bromo-3-p-tolyl-1,4-dihydro-[1,2,4]triazolo[5,1-c][1,2,4]triazine 5b

Yield: 73%; white solid; m.p.: 282-286°C; FT-IR (KBr, cm⁻¹): 3120.26 (-NH stretch), 1581.34 (-NH bending); 1H NMR (400 MHz, DMSO-d₆): δ ppm 2.36 (s, 3H), 5.21 (s, 2H), 7.28 (d, J = 8.12 Hz, 2H), 7.65 (d, J = 8.2 Hz, 2H), 11.62 (s, 1H, NH). 13C NMR (100 MHz, DMSO-d₆): δ ppm 147.40, 139.36, 137.88, 131.25, 129.11, 128.33, 124. 92, 44.75, 20.87. ESI-MS calculated for (C₁₁H₁₀BrN₃S) [M+1]+ 292.01, found 292.1 [M+1]+, 294.1 [M+3]+

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4.3.3.4. 7-bromo-3-(4-chlorophenyl)-1,4-dihydro-[1,2,4]triazolo[5,1-c][1,2,4]triazine 5d

Yield: 73%; white solid; m.p. >300°C; FT-IR (KBr, cm⁻¹): 3211.86 (-NH stretch), 1588.09 (-NH bending); ¹H NMR (400 MHz, DMSO-d₆): δ ppm 5.25 (s, 2H), 7.77 (d, J = 8.68 Hz, 2H), 7.54 (d, J = 8.72 Hz, 2H), 7.17 (s, 1H, NH). ¹³C NMR (100 MHz, DMSO-d₆): δ ppm 147.16, 137.95, 136.78, 134.21, 132.84, 128.96, 126.73, 44.79. ESI-MS calculated for (C₁₀H₇BrClN₅) [M+1]+ 311.96, found 312.1 [M+1]+, 314.1 [M+3]+, 316.0 [M+5]+

4.3.3.5. 7-bromo-3-(4-phenylphenyl)-1,4-dihydro-[1,2,4]triazolo[5,1-c][1,2,4]triazine 5e

Yield: 76%; white solid; m.p.: 279-283°C; FT-IR (KBr, cm⁻¹): 3113.51 (-NH stretch), 1574.56 (-NH bending); ¹H NMR (400 MHz, DMSO-d₆): δ ppm 5.29 (s, 2H), 7.85-7.72 (m, 6H), 7.51 (t, 2H), 7.42 (t, 1H), 7.17 (s, 1H, NH). ¹³C NMR (100 MHz, DMSO-d₆): δ ppm 147.32, 141.07, 140.10, 139.32, 137.95, 137.44, 133.00, 129.01, 127.85, 126.84, 24.84. ESI-MS calculated for (C₁₆H₁₂BrN₃) [M+1]+ 354.04, found 354.3 [M+1]+, 356.2 [M+3]+

4.3.3.6. 7-bromo-3-(3-fluorophenyl)-1,4-dihydro-[1,2,4]triazolo[5,1-c][1,2,4]triazine 5f

Yield: 72%; white solid; m.p.: 270-273°C; FT-IR (KBr, cm⁻¹): 314.75 (-NH stretch), 1574.59 (-NH bending); ¹H NMR (400 MHz, DMSO-d₆): δ ppm 4.30 (s, 2H), 7.32-7.27 (t, 1H), 7.59-7.48 (m, 3H), 7.17 (s, 1H, NH). ¹³C NMR (100 MHz, DMSO-d₆): δ ppm 163.47; 161.05, 147.16, 137.96, 136.73, 130.63, 121.16, 116.47, 111.74, 111.51, 44.93 ESI-MS calculated for (C₁₀H₇BrF₃) [M+1]+ 295.99, found 296.0 [M+1]+, 298.2 [M+3]+

4.3.3.7. 4-(7-bromo-1,4-dihydro-[1,2,4]-triazolo[5,1-c][1,2,4]triazin-3-yl)phenol 5g

Yield: 67%; white solid; m.p. >300°C; FT-IR (KBr, cm⁻¹): 3159.79 (-OH stretch), 1598.7 (-NH bending); ¹H NMR (400 MHz, DMSO-d₆): δ ppm 5.17 (s, 2H), 6.85 (d, J = 8.68 Hz, 2H), 7.60 (d, J = 8.64, 2H), 9.90 (s, 1H, OH), 11.48 (s, 1H, NH). ¹³C NMR (100 MHz, DMSO-d₆): δ ppm 158.93, 147.52, 137.84, 133.54, 130.06, 126.63, 114.30, 44.62. ESI-MS calculated for (C₁₀H₇BrN₃O) [M+1]+ 293.99, found 294.3 [M+1]+, 296.1 [M+3]+

