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Original article

Synthesis of novel alkyltriazole tagged pyrido[2,3-d]pyrimidine derivatives and their anticancer activity

C. Kurumurthy [a], P. Sambasiva Rao [a], B. Veera swamy [a], G. Santhosh kumar [a], P. Shanthan Rao [a], B. Narsaiah [a,*], L.R. Velatooru [b], R. Pamanji [b], J. Venkateswara Rao [b]

[a] Fluoroorganic Division, Indian Institute of Chemical Technology, Tarnaka, Hyderabad 500607, India
[b] Biology Division, Indian Institute of Chemical Technology, Tarnaka, Hyderabad 500607, India

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ABSTRACT

A series of novel alkyltriazole tagged pyrido[2,3-d]pyrimidine derivatives **5** and **6** was prepared starting from 2,3-active functional pyridine **1** *via* cyclization, propargylation followed by reaction with alkyl or perfluoroalkyl azides under Sharpless conditions. All the compounds **5** and **6** were screened for anticancer activity against three cancer cell lines such as U_{937}, THP-1 and Colo205. The promising compounds **5b** and **5e** have been identified.

1. Introduction

The pyrido[2,3-d]pyrimidine nucleus represents in many biologically active compounds which includes antitumour [1], antibacterial [2], anticonvulsant [3], antipyretic [4], analgesic [5], and CNS depressant activity [6]. Specifically pyrido[2,3-d]pyrimidines known to inhibit *Pneumocystis carinii*(pc), *Toxoplasma gondii*(tg) of tumour cell lines in culture [7] and the activity is attributed to inhibition of dihydrofolate reductase (DHFR) [8,9]. In recent past it was found that the fluorine [10] or trifluoromethyl [11,12] group at a strategic position of an organic molecule dramatically alters the properties of molecule in terms of lipid solubility, oxidative thermal stability, permeability, oral bioavailability thereby enhancement of transport mechanism.

The perfluoroalkyl triazoles [13,14] perfluoroalkyl tetrazole [15] and other fluoroalkyl derivatives [16–18] also found to show promising biological activity. Based on the importance and further to our ongoing research on the synthesis of hetero ring fused pyridines as potential molecules [19–21], we commenced our synthetic strategy towards a series of novel pyrido[2,3-d]pyrimidines, N-/O-propargyl and perfluoroalkyl triazole tagged pyrido[2,3-d]pyrimidines and screened for anticancer activity against three cancer cell lines such as U_{937}, THP-1 and Colo205. The promising compounds **5b**, **5e** have been identified and reporting here for the first time.

2. Chemistry

The 2-amino-3-cyano-4-trifluoromethyl-6-phenyl pyridine **1** [22] was reacted with various aliphatic acids in presence of catalytic amount of sulphuric acid at 120–130 °C for 12–20 h, obtained pyrido[2,3-d]pyrimidine derivatives **2**. The reaction sequence involves acylation of amine, hydrolysis of nitrile to amide followed by cyclization and dehydration to form products **2** in single step. However, reaction with aromatic acids under same conditions could not drive the reaction to the products. Alternatively compound **1** was reacted with benzaldehyde followed by MnO_2 oxidation resulted 2-phenyl pyrido[2,3-d]pyrimidine [23]. Efforts to introduce CF_3 or perfluoroalkyl group in second position of pyrido[2,3-d]pyrimidine under different set of conditions such as (i) CF_3COOH, conc. H_2SO_4, reflux, 14–16 h (ii) $(CF_3CO)_2O$, PTSA, reflux, 14–16 h (iii) $CF_3COOC_2H_5$, conc. H_2SO_4, reflux, 20 h. (iv) CF_3CF_2–COOH, PTSA, 100 °C, 15–16 h (v) CF_3COOH + $(CF_3CO)_2O$, PTSA, 60–70 °C, 15–16 h (vi) CF_3CF_2–COOH, conc. H_2SO_4, sealed tube, 120 °C, 10–12 h (vii) $(CF_3CO)_2O$, conc. H_2SO_4, 100 °C, sealed

* Corresponding author. Tel.: +91 40 27193630; fax: +91 40 27160387.
E-mail address: narsaiah_1984@yahoo.co.in (B. Narsaiah).

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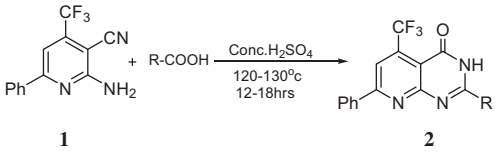
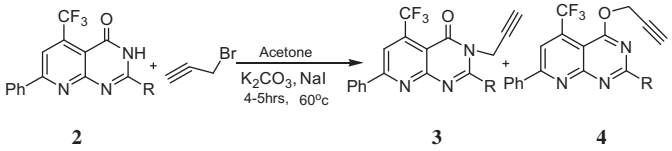


1 2

R = H, CH_3, C_2H_5, $(CH_3)_2CH$-, Ph, CH_3-$(CH_2)_4$-CH_2-



2 3 4

3 : 1

1 : 3 if R = Ph

R = H, CH_3, C_2H_5, $(CH_3)_2CH$-, Ph, CH_3-$(CH_2)_4$-CH_2-

Scheme 1. Synthesis of 2-substituted-5-trifluoromethyl-7-phenyl pyrido[2,3-d] pyrimidine-4(3H)-ones (**2a**–**f**) and N-/O-propargyl-2-substituted-5-trifluoromethyl-7-phenyl pyrido[2,3-d]pyrimidine-4(3H)-ones (**3a**–**f**)/pyrido[2,3-d]pyrimidines (**4a**–**f**).

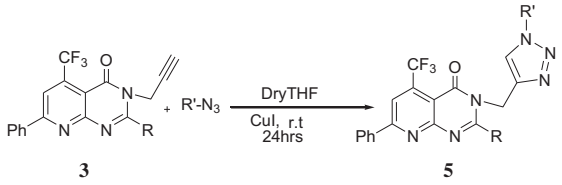
tube, 10–12 h could not give the product and recovered the starting material. Compound **2** when R = alkyl was further reacted with propargyl bromide in acetone using potassium carbonate as a base, obtained two regioisomers **3** and **4** (N-propargylated and O-propargylated) in definite proportion of 3:1 ratio. If R = aryl, the ratio is reversed to 1:3 and is attributed to steric hindrance as a result O-propargylated product as major. The ratio of products was established based on ^{1}H NMR data in which the signal for N–CH_2 appeared in the range δ 4.71 ppm–4.97 ppm, whereas signal for O–CH_2 appeared in the range δ 5.25 ppm to 5.44 ppm. The intensity of each signal is considered for estimating the ratio of each compound. The synthetic sequence is drawn in a Scheme 1 and products are tabulated in Table 1.

The two regioisomers such as N- and O-propargylated pyrido [2,3-d]pyrimidines **3** and **4** were separated based on their difference in polarity and each isomer was independently reacted with alkyl or perfluoroalkyl azide in THF, using copper(I) iodide as catalyst [24] and resulted exclusively 1,4-disubstituted-1,2,3-triazole derivatives **5** and **6** respectively as outlined in Schemes 2 and 3.

Table 1
Preparation of pyrido[2,3-d]pyrimidine derivatives **2a**–**f**, **3a**–**f**, and **4a**–**f**.

