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ORIGINAL ARTICLE

Synthesis and biological evaluation of novel flavone/triazole/benzimidazole hybrids and flavone/isoxazole-annulated heterocycles as antiproliferative and antimycobacterial agents

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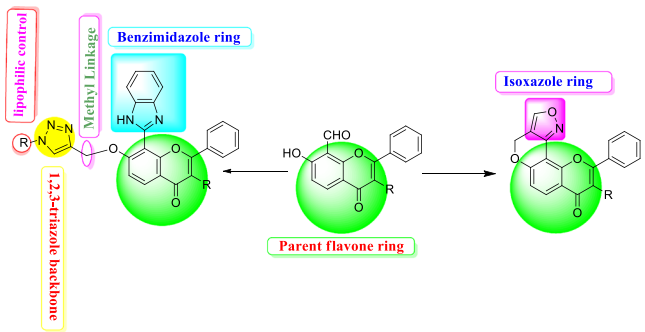
Abstract

A series of new flavone/isoxazole fused heterocycles **5a–f** and flavone/1,2,3-triazole/benzimidazole hybrid heterocycles compounds **7a–t** were synthesized via an intramolecular cyclization and Cu(I)-catalyzed click 1,3-dipolar cycloaddition. The products were evaluated for their antiproliferative activity against human breast cancer cell line (MCF-7) using sulforhodamine B assay (SRB) and antimycobacterial activity using turbidometric assay. The majority of the tested compounds exhibited antiproliferative activity and antimycobacterial activity. Compounds **7l**, **7q** and **7r** showed moderate antiproliferative activity with IC50 values 17.9, 14.2, 19.1 μM, respectively, and compound **5a** showed moderate antimycobacterial activity with 41.7% of inhibition at 30 μM concentration.

Graphical Abstract



Keywords 8-Formyl-7-hydroxy flavones · Click chemistry · 1, 2, 3-Triazole · Antimycobacterial activity · Antiproliferative activity · Cycloaddition

Introduction

The synthesis and functionalization of flavones has become a major area of focus for synthetic organic chemists because of the varied pharmacological activities flavone derivatives exhibit [1]. Flavones are a class of natural products most

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Fig. 1 Chemical structures of some anti cancerous flavones

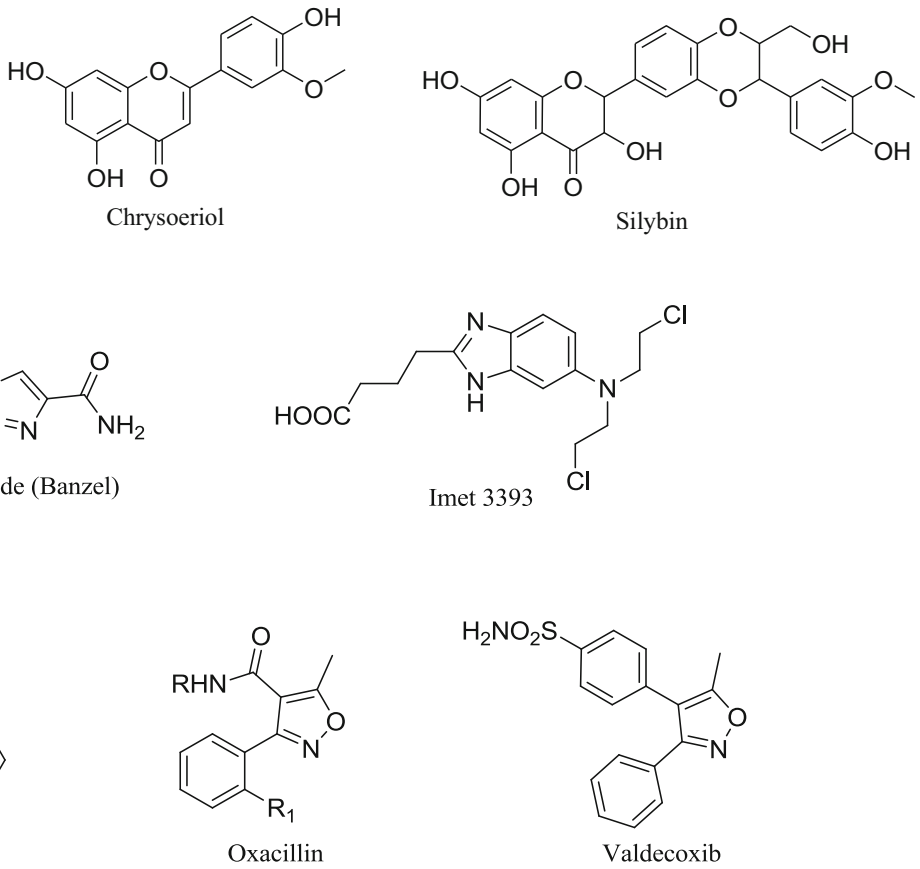


Fig. 2 Pharmacologically important triazole, benzimidazole and isoxazole based drugs