4.3.3.8. 7-bromo-3-(3-nitrophenyl)-1,4-dihydro-[1,2,4]triazolo[5,1-c][1,2,4]triazine 5h
Yield: 62%; white solid; m.p. 275-278°C; FT-IR (KBr, cm⁻¹): 3099.05 (-NH stretch), 1581.34 (-NH bending); ¹H NMR (400 MHz, DMSO-d₆): δ ppm 4.34 (s, 2H), 7.79-7.74 (t, 1H), 8.14 (d, J = 8.28, 2H), 8.29 (d, J = 8.2, 2H), 8.53 (s, 1H), 11.94 (s, 1H, NH). ¹³C NMR (100 MHz, DMSO-d₆): δ ppm 148.01, 147.02, 138.04, 136.06, 135.61, 131.20, 130.27, 123.86, 119.28, 45.00. ESI-MS calculated for (C₁₀H₂BrN₆O₂) [M+1]⁺ 322.98, found 323.0 [M+1]⁺, 325.2 [M+3]⁺

4.3.3.9. 7-bromo-3-(3-methoxyphenyl)-1,4-dihydro-[1,2,4]triazolo[5,1-c][1,2,4]triazine 5i

Yield: 75%; white solid; m.p. 250-253°C; FT-IR (KBr, cm⁻¹): 3094.23 (-NH stretch), 1559.17 (-NH bending); ¹H NMR (400 MHz, DMSO-d₆): δ ppm 3.80 (s, 3H), 5.24 (s, 2H), 7.04 (s, 1H), 7.31 (d, J = 8.12, 2H), 7.40-7.36 (t, 1H), 11.69 (s, 1H, NH). ¹³C NMR (100 MHz, DMSO-d₆): δ ppm 159.30, 147.30, 137.92, 134.37, 131.20, 129.65, 117.47, 114.50, 110.01, 55.14, 44.88. ESI-MS calculated for (C₁₁H₁₀BrN₅O) [M+1]⁺ 308.01, found 307.6 [M+1]⁺, 309.6 [M+3]⁺

4.3.3.10. 3-(7-bromo-1,4-dihydro-[1,2,4]triazolo[5,1-c][1,2,4]triazin-3-yl)benzonitrile 5j

Yield: 69%; white solid; m.p. 287-291°C; FT-IR (KBr, cm⁻¹): 3119.3 (-NH stretch), 2228.34 (-CN stretch), 1564.95 (-NH bending); ¹H NMR (400 MHz, DMSO-d₆): δ ppm 5.28 (s, 2H), 7.69-7.65 (t, 1H), 7.92 (d, J = 7.68, 1H), 8.10 (d, J = 8.12, 1H), 8.12 (s, 1H, NH). ¹³C NMR (100 MHz, DMSO-d₆): δ ppm 147.04, 138.01, 136.16, 135.15, 132.93, 129.79, 129.44, 128.50, 118.44, 111.81, 44.90. ESI-MS calculated for (C₁₁H₁₂BrN₅O) [M+1]⁺ 302.99, found 302.5 [M+1]⁺, 304.6 [M+3]⁺

4.3.3.11. 7-bromo-3-(2,4-difluorophenyl)-1,4-dihydro-[1,2,4]triazolo[5,1-c][1,2,4]triazine 5k

Yield: 72%; pale yellow solid; m.p. 214-218°C; FT-IR (KBr, cm⁻¹): 3209.3 (-NH stretch), 1564.95 (-NH bending); ¹H NMR (400 MHz, DMSO-d₆): δ ppm 5.21 (s, 2H), 7.22-7.18 (t, 1H), 7.41-7.35 (t, 1H), 7.83-7.77 (m, 1H), 11.76 (s, 1H, NH). ¹³C NMR (100 MHz, DMSO-d₆): δ ppm 164.08; 163.96, 161.60, 161.48, 159.08, 158.96, 147.32, 137.92, 135.15, 130.34; 130.29, 130.24, 130.19, 112.19, 112.01, 111.97, 105.15; 104.89, 104.62, 46.38. ESI-MS calculated for (C₁₀H₆BrF₂N₅) [M+1]⁺ 313.98, found 314.8 [M+1]⁺
4.3.3.12. 7-bromo-3-(3,4-dichlorophenyl)-1,4-dihydro-[1,2,4]triazolo[5,1-c][1,2,4]triazine 5l