Compound no.	R	m. pt. °C	Yield (%)
2a	H	245–248	78
2b	CH_3	291–292	90
2c	C_2H_5	272–274	93
2d	$(CH_3)_2CH$	283–286	95
2e	CH_3-$(CH_2)_4$-CH_2	233–235	96
2f	C_6H_5	309–311	72
3a	H	220–223	73.5
4a	H	204–206	24.5
3b	CH_3	212–214	72.6
4b	CH_3	208–211	24.2
3c	C_2H_5	205–207	72
4c	C_2H_5	199–202	24
3d	$(CH_3)_2CH$	168–170	71.4
4d	$(CH_3)_2CH$	248–250	23.8
3e	CH_3-$(CH_2)_4$-CH_2	167–168	70.5
4e	CH_3-$(CH_2)_4$-CH_2	150–152	23.5
3f	C_6H_5	203–205	24.38
4f	C_6H_5	227–229	73.14

*All are new compounds except **2f**[23].



3 5

5a) R = H, R'= C_8F_{17}-CH_2-CH_2-, **5b**) R = H, R'= CH_3-$(CH_2)_8$-CH_2-

5c) R = CH_3, R' = C_6F_{13}-CH_2-CH_2, **5d**) R = CH_3, R' = C_8F_{17}-CH_2-CH_2-

5e) R = CH_3, R'= CH_3-$(CH_2)_8$-CH_2-, **5f**) R = CH_3, R'= Ph-

5g) R = C_2H_5, R' = C_6F_{13}-CH_2-CH_2, **5h**) R = C_2H_5, R' = C_8F_{17}-CH_2-CH_2-

5i) R = C_2H_5, R'= CH_3-$(CH_2)_8$-CH_2-, **5j**) R = $(CH_3)_2$ CH-, R'= C_8F_{17}-CH_2-CH_2-

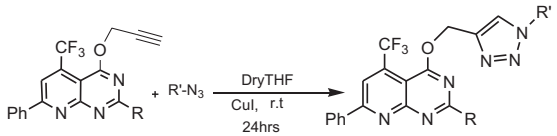
5k) R = CH_3-$(CH_2)_4$-CH_2-, R' = C_6F_{13}-CH_2-CH_2-, **5l**) R = CH_3-$(CH_2)_4$-CH_2-, R' = C_8F_{17}-CH_2-CH_2

Scheme 2. Synthesis of N-alkyltriazole tagged pyrido[2,3-d]pyrimidine derivatives (**5a**–**5l**).

The reaction is considered to take place *via* 1,3-dipolar cycloaddition of perfluoroalkyl azide to alkyne through a preformed copper acetylide complex formation under sharpless conditions [25,26]. The product yields were tabulated in Table 2.

3. Results and discussion

The compounds **5a**–**l** and **6a**–**c** were screened *in vitro* against three cancer cell lines such as U_{937} (human leukemic monocytic lymphoma), THP-1 (human acute monocytic leukemia) and Colo205 (human colourectal cancer) using MTT assay method [27]. IC_{50} values of the test compound for 24 h on each cell line was calculated and presented in a Table 3. Compound **5b** and **5e** exhibited decrease in cell viability in all the three test cell lines, however compound **5i**, **5j** and **6a** are active only on U_{937} and THP-1 cells. Similarly compound **5k** show activity against U_{937} and Colo205 cell lines and compound **5a** show against THP-1 and Colo205. Other compounds such as **5d**, **6b** and **6c** show activity only on U_{937} cell lines. The order of toxicity over U_{937} cell lines is **5e > 5k > 5b > 5j > 6b>**Etoposide. Structure activity relationship studies revealed that the presence of hydrogen or methyl group in second position and alkyltriazole tag in 3rd position promote high activity as experienced in compound **5b**, **5e** and is better than standard Etoposide against U_{937} cell lines. Among all the compounds, compound **5e** with methyl in 2nd position and alkyl tag in 3rd position found to show high activity. However, perfluoroalkyl tag on triazole is considered to be detrimental to the activity. Increase in aliphatic chain length on 2nd position has no additional advantage in promoting activity. Thus, promising compounds **5b** and **5e** will be further optimized in order to find a potential candidate for further evaluation. The activity data is tabulated in Table 3.



4 6

6a) R = CH_3, R'= C_8F_{17}-CH_2-CH_2-, **6b**) R = C_2H_5, R'= C_6F_{13}-CH_2-CH_2-

6c) R = Ph, R' = C_8F_{17}-CH_2-CH_2, **6d**) R = $(CH_3)_2$ CH-, R' = C_6F_{13}-CH_2-CH_2-

Scheme 3. Synthesis of O-alkyltriazole tagged pyrido[2,3-d]pyrimidine derivatives (**6a**–**6d**).

Table 2
Preparation of N-/O-alkyltriazole tagged pyrido[2,3-d]pyrimidine derivatives.

Compound no.	R	R′	m. pt. °C	Yield (%)
5a	H	$C_8F_{17}CH_2$-CH_2	180–182	95
5b	H	$CH_3(CH_2)_8$-CH_2	143–144	92
5c	CH_3	$C_6F_{13}CH_2$-CH_2	209–212	90
5d	CH_3	$C_8F_{17}CH_2$-CH_2	203–206	89
5e	CH_3	$CH_3(CH_2)_8$-CH_2	123–124	95
5f	CH_3	C_6H_5	215–217	96
5g	C_2H_5	$C_6F_{13}CH_2$-CH_2	141–144	86
5h	C_2H_5	$C_8F_{17}CH_2$-CH_2	182–185	90
5i	C_2H_5	$CH_3(CH_2)_8$-CH_2	145–147	93
5j	$(CH_3)_2CH$	$C_8F_{17}CH_2$-CH_2	186–188	91
5k	CH_3-$(CH_2)_4$-CH_2	$C_6F_{13}CH_2$-CH_2	149–151	89
5l	CH_3-$(CH_2)_4$-CH_2	$C_8F_{17}CH_2$-CH_2	147–148	87
6a	CH_3	$C_8F_{17}CH_2$-CH_2	156–159	85
6b	C_2H_5	$C_6F_{13}CH_2$-CH_2	124–126	88
6c	C_6H_5	$C_8F_{17}CH_2$-CH_2	290–293	89
6d	$(CH_3)_2CH$	$C_6F_{13}CH_2$-CH_2	173–175	86

4. Conclusion

A series of novel pyrido[2,3-d]pyrimidine derivatives **2** were prepared by an efficient route and subjected to alkylation to obtain two regioisomers **3** and **4** in definite proportion. Each regioisomer was separated based on their difference in polarity and cyclised with various azides to obtain alkyltriazole tagged final products **5** and **6**. The products **5** and **6** were screened for anticancer activity and promising compounds **5b** and **5e** have been identified.

5. Experimental

Melting points of all the compounds was recorded on Casia–Siamia (VMP-AM) melting point apparatus and are uncorrected. IR spectra were recorded on a Perkin–Elmer FT-IR 240-C spectrophotometer using KBr optics. ^{1}H NMR spectra were recorded on Bruker AV 300 MHz and INOVA 400 MHz in DMSO-d_6 or $CDCl_3$ using TMS as internal standard. Electron impact (EI) and chemical ionization mass spectra were recorded on a VG 7070 H instrument at 70 eV. All high-resolution spectra were recorded on QSTARXL hybrid MS/MS system (Applied Biosystems, USA) under electrospray ionization. All the reactions were monitored by thin layer chromatography (TLC) on precoated silica gel 60 F_{254} (mesh); spots were visualized with UV light. Merck silica gel (60–120 mesh) was used for column chromatography. CHN analyses were recorded on a vario EL analyzer.

Table 3
In vitro cytotoxicity of pyrido[2,3-d]pyrimidine derivatives against U_{937}, THP-1 and Colo205 cancer cell lines.