abundant in fruits, seeds, nuts and flowers and are important components of human diet. These compounds exhibit antioxidant [2], antihypertensive [3], antiallergic [4], antibacterial and antifungal [5,6] activities, and can act as plant growth regulators [7]. Flavones have a number of positive features among which the most significant is their antioxidant effects, which provide enhanced protection of the body against the harmful effects of free radical reactions [8]. Hydroxy flavones and their derivatives are potential anticancer as well as antibacterial and antifungal agents [9,10]. Moreover, heterocyclic ring pendent or fused at 7/8 positions of flavones is reported to possess anticancer activities [11]. Examples of commercially available anticancer flavone drugs are depicted in Fig. 1 [12].

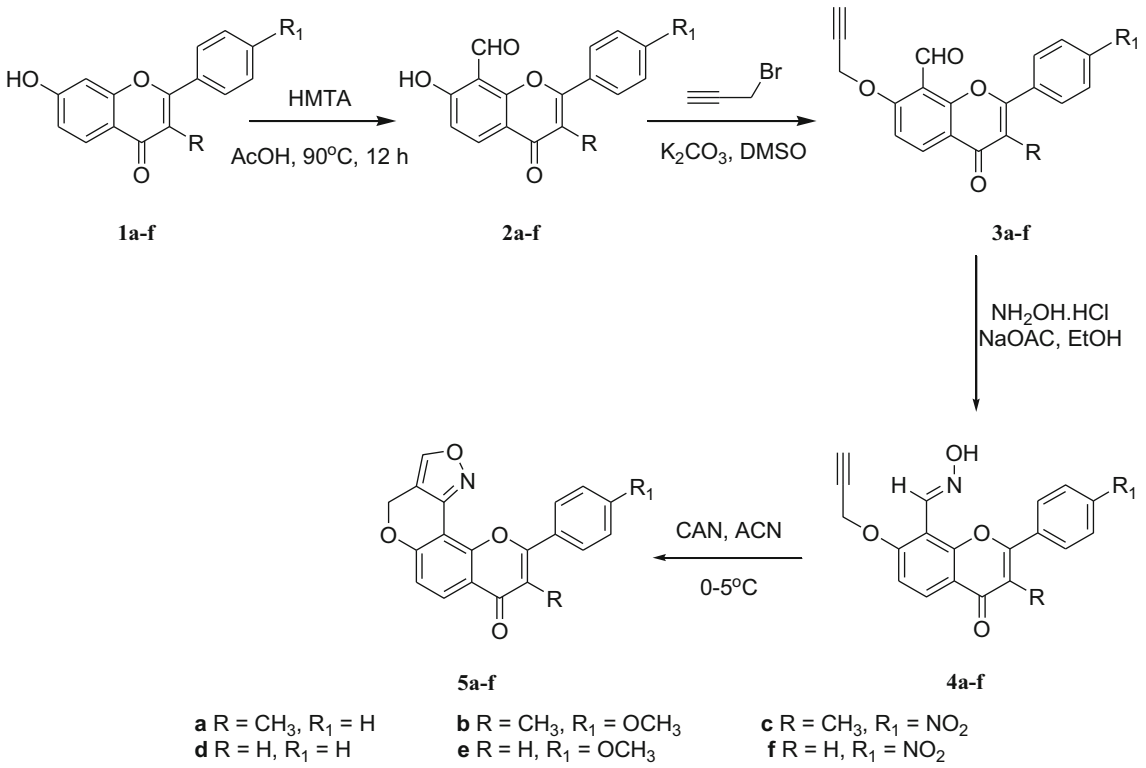
Triazoles are a class of N-heterocyclic compounds with diversified biological activities such as antimicrobial [13], antiprotozoal [14], anti-inflammatory [15], antitubercular [16], antibacterial [17], anticancer [18], antimalarial [19], antiviral [20] and antiproliferative activities [21,22]. Benzimidazole scaffolds are of special interest to medicinal chemistry because its derivatives possess antioxidant, antimicrobial, antihelmintic, anticancer, antihypertensive, antineoplastic, anti-inflammatory, analgesic, antiprotozoal, antihepatitis B virus, antiulcer, antiviral, antifungal and anticonvulsant activities [23,24]. Isoxazoles are important structural moieties, which are largely used in pharmaceuticals and therapeutics [25–27]. Examples of pharmacologically important triazole-, benzimidazole- and isoxazole- based drugs are shown in Fig. 2.