Yield: 68%; white solid; m.p.: 272-276°C; FT-IR (KBr, cm\(^{-1}\)): 3206.08 (-NH stretch), 1588.09 (-NH bending); \(^1\)H NMR (400 MHz, DMSO-d\(_6\)): δ ppm 5.23 (s, 2H), 7.90 (s, 1H), 7.75 (d, 2H), 11.86 (s, 1H, NH). \(^1^C\) NMR (100 MHz, DMSO-d\(_6\)): δ ppm 146.98 (1C), 138.01 (1C), 135.77 (1C), 134.58 (1C), 130.68 (1C), 129.64 (1C), 128.42 (1C), 126.67 (1C), 125.05 (1C), 44.84 (1C). ESI-MS calculated for (C\(_{10}\)H\(_6\)BrCl\(_2\)N\(_3\)) [M+1]\(^+\) 345.92, found 344.5 [M+1]\(^+\) 347.5 [M+3]\(^+\) 349.5 [M+5]\(^+\)

4.3.3.13. 7-bromo-3-(4-(trifluoromethyl)phenyl)-1,4-dihydro-[1,2,4]triazolo[5,1-c][1,2,4]triazine 5m

Yield: 73%; white solid; m.p. 275-278°C; FT-IR (KBr, cm\(^{-1}\)): 3146.08 (-NH stretch), 1583.45 (-NH bending); \(^1\)H NMR (400 MHz, DMSO-d\(_6\)): δ ppm 5.29 (s, 2H), 7.95 (d, J=8.24 Hz, 1H), 7.81 (d, J=8.44 Hz, 2H), 11.92 (s, 1H, NH). \(^1^C\) NMR (100 MHz, DMSO-d\(_6\)): δ ppm 147.01, 140.66, 138.01, 136.42, 128.80, 125.64, 122.69, 119.99, 44.92. ESI-MS calculated for (C\(_{11}\)H\(_7\)BrF\(_3\)N\(_5\)) [M+1]\(^+\) 345.98, found 346.0 [M+1]\(^+\) 347.8 [M+3]\(^+\)

4.3.3.14. 7-bromo-3-(3-chloro-4-fluorophenyl)-1,4-dihydro-[1,2,4]triazolo[5,1-c][1,2,4]triazine 5n

Yield: 70%; faint yellow solid; m.p.: 242-246°C; FT-IR (KBr, cm\(^{-1}\)): 3196.08 (-NH stretch), 1579.02 (-NH bending); \(^1\)H NMR (400 MHz, DMSO-d\(_6\)): δ ppm 5.24 (s, 2H), 7.91 (d, J=7.12 Hz, 1H), 7.76 (s, 1H), 7.53-7.49 (t, 1H), 11.81 (s, 1H, NH). \(^1^C\) NMR (100 MHz, DMSO-d\(_6\)): δ ppm 159.00; 156.52, 147.05, 137.99, 135.91, 131.89, 127.36, 125.86, 120.07, 117.20, 44.88 ESI-MS calculated for (C\(_{10}\)H\(_6\)BrClF\(_3\)N\(_3\)) [M+1]\(^+\) 329.95, found 330.0 [M+1]\(^+\) 332.2 [M+3]\(^+\)

4.3.3.15. 7-bromo-3-(2-methoxyphenyl)-1,4-dihydro-[1,2,4]triazolo[5,1-c][1,2,4]triazine 5o

Yield: 78%; white solid; m.p. 157-160°C; FT-IR (KBr, cm\(^{-1}\)): 3102.20 (-NH stretch), 1564.34 (-NH bending); \(^1\)H NMR (400 MHz, DMSO-d\(_6\)): δ ppm 3.85 (s, 3H), 5.13 (s, 2H), 7.04-6.99 (t, 1H), 7.13 (d, J = 8.36 Hz, 1H), 7.45-7.42 (t, 1H), 7.51 (d, J=7.2 Hz, 1H), 11.49 (s, 1H, NH). \(^1^C\) NMR (100 MHz, DMSO-d\(_6\)): δ ppm 157.58, 148.06, 139.92, 137.84, 131.08, 129.36, 123.92, 120.59, 111.87, 55.69, 46.72. ESI-MS calculated for (C\(_{11}\)H\(_{10}\)BrN\(_3\)O) [M+1]\(^+\) 308.01, found 308.0 [M+1]\(^+\), 309.9 [M+3]\(^+\)
4.3.3.16. 7-bromo-3-(naphthalene-2-yl)-1,4-dihydro-[1,2-4]triazolo[5,1-c][1,2,4]triazine 5p