Compound no.	IC_{50} (μg/ml)		
	U_{937}	THP-1	Colo205
5a	–	79.12 ± 2.41	162.52 ± 5.62
5b	8.16 ± 0.68	16.91 ± 1.42	19.25 ± 1.46
5c	–	–	–
5d	124.23 ± 3.67	–	–
5e	6.20 ± 0.68	11.27 ± 1.67	15.01 ± 1.54
5f	–	–	–
5g	–	–	–
5h	–	–	–
5i	17.83 ± 1.35	82.65 ± 3.41	–
5j	8.35 ± 0.34	142.23 ± 5.24	–
5k	7.56 ± 0.74	–	132.42 ± 4.26
5l	–	–	–
6a	22.37 ± 1.73	107 ± 4.62	–
6b	9.22 ± 0.76	–	–
6c	44.35 ± 2.14	–	–
Etoposide (positive control)	17.94 ± 1.19	2.16 ± 0.15	7.24 ± 1.26

–indicated IC_{50} values are more than 200 μg/ml.

5.1. Preparation of 2-substituted-5-trifluoromethyl-7-phenyl pyrido [2,3-d]pyrimidin-4(3H)-ones (2a–f)

5.1.1. General procedure

A mixture of 2-amino-3-cyano-4-trifluoromethyl-6-phenyl pyridine (**1**) (1.9 mmol), aliphatic acids (5 ml) and a catalytic amount of conc. sulphuric acid was heated at 120–130 °C for 12–20 h. The reaction mixture was allowed to cool to room temperature, poured into ice cold water. The separated solid was collected by filtration, dried and recrystallized from ethanol.

*5.1.1.1. 7-Phenyl-5-(trifluoromethyl) pyrido[2,3-d]pyrimidine-4(3H)-one(**2a**).* IR (KBr, cm^{-1}): 3180 (-NH-), 1679 (lactam CO), 1344 (-C-F); ^{1}H NMR (DMSO, 300 MHz): δ 7.53–7.57 (m, 3H, Ar-H), 7.98 (s, 1H, Py-HC(2)), 8.20–8.29 (m, 3H, Ar-H), 12.72 (br, s, -NH-); MS (EI, 70 eV): *m*/*z*(%) M^+, 291(100), 263(40) (M^+-28), 222(25) (M^+-69); Anal. Calcd. for $C_{14}H_8F_3N_3O$: C 57.74, H 2.77, N 14.43%. Found: C 57.68, H 2.72, N 14.41%.

*5.1.1.2. 2-Methyl-7-phenyl-5-(trifluoromethyl) pyrido[2,3-d]pyrimidin-4(3H)-one(**2b**).* IR (KBr, cm^{-1}): 3188 (-NH-), 1688 (lactam CO), 1372 (-C-F); ^{1}H NMR (DMSO, 300 MHz): δ 2.51 (s, 3H, $-CH_3$), 7.50–7.55 (m, 3H, Ar-H), 8.10 (s, 1H, Py-HC(6)), 8.21–8.25 (m, 2H, Ar-H), 12.68 (br, s, -NH-); MS (EI, 70eV): *m*/*z*(%) M^+, 305(100), 277(25) (M^+-28), 236(40) (M^+-69); Anal. Calcd. for $C_{15}H_{10}F_3N_3O$: C 59.02, H 3.30, N 13.77%. Found: C 58.98, H 3.27, N 13.73%.

*5.1.1.3. 2-Ethyl-7-phenyl-5-(trifluoromethyl) pyrido[2,3-d]pyrimidin-4(3H)-one(**2c**).* IR (KBr, cm^{-1}): 3186 (-NH-), 1690 (lactam CO), 1370 (-C-F); ^{1}H NMR (DMSO, 300 MHz): δ 1.30 (t, 3H, J = 7.55 Hz, -CH_3), 2.71 (q, 2H, J = 7.55 Hz, -CH_2-), 7.58–7.61 (m, 3H, Ar-H), 8.28–8.33 (m, 3H, Ar-H), 12.65 (br, s, -NH-); MS (EI, 70 eV): *m*/*z*(%) M^+, 319(100), 291(15) (M^+-28), 250(40) (M^+-69); Anal. Calcd. for $C_{16}H_{12}F_3N_3O$: C 60.19, H 3.79, N 13.16%. Found: C 60.20, H 3.77, N 13.17%.

*5.1.1.4. 2-Isopropyl-7-phenyl-5-(trifluoromethyl) pyrido[2,3-d]pyrimidin-4(3H)-one(**2d**).* IR (KBr, cm^{-1}): 3187 (-NH-), 1677 (lactam CO), 1591 (-C=N), 1368 (-C-F); ^{1}H NMR (DMSO, 400 MHz): δ 1.36 (d, 6H, J = 7.168Hz, (-CH_3)$_2$), 2.99 (m, 1H, -CH-), 7.53–7.57 (m, 3H, Ar-H), 8.14 (s, 1H, Py-HC(6)), 8.23–8.28 (m, 2H, Ar-H), 12.57 (br, s, -NH-); MS (EI, 70 eV): *m*/*z*(%) M^+, 333(45), 318(100) (M^+-15), 305(20) (M^+-28), 264(15) (M^+-69); Anal. Calcd. for $C_{17}H_{14}F_3N_3O$: C 61.26, H 4.23, N 12.61%. Found: C 61.20, H 4.19, N 12.59%.

*5.1.1.5. 2-n-Hexyl-7-phenyl-5-(trifluoromethyl) pyrido[2,3-d]pyrimidin-4(3H)-one(**2e**).* IR (KBr, cm^{-1}): 3184 (-NH-), 1683 (lactam CO), 1591 (-C=N), 1368 (-C-F); ^{1}H NMR (DMSO, 300 MHz): δ 0.91 (t, 3H, J = 6.798Hz, -CH_3), 1.30–1.44 (m, 6H, -CH_2-CH_2-CH_2-), 1.79–1.88 (m, 2H, -CH_2-), 2.71 (t, 2H, J = 7.554Hz -CH_2-), 7.50–7.54 (m, 3H, Ar-H), 8.10 (s, 1H, Py-HC(6)), 8.21–8.26 (m, 2H, Ar-H), 12.60 (br, s, -NH-); MS (EI, 70 eV): *m*/*z*(%) M^+, 375(20), 360(10) (M^+-15), 346(25) (M^+-29), 305(100) (M^+-70); Anal. Calcd. for $C_{20}H_{20}F_3N_3O$: C 63.99, H 5.37, N 11.19%. Found: C 64.01, H 5.29, N 11.21%.

2,7-Diphenyl-5-(trifluoromethyl) pyrido[2,3-d]pyrimidine-4(3H)-one(**2f**)[23].

5.2. Preparation of N-/O-propargyl-2-substituted-5-trifluoromethyl-7-phenyl pyrido[2,3-d]pyrimidine-4(3H)-ones (3a–f/pyrido[2,3-d] pyrimidines (4a–f)

5.2.1. General procedure

Quinazol-4-one **2** (0.65 mmol) was dissolved in acetone (10 ml), K_2CO_3 (1.3 mmol), NaI (2 mg) was added. The propargyl bromide (1.9 mmol) was slowly added drop wise to the above mixture over

a period of 15 min at room temperature. Reaction mixture was refluxed for 3–4hrs and solvent was removed under reduced pressure. The residue was washed with n-hexane repeatedly to remove excess propargyl bromide, treated with distilled water to remove excess potassium carbonate and solid product separated was filtered, purified by eluting through column using 12% ethyl acetate in n-hexane for N-propargylated compound **3** and 30% ethyl acetate in n-hexane for O-propargylated compound **4**.