Yield: 75%; white solid; m.p. 254-257°C; FT-IR (KBr, cm⁻¹): 3112.55 (NH stretch), 1576.52 (NH bending); ¹H NMR (400 MHz, DMSO-d₆): δ ppm 4.39 (s, 2H), 7.58 (s, 1H), 8.43-7.96 (m, 6H), 11.81 (s, 1H, NH). ¹³C NMR (100 MHz, DMSO-d₆): δ ppm 147.31, 137.59, 133.24, 131.40, 128.49, 128.00, 127.58, 127.26, 127.05, 126.67, 125.97, 124.99, 122.13, 44.84. ESI-MS calculated for (C₁₄H₁₀BrN₅) [M+1]⁺ 328.01, found 328.1 [M+1]⁺, 330.1 [M+3]⁺
4.4 Representative Spectra

4.4.1 $^1$H NMR Spectrum (400 MHz, CDCl$_3$) of compound 4a

4.4.2 ESI-MS spectrum of compound 4a
4.4.3 $^1$H NMR Spectrum (400 MHz, CDCl$_3$) of compound 4e

4.4.4 ESI-MS spectrum of compound 4e
4.4.5 $^1$H NMR Spectrum (400 MHz, CDCl$_3$) of compound 4n

4.4.6 ESI-MS spectrum of compound 4n
4.4.7 $^1$H NMR Spectrum (400 MHz, CDCl$_3$) of compound 4q
4.4.8 ESI-MS spectrum of compound 4q

![ESI-MS spectrum of compound 4q](image)

4.4.9 $^1$H NMR Spectrum (400 MHz, DMSO) of compound 5b

![$^1$H NMR Spectrum of compound 5b](image)
4.4.10 $^{13}$C NMR Spectrum (100 MHz, DMSO) of compound 5b

4.4.11 ESI-MS spectrum of compound 5b
4.4.12 $^1$H NMR Spectrum (400 MHz, DMSO) of compound 5c
4.4.13 $^{13}\text{C}$ NMR Spectrum (100 MHz, DMSO) of compound 5c

4.4.14 ESI-MS spectrum of compound 5c
4.4.15 $^1$H NMR Spectrum (400 MHz, DMSO) of compound 5g
4.4.16 $^{13}$C NMR Spectrum (100 MHz, DMSO) of compound 5g

![13C NMR Spectrum](image1)

4.4.17 ESI-MS spectrum of compound 5g

![ESI-MS Spectrum](image2)
4.4.18 $^1$H NMR Spectrum (400 MHz, DMSO) of compound 5h
4.4.19 $^{13}$C NMR Spectrum (100 MHz, DMSO) of compound 5h

4.4.20 ESI-MS spectrum of compound 5h
4.4.21 $^1$H NMR Spectrum (400 MHz, DMSO) of compound 5q
4.4.22 $^{13}$C NMR Spectrum (100 MHz, DMSO) of compound 5q

4.4.23 ESI-MS spectrum of compound 5q
4.5 X-Ray Crystallographic Analysis of Compound 5p

4.5.1 Data Collection

The X-ray crystallographic analysis of compound 5p was determined on a colorless crystal, with approximate dimensions of 0.66 mm x 0.40 mm x 0.30 mm, grown from the vapor diffusion method using THF and hexane. All the measurements were made on a RIGAKU SCX mini diffractometer using graphite monochromated Mo-Kα radiation. The crystal to detector distance was fixed at 52 mm. The data were collected at a temperature of 20±1°C to a maximum 2θ value of 54.9° and a total of 540 oscillation images were collected. A sweep of data was done using ω oscillations from -120.0 to 60.0° in 1.0° steps. The exposure rate was 7.0 [sec./°]. The detector swing angle was -30.80°. A second sweep was performed using ω oscillations from -120.0 to 60.0° in 1.0° steps. The exposure rate was 7.0 [sec./°]. The detector swing angle was -30.80°. Another sweep was performed using ω oscillations from -120.0 to 60.0° in 1.0° steps. The exposure rate was 7.0 [sec./°]. The detector swing angle was -30.80°. Readout was performed in the 0.146 mm pixel mode.