*5.2.1.1. 7-Phenyl-3-(prop-2-ynyl)-5-(trifluoromethyl) pyrido[2,3-d] pyrimidine-4(3H)-one(**3a**).* IR (KBr, cm^{-1}): 2117 (-C≡C-), 1696 (lactam CO), 1594 (-C=N), 1368 (-C-F); ^1H NMR ($CDCl_3$, 300 MHz): δ 2.57 (t, 1H, $J = 2.45$ Hz, -C≡CH), 4.84 (d, 2H, $J = 2.45$ Hz, $-CH_2$-N-), 7.52–7.55 (m, 3H, Ar-H), 8.21–8.26 (m, 3H, Ar-H), 8.63 (s, 1H, Py-HC(2)), MS (EI, 70eV): *m/z*(%) M^+, 329(100), 301(15) (M^+-28); Anal. Calcd. for $C_{17}H_{10}F_3N_3O$: C 62.01, H 3.06, N 12.76%. Found: C 61.91, H 3.02, N 12.69%.

*5.2.1.2. 2-Methyl-7-phenyl-3-(prop-2-ynyl)-5-(trifluoromethyl) pyrido[2,3-d]pyrimidine-4(3H)-one(**3b**).* IR (KBr, cm^{-1}): 2118 (-C≡C-), 1696 (lactam CO), 1593 (-C=N), 1368 (-C-F); ^1H NMR ($CDCl_3$, 300 MHz): δ 2.32 (t, 1H, $J = 2.45$ Hz, -C≡CH), 2.88 (s, 3H, $-CH_3$), 4.93 (d, 2H, $J = 2.45$ Hz, $-CH_2$-N-), 7.51–7.53 (m, 3H, Ar-H), 8.15 (s, 1H, Py-HC(6)), 8.21–8.24 (m, 2H, Ar-H), MS (EI, 70 eV): *m/z*(%) M^+, 343(100), 315(20) (M^+-28), 274(28) (M^+-69); Anal. Calcd. for $C_{18}H_{12}F_3N_3O$: C 62.97, H 3.52, N 12.24%. Found: C 63.01, H 3.49, N 12.26%.

*5.2.1.3. 2-Ethyl-7-phenyl-3-(prop-2-ynyl)-5-(trifluoromethyl) pyrido [2,3-d]pyrimidine-4(3H)-one(**3c**).* IR (KBr, cm^{-1}): 2126 (-C≡C-), 1687 (lactam CO), 1595 (-C=N), 1371 (-C-F); ^1H NMR ($CDCl_3$, 300 MHz): δ 1.55 (t, 3H, $J = 7.55$ Hz, $-CH_3$), 2.30 (t, 1H, $J = 2.26$ Hz, -C≡CH), 3.11 (q, 2H, $J = 7.55$ Hz, $-CH_2$-), 4.94 (d, 2H, $J = 2.26$ Hz, $-CH_2$-N-), 7.51–7.55 (m, 3H, Ar-H), 8.14 (s, 1H, Py-HC(6)), 8.20–8.25 (m, 2H, Ar-H), MS (EI, 70eV): *m/z*(%) M^+, 357(100), 329(23) (M^+-28), 288(10) (M^+-69); Anal. Calcd. for $C_{19}H_{14}F_3N_3O$: C 63.86, H 3.95, N 11.76%. Found: C 63.81, H 4.01, N 11.71%.

*5.2.1.4. 2-Isopropyl-7-phenyl-3-(prop-2-ynyl)-5-(trifluoromethyl) pyrido[2,3-d]pyrimidine-4(3H)one(**3d**).* IR (KBr, cm^{-1}): 2127 (-C≡C-), 1698 (lactam CO), 1590 (-C=N), 1371 (-C-F); ^1H NMR ($CDCl_3$, 300 MHz): δ 1.52 (d, 6H, $J = 6.04$ Hz, $(-CH_3)_2$), 2.31 (t, 1H, $J = 2.26$ Hz, -C≡CH), 3.44 (m, 1H, -CH-), 4.97 (d, 2H, $J = 2.26$ Hz, $-CH_2$-N-), 7.50–7.53 (m, 3H, Ar-H), 8.12 (s, 1H, Py-HC(6)), 8.19–8.23 (m, 2H, Ar-H), MS (EI, 70eV): *m/z*(%) M^+, 371(48), 356(100) (M^+-15); Anal. Calcd. for $C_{20}H_{16}F_3N_3O$: C 64.69, H 4.34, N 11.32%. Found: C 64.70, H 4.31, N 11.29%.

*5.2.1.5. 2-n-Hexyl-7-phenyl-3-(prop-2-ynyl)-5-(trifluoromethyl) pyrido[2,3-d]pyrimidine-4(3H)-one(**3e**).* IR (KBr, cm^{-1}): 2113 (-C≡C-), 1701 (lactam CO), 1593 (-C=N), 1370 (-C-F); ^1H NMR ($CDCl_3$, 300 MHz): δ 0.93 (t, 3H, $J = 6.79$ Hz, $-CH_3$), 1.36–1.42 (m, 4H, $-CH_2$-CH_2-), 1.49–1.57 (m, 2H, $-CH_2$-), 1.99 (pentet, 2H, $J = 8.31$ Hz, $-CH_2$-), 2.31 (t, 1H, $J = 2.26$ Hz, -C≡CH), 3.04 (t, 2H, $J = 8.31$ Hz, $-CH_2$-), 4.93 (d, 2H, $J = 2.26$ Hz, $-CH_2$-N-), 7.50–7.54 (m, 3H, Ar-H), 8.14 (s, 1H, Py-HC(6)), 8.20–8.25 (m, 2H, Ar-H), MS (EI, 70 eV): *m/z*(%) M^+, 413(10), 342(100) (M^+-71); Anal. Calcd. for $C_{23}H_{22}F_3N_3O$: C 66.82, H 5.36, N 10.16%. Found: C 66.79, H 5.32, N 10.13%.

*5.2.1.6. 2,7-Diphenyl-3-(prop-2-ynyl)-5-(trifluoromethyl) pyrido [2,3-d]pyrimidine-4(3H)-one(**3f**).* IR (KBr, cm^{-1}): 2117 (-C≡C-), 1685 (lactam CO), 1583 (-C=N), 1372 (-C-F); ^1H NMR ($CDCl_3$, 300 MHz): δ 2.34 (t, 1H, $J = 2.26$ Hz, -C≡CH), 4.71 (d, 2H, $J = 2.26$ Hz, $-CH_2$-N-), 7.50–7.58 (m, 6H, Ar-H), 7.82–7.86 (m, 2H, Ar-H), 8.20–8.27 (m, 3H, Ar-H); MS (EI, 70 eV): *m/z*(%) M^+, 405(100), 336(10) (M^+-69); Anal. Calcd. for $C_{23}H_{14}F_3N_3O$: C 68.15, H 3.48, N 10.37%. Found: C 68.18, H 3.42, N 10.32%.

*5.2.1.7. 7-Phenyl-4-(prop-2-ynyloxy)-5-(trifluoromethyl) pyrido[2,3-d]pyrimidine(**4a**).* IR (KBr, cm^{-1}): 2116 (-C≡C-), 1658 (-C=N), 1371 (-C-F), 1272 (C-O-C); ^1H NMR ($CDCl_3$, 400 MHz): δ 2.55 (s, 1H, -C≡CH), 5.25 (s, 2H, $-CH_2$-O-), 7.55–7.57 (m, 3H, Ar-H), 8.20–8.22 (m, 3H, Ar-H), 8.72 (s, 1H, Py-HC(2)), MS (EI, 70 eV): *m/z*(%) M^+, 329(15), 301(20) (M^+-28), 149(100); Anal. Calcd. for $C_{17}H_{10}F_3N_3O$: C 62.01, H 3.06, N 12.76%. Found: C 61.98, H 3.04, N 12.72%.