4.5.2 Data Reduction

During data reduction of the 12991 reflections that were collected, 6058 were unique (Rint = 0.0392) and so equivalent reflections were merged. Data were collected and processed using CrystalClear (Rigaku) [17]. The linear absorption coefficient, µ, for Mo-Kα radiation is 30.785 cm⁻¹. An empirical absorption correction was applied which resulted in transmission factors ranging from 0.189 to 0.397. The data were corrected for Lorentz and polarization effects.

4.5.3 Structure Solution and Refinement

The structure was solved by direct methods [18] and expanded using Fourier techniques. The non-hydrogen atoms were refined anisotropically. Hydrogen atoms were refined using the riding model. The final cycle of full-matrix least-squares refinement [19] on F² was based on 6058 observed reflections and 361 variable parameters and converged. The standard deviation of an observation of unit weight [20] was 0.99. Unit weights were used. The maximum and minimum peaks on the final difference Fourier map corresponded to 0.40 and -0.43 e /Å³, respectively. Neutral atom scattering factors were taken from Cromer and Hubbell [22]. Anomalous dispersion effects were included in F calculation [22] the values for Δf' and Δf" were those of Creagh and McAuley [23]. The values for the mass attenuation coefficients are those of Creagh and Hubbell [24]. All calculations were performed using the CrystalStructure
[25], a crystallographic software package except for refinement, which was performed using SHELXL-97 [26]. Parameters obtained during the XRD study is given in Table 4.2 and ORTEP image of compound 5p is given in Fig. 4.16.

**Table 4.2. Single crystal XRD parameters of compound 5p**

<table>
<thead>
<tr>
<th>Sr. no.</th>
<th>Parameter</th>
<th>Compound 5p</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Empirical formula</td>
<td>C$<em>{14}$H$</em>{10}$BrN$_{5}$</td>
</tr>
<tr>
<td>2</td>
<td>MW</td>
<td>327.01</td>
</tr>
<tr>
<td>3</td>
<td>Color, Habit</td>
<td>Colorless, Chip</td>
</tr>
<tr>
<td>4</td>
<td>Crystal system</td>
<td>Triclinic</td>
</tr>
<tr>
<td>5</td>
<td>Crystal dimensions</td>
<td>0.660 X 0.400 X 0.300 mm</td>
</tr>
<tr>
<td>6</td>
<td>Space group</td>
<td>P-1 (#2)</td>
</tr>
<tr>
<td>7</td>
<td>Z</td>
<td>4</td>
</tr>
<tr>
<td>8</td>
<td>a (°Å)</td>
<td>6.7005(8) Å</td>
</tr>
<tr>
<td>9</td>
<td>b (°Å)</td>
<td>9.742(2) Å</td>
</tr>
<tr>
<td>10</td>
<td>c (°Å)</td>
<td>20.978(3) Å</td>
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<tr>
<td>11</td>
<td>α (deg)</td>
<td>90.0000 °</td>
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<tr>
<td>12</td>
<td>β (deg)</td>
<td>90.0000 °</td>
</tr>
<tr>
<td>13</td>
<td>γ (deg)</td>
<td>102.348(3) °</td>
</tr>
<tr>
<td>14</td>
<td>V (°Å$^3$)</td>
<td>1337.7(3) Å$^3$</td>
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<tr>
<td>15</td>
<td>Density (g/cm$^3$)</td>
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<td>T</td>
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<td>17</td>
<td>X-ray wavelength</td>
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<td>18</td>
<td>μ(MoKα)</td>
<td>30.785 cm$^{-1}$</td>
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<tr>
<td>19</td>
<td>No. of reflns collected</td>
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</tr>
<tr>
<td></td>
<td></td>
<td>Unique: 6058 (R$_{int}$ = 0.0392)</td>
</tr>
<tr>
<td>20</td>
<td>R1 (obsd)</td>
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</tr>
<tr>
<td>21</td>
<td>wR2 (all)</td>
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<tr>
<td>22</td>
<td>2θ$_{max}$</td>
<td>54.9°</td>
</tr>
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</table>
4.6 Determination of Purity of Final Compounds 5a-q