*5.2.1.8. 2-Methyl-7-phenyl-4-(prop-2-ynyloxy)-5-(trifluoromethyl) pyrido[2,3-d]pyrimidine(**4b**).* IR (KBr, cm^{-1}): 2127 (-C≡C-), 1667 (-C=N), 1373 (-C-F), 1262 (C-O-C); ^1H NMR ($CDCl_3$, 300 MHz): δ 2.51 (s, 1H, -C≡CH), 3.13 (s, 3H, $-CH_3$), 5.44 (s, 2H, $-CH_2$-O-), 7.49–7.54 (m, 3H, Ar-H), 8.13 (s, 1H, Py-HC(6)), 8.20–8.26 (m, 2H, Ar-H), MS (EI, 70eV): *m/z*(%) (M^+-H), 342(29), 301(100) (M^+-41), 273(38) (M^+-69); Anal. Calcd. for $C_{18}H_{12}F_3N_3O$: C 62.97, H 3.52, N 12.24%. Found: C 62.99, H 3.50, N 12.23%.

*5.2.1.9. 2-Ethyl-7-phenyl-4-(prop-2-ynyloxy)-5-(trifluoromethyl) pyrido[2,3-d]pyrimidine(**4c**).* IR (KBr, cm^{-1}): 2128 (-C≡C-), 1664 (-C=N), 1378 (-C-F), 1260 (C-O-C); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.43 (t, 3H, $J = 7.22$ Hz, $-CH_3$), 2.54 (t, 1H, $J = 2.71$ Hz, -C≡CH), 3.08 (q, 2H, J=7.22 Hz, $-CH_2$-), 5.37 (d, 2H, $J = 2.71$ Hz, $-CH_2$-O-), 7.55–7.58 (m, 3H, Ar-H), 8.19–8.23 (m, 3H, Ar-H), MS (EI, 70 eV): *m/z*(%) (M^+-H), 356(33), 301(100) (M^+-55), 273(32) (M^+-83); Anal. Calcd. for $C_{19}H_{14}F_3N_3O$: C 63.86, H 3.95, N 11.76%. Found: C 63.83, H 3.98, N 11.73%.

*5.2.1.10. 2-Isopropyl-7-phenyl-4-(prop-2-ynyloxy)-5-(trifluoromethyl) pyrido[2,3-d]pyrimidine(**4d**).* IR (KBr, cm^{-1}): 2117 (-C≡C-), 1664 (-C=N), 1372 (-C-F), 1258 (C-O-C); ^1H NMR ($CDCl_3$, 300 MHz): δ 1.47 (d, 6H, $J = 6.79$ Hz, $(-CH_3)_2$), 2.34 (t, 1H, $J = 2.26$ Hz, -C≡CH), 3.39 (m, 1H, -CH-), 5.34 (d, 2H, $J = 2.26$ Hz, $-CH_2$-O-), 7.54–7.57 (m, 3H, Ar-H), 8.11–8.15 (m, 3H, Ar-H); MS (EI, 70 eV): *m/z*(%) (M^+-H), 370(22), 356(68) (M^+-14), 301(100) (M^+-69); Anal. Calcd. for $C_{20}H_{16}F_3N_3O$: C 64.69, H 4.34, N 11.32%. Found: C 64.67, H 4.32, N 11.30%.

*5.2.1.11. 2-n-Hexyl-7-phenyl-4-(prop-2-ynyloxy)-5-(trifluoromethyl) pyrido[2,3-d] pyrimidine(**4e**).* IR (KBr, cm^{-1}): 2128 (-C≡C-), 1665 (-C=N), 1367 (-C-F), 1267 (C-O-C); ^1H NMR ($CDCl_3$, 400 MHz): δ 0.92 (t, 3H, $J = 6.59$Hz, $-CH_3$), 1.34–1.53 (m, 6H, $-CH_2$-CH_2-CH_2-), 1.94 (pentet, 2H, $J = 7.32$ Hz,$-CH_2$), 2.34 (s, 1H, -C≡CH), 2.95 (t, 2H, $J = 7.32$ Hz,$-CH_2$), 5.29 (s, 2H, $-CH_2$-O-), 7.53–7.58 (m, 3H, Ar-H), 8.11–8.15 (m, 3H, Ar-H); MS (EI, 70 eV): *m/z*(%) (M^+-H), 412(12), 342(100) (M^+-70); Anal. Calcd. for $C_{23}H_{22}F_3N_3O$: C 66.82, H 5.36, N 10.16%. Found: C 66.79, H 5.37, N 10.14%.

*5.2.1.12. 2,7-Diphenyl -4-(prop-2-ynyloxy)-5-(trifluoromethyl) pyrido[2,3-d]pyrimidine(**4f**).* IR (KBr, cm^{-1}): 2124 (-C≡C-), 1577 (-C=N), 1321 (-C-F); 1258 (C-O-C); ^1H NMR ($CDCl_3$, 300 MHz): δ 2.52 (t, 1H, $J = 2.26$ Hz, -C≡CH), 5.39 (d, 2H, $J = 2.26$ Hz, $-CH_2$-O-), 7.51–7.58 (m, 6H, Ar-H), 8.26–8.34 (m, 3H, Ar-H), 8.67–8.72 (m, 2H, Ar-H), MS (EI, 70 eV): *m/z*(%) (M^+-H), 404(100), 368(12); Anal. Calcd. for $C_{23}H_{14}F_3N_3O$: C 68.15, H 3.48, N 10.37%. Found: C 68.16, H 3.45, N 10.35%.

5.3. Preparation of N-/O-alkyltriazole-5-trifluoromethyl-7-phenyl pyrido[2,3-d]pyrimidin-4(3H)-ones (5a-l/pyrido[2,3-d]pyrimidines (6a–d)

5.3.1. General procedure

The propargylated quinazoline **3** or **4** (0.36 mmol) was dissolved in dry THF (5 ml) and catalytic amount of CuI was added. Then alkyl/perfluoroalkyl/aryl azides in dry THF was slowly added at

room temperature under nitrogen atmosphere and continued stirring for 24 h. The solvent was removed under reduced pressure, the residue was diluted with distilled water and extracted twice with ethyl acetate. The combined organic layer was dried over anhydrous Na_2SO_4 and concentrated to get the product. The crude product was purified by column chromatography.

*5.3.1.1. 3-((1-(3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-Heptadecafluorodecyl)-1H-1,2,3-triazol-4-yl)methyl)-7-phenyl-5-(trifluoromethyl) pyrido[2,3-d]pyrimidine-4(3H)-one(**5a**).* IR (KBr, cm^{-1}): 1696 (lactam CO), 1599 (-C=N), 1369 (-C-F); UV λ_{max} (nm): 334, 258; ^{1}H NMR ($CDCl_3$, 400 MHz): δ 2.76–2.87 (m, 2H, -CH_2-CF_2), 4.66 (t, 2H, J = 6.83 Hz, -CH_2-), 5.27 (s, 2H, -CH_2-N-), 7.51–7.54 (m, 3H, Ar-H), 7.88 (s, 1H, Triazol-H), 8.18–8.24 (m, 2H, Ar-H), 8.67 (s, 1H, Py-HC(2)); ^{13}C NMR ($CDCl_3$, 75 MHz): δ 31.67, 42.09, 42.53, 117.44, 117.53, 120.29, 122.13, 124.62, 124.70, 127.96, 129.11, 131.58, 136.32, 139.17, 141.70, 150.19, 150.46, 163.17, 163.30, 163.52; HRMS m/z Calcd. for $C_{27}H_{14}F_{20}N_6ONa$ ($[M + Na]^+$): 841.0807, Found 841.0835.