Purity of final compounds 5a-q was checked by HPLC analysis. A solution of compounds prepared in water: ACN (1:1, 5µl) was injected into a C\textsubscript{18} column (J'SPHERE 4 µ; 150 X 4.6 mm) and eluted with a linear gradient of ACN in water, both buffered with 0.05 % TFA, using a flow rate of 1 ml / min, with effluent monitoring by PDA detector at 220 nm. A typical gradient of 20 % to 70 % of water-ACN mixture, buffered with 0.05 % TFA was used, over a period of 100 minutes, with 1% gradient change per minute. The retention time and peak purity of chromatographic peak of various compounds was noted down. Table 4.3. shows the retention time and area % of each compound. The area% almost corresponds to the %purity of compounds.

Table 4.3. Results of the purity of compounds by HPLC analysis

<table>
<thead>
<tr>
<th>Compound</th>
<th>Retention Time</th>
<th>Area %</th>
<th>Compound</th>
<th>Retention Time</th>
<th>Area %</th>
</tr>
</thead>
<tbody>
<tr>
<td>5a</td>
<td>16.35</td>
<td>94.57</td>
<td>5j</td>
<td>14.58</td>
<td>96.82</td>
</tr>
<tr>
<td>5b</td>
<td>16.66</td>
<td>94.76</td>
<td>5k</td>
<td>14.63</td>
<td>94.56</td>
</tr>
<tr>
<td>5c</td>
<td>16.77</td>
<td>97.21</td>
<td>5l</td>
<td>9.79</td>
<td>96.60</td>
</tr>
<tr>
<td>5d</td>
<td>10.58</td>
<td>94.44</td>
<td>5m</td>
<td>10.47</td>
<td>97.51</td>
</tr>
<tr>
<td>5e</td>
<td>12.68</td>
<td>98.29</td>
<td>5n</td>
<td>10.18</td>
<td>96.87</td>
</tr>
<tr>
<td>5f</td>
<td>10.99</td>
<td>96.27</td>
<td>5o</td>
<td>10.14</td>
<td>98.42</td>
</tr>
<tr>
<td>5g</td>
<td>11.27</td>
<td>98.43</td>
<td>5p</td>
<td>15.99</td>
<td>99.25</td>
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### 4.6.1. Chromatogram of Compound 5c

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<th>RetTime [min]</th>
<th>Width [min]</th>
<th>Area [mAU]</th>
<th>Height [mAU]</th>
<th>Area %</th>
</tr>
</thead>
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<tr>
<td>2</td>
<td>12.775</td>
<td>0.1065</td>
<td>2.79531</td>
<td>3.72277e-1</td>
<td>0.0363</td>
</tr>
<tr>
<td>3</td>
<td>13.166</td>
<td>0.1086</td>
<td>1.41470</td>
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<tr>
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<td>2.62768</td>
<td>0.2115</td>
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4.6.2. Chromatogram of Compound 5p

Below is a chromatogram of Compound 5p. The area percent report provides the following information:

<table>
<thead>
<tr>
<th>Peak</th>
<th>RetTime</th>
<th>Type</th>
<th>Width</th>
<th>Area</th>
<th>Height</th>
<th>Area %</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1.360</td>
<td>PB</td>
<td>0.0710</td>
<td>4.98226</td>
<td>1.08557</td>
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</tr>
<tr>
<td>2</td>
<td>14.124</td>
<td>BP</td>
<td>0.0979</td>
<td>9.63518e-1</td>
<td>1.59976e-1</td>
<td>0.0139</td>
</tr>
<tr>
<td>3</td>
<td>15.440</td>
<td>PBA</td>
<td>0.1303</td>
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<td>0.0863</td>
</tr>
<tr>
<td>4</td>
<td>15.998</td>
<td>MF R</td>
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<td>6888.30664</td>
<td>994.57733</td>
<td>99.2526</td>
</tr>
<tr>
<td>5</td>
<td>16.375</td>
<td>FM R</td>
<td>0.0945</td>
<td>8.25475</td>
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<tr>
<td>6</td>
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<tr>
<td>7</td>
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<td>3.49191</td>
<td>0.3542</td>
</tr>
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</table>