*5.3.1.2. 3-((1-Decyl-1H-1,2,3-triazol-4-yl)methyl)-7-phenyl-5-(trifluoromethyl) pyrido[2,3-d]pyrimidine-4(3H)-one(**5b**).* IR (KBr, cm^{-1}): 1690(lactam CO), 1594 (-C=N), 1366 (-C-F); UV λ_{max} (nm): 334, 258; ^{1}H NMR ($CDCl_3$, 300 MHz): δ 0.87 (t, 3H, J = 6.79 Hz, -CH_3), 1.21–1.33 (m, 16H, -$(CH_2)_7$-), 1.89 (m, 2H, -CH_2-), 4.32 (t, 2H, J = 7.55 Hz, -CH_2-N-), 5.27 (s, 2H, -CH_2-N-), 7.50–7.55 (m, 3H, Ar-H), 7.76 (s, 1H, Triazol-H), 8.17–8.25 (m, 3H, Ar-H), 8.70 (s, 1H, Py-HC(2)); HRMS m/z Calcd. for $C_{27}H_{31}F_3N_6ONa$ ($[M + Na]^+$): 535.2409, Found 535.2412.

*5.3.1.3. 2-Methyl-7-phenyl-3-((1-(3,3,4,4,5,5,6,6,7,7,8,8,8-tridecafluorooctyl)-1H-1,2,3-triazol-4-yl)methyl)-5-(trifluoromethyl) pyrido[2,3-d]pyrimidine-4(3H)-one(**5c**).* IR (KBr, cm^{-1}): 1694 (lactam CO), 1592 (-C=N), 1368 (-C-F); UV λ_{max} (nm): 334, 258; ^{1}H NMR ($CDCl_3$, 400 MHz): δ 2.77–2.86 (m, 2H, -CH_2-CF_2), 3.07 (s, 3H, -CH_3), 4.65 (t, 2H, J = 6.86Hz, -CH_2-CH_2-), 5.34 (s, 2H, -CH_2-N-), 7.50–7.52 (m, 3H, Ar-H), 7.86 (s, 1H, Triazol-H), 8.13 (s, 1H, Py-HC(6)), 8.20–8.23 (m, 2H, Ar-H); ^{13}C NMR ($CDCl_3$, 75 MHz): δ 23.86, 31.65, 40.07, 42.39, 109.65, 116.80, 120.38, 123.90, 124.92, 127.93, 128.98, 131.41, 136.48, 142.34, 159.22, 159.65, 163.53; HRMS m/z Calcd. for $C_{26}H_{17}F_{16}N_6O$ ($[M + H]^+$): 733.1123, Found 733.1125.

*5.3.1.4. 3-((1-(3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-Heptadecafluorodecyl)-1H-1,2,3-triazol-4-yl)methyl)-2-methyl-7-phenyl-5-(trifluoromethyl) pyrido[2,3-d]pyrimidine-4(3H)-one(**5d**).* IR (KBr, cm^{-1}): 1692 (lactam CO), 1598 (-C=N), 1372 (-C-F); UV λ_{max} (nm): 334, 258; ^{1}H NMR ($CDCl_3$, 400 MHz): δ 2.77–2.88 (m, 2H, -CH_2-CF_2), 3.07 (s, 3H, -CH_3), 4.65 (t, 2H, J = 7.45Hz, -CH_2-CH_2-), 5.34 (s, 2H, -CH_2-N-), 7.49–7.53 (m, 3H, Ar-H), 7.87 (s, 1H, Triazol-H), 8.13 (s, 1H, Py-HC(6)), 8.20–8.23 (m, 2H, Ar-H); ^{13}C NMR ($CDCl_3$, 75 MHz): δ 23.91, 31.70, 40.10, 42.43, 109.69, 116.86, 120.40, 124.01, 124.95, 127.97, 129.02, 131.44, 136.53, 142.36, 159.22, 159.67, 163.58; HRMS m/z Calcd. for $C_{28}H_{17}F_{20}N_6O$ ($[M + H]^+$): 833.1144, Found 833.1136.

*5.3.1.5. 3-((1-Decyl-1H-1,2,3-triazol-4-yl)methyl)-2-methyl-7-phenyl-5-(trifluoromethyl) pyrido[2,3-d]pyrimidin-4(3H)-one(**5e**).* IR (KBr, cm^{-1}): 1688 (lactam CO), 1587 (-C=N), 1366 (-C-F); UV λ_{max} (nm): 334, 258; ^{1}H NMR ($CDCl_3$, 400 MHz): δ 0.87 (t, 3H, J = 6.21Hz, -CH_3), 1.22–1.32 (m, 14H, -$(CH_2)_7$-), 1.90 (m, 2H, -CH_2-), 3.08 (s, 3H, -CH_3), 4.31 (t, 2H, J = 7.10Hz, -CH_2-N-), 5.34 (s, 2H, -CH_2-N-), 7.50–7.54 (m, 3H, Ar-H), 7.77 (s, 1H, Triazol-H), 8.13 (s, 1H, Py-HC(6)), 8.21–8.24 (m, 2H, Ar-H); HRMS m/z Calcd. for $C_{28}H_{33}F_3N_6ONa$ ($[M + Na]^+$): 549.2565, Found 549.2561.

*5.3.1.6. 2-Methyl-7-phenyl-3-((1-phenyl-1H-1,2,3-triazol-4-yl) methyl)- 5-(trifluoromethyl) pyrido[2,3-d]pyrimidin-4(3H)-one(**5f**).* IR (KBr, cm^{-1}): 1683 (lactam CO), 1591 (-C=N), 1370 (-C-F); UV λ_{max} (nm): 334, 256; ^{1}H NMR ($CDCl_3$, 300 MHz): δ 3.13 (s, 3H, -CH_3), 5.44 (s, 2H, -CH_2-N-), 7.42 (m, 1H, Ar-H), 7.48–7.54 (m, 5H, Ar-H), 7.73 (d, 2H, J = 7.74 Hz, Ar-H), 8.14 (s, 1H, Py-HC(6)), 8.20–8.26 (m, 3H, Ar-H); ^{13}C NMR ($CDCl_3$, 75 MHz): δ 23.91, 40.11, 109.68, 116.79, 120.49, 122.54, 127.92, 128.94, 129.70, 131.35, 136.47, 136.67, 142.57, 159.19, 159.71, 163.45; HRMS m/z Calcd. for $C_{24}H_{18}F_3N_6O$ ($[M + H]^+$): 463.1494, Found 463.1496.

*5.3.1.7. 2-Ethyl-7-phenyl-3-((1-(3,3,4,4,5,5,6,6,7,7,8,8,8-tridecafluorooctyl)-1H-1,2,3-triazol-4-yl)methyl)- 5-(trifluoromethyl) pyrido[2,3-d] pyrimidin-4(3H)-one(**5g**).* IR (KBr, cm^{-1}): 1690 (lactam CO), 1596 (-C=N), 1373 (-C-F); UV λ_{max} (nm): 335, 258; ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.56 (t, 3H, J = 7.36Hz, -CH_3), 2.73–2.90 (m, 2H, -CH_2-CF_2), 3.36 (q, 2H, J = 7.36Hz, -CH_2-), 4.65 (t, 2H, J = 7.74 Hz, -CH_2-N-), 5.36 (s, 2H, -CH_2-N-), 7.50–7.53 (m, 3H, Ar-H), 7.87 (s, 1H, Triazol-H), 8.12 (s, 1H, Py-HC(6)), 8.19–8.23 (m, 2H, Ar-H); ^{13}C NMR ($CDCl_3$, 75 MHz): δ 11.26, 28.90, 31.63, 39.27, 42.38, 109.62, 116.90, 124.95, 127.99, 128.94, 131.29, 136.63, 138.38, 138.89, 142.53, 159.35, 159.43, 163.13, 163.65; HRMS m/z Calcd. for $C_{27}H_{18}F_{16}N_6ONa$ ($[M + Na]^+$): 769.1184, Found 769.1176.