Totals: 6940.17598 1002.45504

4.7 Determination of LogP of Final Compounds 5a-q

Lipophilicity profiling of a newly synthesized compounds provides useful information for the forecast of oral bioavailability and biological effect. Therefore, LogP of all final compounds was determined experimentally by shake flask method [27]. The values were also predicted from the software ChemDraw Pro 13.0 [28] and compared with the experimental values.
4.7.1 Materials and Method

The n-octanol-water partition coefficients of compounds were measured using the traditional shake-flask technique at room temperature. The two phases, n-octanol and water were mutually saturated before the experiments. Stock solution of each test compound was prepared individually in n-octanol as well as in water by dissolving 1 mg of substance in 5 mL solvent by sonication. A standard curve of absorbance vs. concentration of test compounds in n-octanol as well as in water was prepared by making various dilutions from stock solution. The absorbance of each dilution was measured at λmax 254 nm using UV Spectrophotometer. The dilutions from the stock solution were chosen such that absorption ranges from 0.2 to 0.8.

Now, for the determination of logP, exactly 1 mg of the test compound was added into the accurately measured volumes of the two solvents together into the separating funnel and shaken vigorously for 45 minutes for complete exchange of the compound between the two phases. After that, both the phases were allowed to separate and then collected into two separate beakers. The n-octanol phase was pre-dried using anhydrous sodium sulfate and filtered. Before measuring the absorbance of each collected phase, they were diluted if required to get the absorbance readings in the range of standard plot. The concentrations of test compounds in each phase were then calculated from the standard curve of respective phase. The partition coefficient, logP was then calculated using following equation.

\[
\log P = \frac{C_0}{C_w}
\]

Where, Co is the concentration of test compound in n-octanol and Cw is the concentration of test compound in water.

The experimental values of logP of all the compounds were then compared with cLogP values predicted by the software ChemDraw Pro 13.0.

4.7.2 Results and Discussion

The results of the study are shown in Table 4.4 which indicates that experimental logP values are almost near to the cLogP values predicted by software. The logP range of 2-3 was considered as acceptable range for the desired activity of the compounds.
Table 4.4 Actual (experimental) and predicted logP values of synthesized molecules

<table>
<thead>
<tr>
<th>Compound</th>
<th>Predicted cLogP</th>
<th>Actual LogP</th>
<th>Compound</th>
<th>Predicted cLogP</th>
<th>Actual LogP</th>
</tr>
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<tr>
<td>5a</td>
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<td>2.56</td>
<td>5j</td>
<td>2.49</td>
<td>2.67</td>
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<tr>
<td>5b</td>
<td>2.94</td>
<td>3.10</td>
<td>5k</td>
<td>2.77</td>
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<td>5c</td>
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<td>2.58</td>
<td>5l</td>
<td>3.57</td>
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<tr>
<td>5d</td>
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<td>3.32</td>
<td>5m</td>
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<tr>
<td>5e</td>
<td>4.13</td>
<td>4.42</td>
<td>5n</td>
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</tr>
<tr>
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</tr>
<tr>
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<td>2.07</td>
<td>2.00</td>
<td>5p</td>
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</tr>
<tr>
<td>5h</td>
<td>--</td>
<td>2.85</td>
<td>5q</td>
<td>2.12</td>
<td>2.04</td>
</tr>
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<td>5i</td>
<td>2.33</td>
<td>2.66</td>
<td></td>
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<td></td>
</tr>
</tbody>
</table>

4.8 Conclusion

Total 17 3,7-disubstituted-1,4-dihydro-[1,2,4]triazolo[5,1-c][1,2,4]triazine derivatives were successfully synthesized in good yield and characterized by FT-IR, Mass (ESI-MS), $^1$H-NMR and $^{13}$C-NMR. Single crystal X-Ray crystallographic analysis of compound 5p was done to enrich the structure elucidation data as well as to know the crystal lattice arrangement of all the atoms in a molecule. Purity of all final compounds were determined using HPLC and was found >95% in each compound. Also, experimental logP was determined for each molecule by traditional shake flask method and compared with predicted clogP values from the software. Both values were found almost near to each other for every compound.

4.9 References

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[19] Least Squares function minimized: (SHELXL97)

[20] Standard deviation of an observation of unit weight: \( \sqrt{\frac{\sum (F_o^2-F_c^2)^2}{N_o-N_v}} \) where:

\( N_o = \) number of observations, \( N_v = \) number of variables


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