*5.3.1.8. 2-Ethyl-3-((1-(3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-heptadecafluorodecyl)-1H-1,2,3-triazol-4-yl)methyl)- 7-phenyl-5-(trifluoromethyl) pyrido[2,3-d]pyrimidin-4(3H)-one(**5h**).* IR (KBr, cm^{-1}): 1691 (lactam CO), 1592 (-C=N), 1369 (-C-F); UV λ_{max} (nm): 334, 258; ^{1}H NMR ($CDCl_3$, 300 MHz): δ 1.56 (t, 3H, J = 7.17 Hz, -CH_3), 2.81 (m, 2H, -CH_2-CF_2), 3.36 (q, 2H, J = 7.17 Hz, -CH_2-), 4.65 (t, 2H, J = 7.17 Hz, -CH_2-N), 5.35 (s, 2H, -CH_2-N-), 7.48–7.53 (m, 3H, Ar-H), 7.87 (s, 1H, Triazol-H), 8.11 (s, 1H, Py-HC(6)), 8.18–8.24 (m, 2H, Ar-H); ^{13}C NMR ($CDCl_3$, 75 MHz): δ 11.29, 28.94, 31.68, 39.30, 42.40, 109.65, 116.94, 124.96, 128.02, 128.96, 131.32, 136.67, 138.41, 138.91,142.56, 159.37, 159.46, 163.16, 163.70; HRMS m/z Calcd. for $C_{29}H_{18}F_{20}N_6ONa$ ($[M + Na]^+$): 869.1120, Found 869.1158.

*5.3.1.9. 3-((1-Decyl-1H-1,2,3-triazol-4-yl)methyl)-2-ethyl-7-phenyl-5-(trifluoromethyl) pyrido[2,3-d]pyrimidin-4(3H)-one(**5i**).* IR (KBr, cm^{-1}): 1684 (lactam CO), 1588 (-C=N), 1368 (-C-F); UV λ_{max} (nm): 335, 275, 259; ^{1}H NMR ($CDCl_3$, 300 MHz): δ 0.87 (t, 3H, J = 6.04 Hz, -CH_3), 1.22–1.33 (m, 14H, -$(CH_2)_7$-), 1.56 (t, 3H, J = 7.55 Hz, -CH_3), 1.89 (m, 2H, -CH_2-),3.38 (q, 2H, J = 7.55 Hz, -CH_2-), 4.30 (t, 2H, J = 6.79 Hz, -CH_2-N-), 5.35 (s, 2H, -CH_2-N), 7.49–7.54 (m, 3H, Ar-H), 7.76 (s, 1H, Triazol-H), 8.11 (s, 1H, Py-HC(6)), 8.19–8.24 (m, 2H, Ar-H); ^{13}C NMR ($CDCl_3$, 75 MHz): δ 11.34, 14.04, 22.59, 26.42, 28.90, 28.94, 29.17, 29.28, 29.38, 30.13, 31.77, 39.43, 50.47, 109.28, 116.89, 124.12, 128.02, 128.95, 131.26, 136.71, 138.48, 138.93, 142.05, 159.40, 159.45, 163.35, 163.60; HRMS m/z Calcd. for $C_{29}H_{35}F_3N_6ONa$ ($[M + Na]^+$): 563.2722, Found 563.2747.

*5.3.1.10. 3-((1-(3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-Heptadecafluorodecyl)-1H-1,2,3-triazol-4-yl)methyl)-2-isopropyl-7-phenyl-5-(trifluoromethyl) pyrido[2,3-d]pyrimidin-4(3H)-one(**5j**).* IR (KBr, cm^{-1}): 1698 (lactam CO), 1591 (-C=N), 1371 (-C-F); UV λ_{max} (nm): 335, 275, 258; ^{1}H NMR ($CDCl_3$, 300 MHz): δ 1.52 (d, 6H, J = 6.61 Hz, -$(CH_3)_2$), 2.73–2.90 (m, 2H, -CH_2-CF_2), 3.90–3.98 (m, 1H, -CH-), 4.65 (t, 2H, J = 7.55 Hz, -CH_2-CH_2-), 5.40 (s, 2H, -CH_2-N-), 7.49–7.54 (m, 3H, Ar-H), 7.87 (s, 1H, Triazol-H), 8.11 (s, 1H, Py-HC(6)), 8.18–8.23 (m, 2H, Ar-H); ^{13}C NMR ($CDCl_3$, 75 MHz): δ 21.02, 31.67, 32.35, 38.99, 42.41, 109.64, 117.02, 117.10, 120.44, 124.20, 124.85, 128.12, 128.95, 131.23, 136.85, 142.79, 159.53, 159.72, 163.89, 166.75; HRMS m/z Calcd. for $C_{30}H_{20}F_{20}N_6ONa$ ($[M + Na]^+$): 883.1276, Found 883.1314.

*5.3.1.11. 2-n-Hexyl-7-phenyl-3-((1-(3,3,4,4,5,5,6,6,7,7,8,8,8-tridecafluorooctyl)-1H-1,2,3-triazol-4-yl)methyl)- 5-(trifluoromethyl) pyrido[2,3-d]pyrimidin-4(3H)-one(**5k**).* IR (KBr, cm^{-1}): 1691 (lactam CO), 1590 (-C=N), 1371 (-C-F); UV λ_{max} (nm): 335, 276, 258; ^{1}H NMR ($CDCl_3$, 300 MHz): δ 0.93 (t, 3H, J = 6.79 Hz, -CH_3), 1.35–1.44 (m, 4H, -$(CH_2)_2$-), 1.53–1.63 (m, 2H, -CH_2-), 1.93–2.03 (m, 2H, -CH_2-),

2.73–2.90 (m, 2H, -CH_2-CF_2-), 3.30 (t, 2H, J = 7.93 Hz, -CH_2-), 4.65 (t, 2H, J = 7.55 Hz, -CH_2-CH_2-), 5.35 (s, 2H, -CH_2-N-), 7.49–7.58 (m, 3H, Ar-H), 7.87 (s, 1H, Triazol-H), 8.11 (s, 1H, Py-HC(6)), 8.18–8.23 (m, 2H, Ar-H); ^{13}C NMR ($CDCl_3$, 75 MHz): δ 13.99, 22.49, 27.44, 29.12, 31.52, 35.73, 39.54, 42.41, 116.92, 124.91, 128.05, 128.98, 131.34, 136.68, 142.62, 159.40, 159.53, 162.68, 162.89, 163.71; HRMS *m/z* Calcd. for $C_{31}H_{27}F_{16}N_6O$ ($[M + H]^+$): 803.1913, Found 803.1915.

*5.3.1.12. 3-((1-(3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-Heptadecafluorodecyl)-1H-1,2,3-triazol-4-yl)methyl)-2-hexyl-7-phenyl-5-(trifluoromethyl) pyrido[2,3-d]pyrimidine-4(3H)-one(**5l**).* IR (KBr, cm^{-1}): 1691 (lactam CO), 1590 (-C=N), 1370 (-C-F); UV λ_{max} (nm): 336, 276, 259; ^{1}H NMR ($CDCl_3$, 300 MHz): δ 0.94 (t, 3H, J = 6.73 Hz, -CH_3), 1.36–1.44 (m, 4H, -$(CH_2)_2$-), 1.55–1.61 (m, 2H, -CH_2-), 1.95–2.02 (m, 2H, -CH_2-), 2.76–2.87 (m, 2H, -CH_2-CF_2-), 3.30 (t, 2H, J = 7.69 Hz, -CH_2-), 4.65 (t, 2H, J = 7.55 Hz, -CH_2-CH_2-), 5.35 (s, 2H, -CH_2-N-), 7.50–7.53 (m, 3H, Ar-H), 7.86 (s, 1H, Triazol-H), 8.12 (s, 1H, Py-HC(6)), 8.20–8.23 (m, 2H, Ar-H); ^{13}C NMR ($CDCl_3$, 75 MHz): δ 13.95, 22.47, 27.40, 29.10, 31.47, 35.63, 39.61, 42.47, 116.79, 124.63, 128.12, 129.08, 131.43, 136.78, 142.73, 159.38, 159.62, 162.71, 162.94, 163.83; HRMS *m/z* Calcd. for $C_{33}H_{26}F_{20}N_6ONa$ ($[M + Na]^+$): 925.1746, Found 925.1779.

*5.3.1.13. 4-((1-(3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-Heptadecafluorodecyl)-1H-1,2,3-triazol-4yl)methoxy)-2-methyl-7-phenyl-5-(trifluoromethyl) pyrido[2,3-d]pyrimidine(**6a**).* IR (KBr, cm^{-1}): 1658 (-C=N), 1372 (-C-F), 1205 (C-O-C); UV λ_{max} (nm): 341, 251; ^{1}H NMR ($CDCl_3$, 400 MHz): δ 2.78–2.89 (m, 2H, -CH_2-CF_2), 3.10 (s, 3H, -CH_3), 4.63 (t, 2H, J = 7.56Hz, -CH_2-N-), 5.89 (s, 2H, -CH_2-O-), 7.53–7.57(m, 3H, Ar-H), 7.68 (s, 1H, Triazol-H), 7.98 (s, 1H, Py-HC(6)), 8.14–8.19 (m, 2H, Ar-H); HRMS *m/z* Calcd. for $C_{28}H_{16}F_{20}N_6ONa$ ($[M + Na]^+$): 855.0963, Found 855.0985.

*5.3.1.14. 2-Ethyl-7-phenyl-4-((1-(3,3,4,4,5,5,6,6,7,7,8,8,8-tridecafluorooctyl)-1H-1,2,3-triazol-4-yl)methoxy)- 5-(trifluoromethyl) pyrido[2,3-d] pyrimidine(**6b**).* IR (KBr, cm^{-1}): 1672 (-C=N), 1373 (-C-F), 1206 (C-O-C); UV λ_{max} (nm): 342, 252; ^{1}H NMR ($CDCl_3$, 300 MHz): δ 1.40 (t, 3H, J = 7.17 Hz, -CH_3), 2.68–2.85 (m, 2H, -CH_2-CF_2), 3.15 (q, 2H, J = 7.17 Hz, -CH_2-), 4.62 (t, 2H, J = 7.36Hz, -CH_2-N-), 5.87 (s, 2H, -CH_2-O-), 7.52–7.56 (m, 3H, Ar-H), 7.67 (s, 1H, Triazol-H), 7.97 (s, 1H, Py-HC(6)), 8.13–8.18 (m, 2H, Ar-H); HRMS *m/z* Calcd. for $C_{27}H_{19}F_{16}N_6O$ ($[M + H]^+$): 747.1312, Found 747.1315.

*5.3.1.15. 4-((1-(3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-Heptadecafluorodecyl)-1H-1,2,3-triazol-4yl)methoxy)-2,7-diphenyl-5-(trifluoromethyl) pyrido[2,3-d]pyrimidine(**6c**).* IR (KBr, cm^{-1}): 1670 (-C=N), 1369 (-C-F), 1203 (C-O-C); UV λ_{max} (nm): 340, 275; ^{1}H NMR ($CDCl_3$, 300 MHz): δ 2.76–2.95 (m, 2H, -CH_2-CF_2-), 4.71 (t, 2H, J = 7.29 Hz, -CH_2-N-), 6.00 (s, 2H, -CH_2-O-), 7.51–7.58 (m, 6H, Ar-H), 7.78 (s, 1H, Triazol-H), 8.24–8.32 (m, 3H, Ar-H), 8.68–8.72 (m, 2H, Ar-H); HRMS *m/z* Calcd. for $C_{33}H_{19}F_{20}N_6O$ ($[M + H]^+$): 895.1300, Found 895.1326.

*5.3.1.16. 2-Isopropyl-7-phenyl-4-((1-(3,3,4,4,5,5,6,6,7,7,8,8,8-tridecafluorooctyl)-1H-1,2,3-triazol-4-yl)methoxy)- 5-(trifluoromethyl) pyrido[2,3-d]pyrimidine(**6d**).* IR (KBr, cm^{-1}): 1664 (-C=N), 1377 (-C-F), 1241 (C-O-C); UV λ_{max} (nm): 342, 252; ^{1}H NMR ($CDCl_3$, 300 MHz): δ 1.24 (d, 6H, J = 6.42Hz, -$(CH_3)_2$), 2.79–2.95 (m, 2H, -CH_2-CF_2), 3.92–4.1 (m, 1H, -CH-), 4.67 (t, 2H, J = 6.23 Hz, -CH_2-N-), 5.92 (s, 2H, -CH_2-O-), 7.52–7.61 (m, 3H, Ar-H), 8.20–8.29 (m, 3H, Ar-H), 8.39 (s, 1H, Triazol-H); HRMS *m/z* Calcd. for $C_{28}H_{21}F_{16}N_6O$ ($[M + H]^+$): 761.1400, Found 761.1406.

5.4. Cell lines and cell culture

The cell lines U_{937} (human leukaemic monocytic lymphoma), THP-1 (human acute monocytic leukaemia) and Colo205 (human colourectal cancer) cell lines were obtained from the National Centre for Cellular Sciences (NCCS), Pune, India. Cells were cultured in RPMI-1640 media, supplemented with 10% heat-inactivated fetal bovine serum (FBS), 1 mM $NaHCO_3$, 2 mM L-glutamine, 100 units/ml penicillin and 100 μg/ml streptomycin. All cell lines were maintained in culture at 37 °C in an atmosphere of 5% CO_2.

5.5. Test concentrations

Initially, the test compound was dissolved in DMSO (8 mg/ml), from which 50 μl of stock was diluted to 1 ml in culture medium to obtain experimental stock concentration of 400 μg/ml. Different aliquots of experimental stock were added to the cultured cells in the medium (final volume of 200 μl) to attain the required concentrations.

5.6. Cytotoxicity

In all experiments, different cell lines were seeded at a final density of 2×10^4 cells/well, in 96 well microtiter plates. Cytotoxicity was measured using the MTT [3-(4,5-dimethylthiazol-2-yl)-2, 5-diphenyl tetrazolium bromide] assay, according to the method of Mosmann (1983) [27]. Briefly, the cells (2×10^4) were seeded in each well containing 0.1 ml of medium in 96 well plates. After overnight incubation, the cells were treated with different test concentrations of test compounds (5–200 μg/ml) at identical conditions with five replicates each. The cell viability was assessed after 24 h, by adding 10 μl of MTT (5 mg/ml) per well. The plates were incubated at 37 °C for additional 3 h. The medium was discarded and the formazan blue, which formed in the cells, was dissolved with 100 μl of DMSO. The rate of colour production was measured at 570 nm in a spectrophotometer (Spectra MAX Plus; Molecular Devices; supported by SOFTmax PRO-5.4). The percent inhibition of cell viability was determined with reference to the control values (without test compound). The data were subjected to linear regression analysis and the regression lines were plotted for the best straight-line fit. The IC_{50} (inhibition of cell viability) concentrations were calculated using the respective regression equation.

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