In continuation of our efforts on structural modification of hydroxy flavone moieties [28,29] to enhance their biological activity, we planned for the synthesis of novel hybrid heterocycles having benzimidazole, triazole and isoxazole scaffolds in a single molecule by adopting Cu(I)-catalyzed click chemistry and intramolecular cyclization reactions.

Results and discussion

Chemistry

A new series of heterocycles have been synthesized by incorporating triazole and isoxazole moieties at the 7 and 8 positions of 8-formyl-7-hydroxy flavone. Further, these new series of compounds were tested for their antimycobacterial activity and antiproliferative activity against human breast cancer cell line (MCF-7). 7-Hydroxy flavones **1a–f** were treated with HMTA/acetic acid at 100 °C to obtain 8-formyl-



Scheme 1 Synthesis of flavone/isoxazole heterocyclic compounds

7-hydroxy flavones **2a–f** [30,31]. Compounds **2a–f** were coupled with propargyl bromide in acetone/K_2CO_3 medium to get 4-oxo-2-phenyl-7-(prop-2-ynyloxy)-4*H*-chromene-8-carbaldehydes **3a–f**. The formyl groups of compounds **3a–f** were converted into corresponding oximes **4a–f** by treating them with hydroxyl amine hydrochloride in sodium acetate/ethanol medium. Compounds **4a–f** were subjected to intramolecular cyclization using ceric ammonium nitrate (CAN) and acetonitrile at 0–5 °C to yield final compounds **5a–f** (Scheme 1). The structures of compounds **5a–f** were confirmed by spectral analysis.

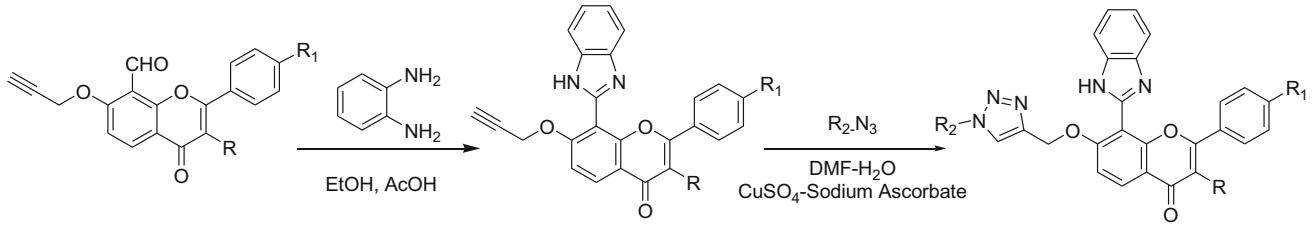
The next step of the present study was the preparation of novel flavone/triazole/benzimidazole hybrid heterocycles **7a–t** from **3a** and **3d**. Compounds **3a** and **3d** underwent smooth condensation with orthophenylenediamine (OPDA) in ethanol/acetic acid medium to yield compounds **6a** and **6d**, respectively. These were further reacted with aromatic and aliphatic azides in the presence of $CuSO_4$ and sodium ascorbate in DMF − H_2O medium. The terminal alkyne of **6a** and **6d** underwent 1,4 regioselective 1,3-dipolar cycloaddition (click chemistry) and produced 7-((1-alkyl/aralkyl-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[*d*]imidazol-2-yl)-3-methyl-2-phenyl-4*H*-chromen-4-ones in good yields **7a–t** (Scheme 2). All the synthesized products **7a–t** were characterized by ^{1}H NMR, ^{13}C NMR, FTIR and mass spectral analysis.

Biological activity

The newly synthesized compounds **5a–f** and **7a–t** were screened for their in vitro antiproliferative activity against breast cancer cell line MCF-7 using standard SRB (Sulforhodamine B), and the results are summarized in Table 1. Among the tested compounds, one compound **7q** showed moderate activity against MCF-7 cell line with IC_{50} value 14.5 μM, two compounds **7l** and **7r** with IC_{50} values of 17.9 and 19.1 μM, respectively, and nine compounds **7j**, **7d**, **7t**, **5a**, **7g**, **7f**, **7s**, **7p** and **5b** showed less activity with IC_{50} values of 25.1, 30.8, 31.9, 34.2, 37.6, 40.2, 41.2, 42.3 and 45.2 μM, respectively. The remaining compounds in the series showed little to no antiproliferation against MCF-7 cell line.

Our structure–activity relationship study revealed that the orientation of a substituent on 1,2,3-triazole ring and flavone/benzimidazole hybrid is crucial for inducing antiproliferative activity against MCF-7 cancer cell line. In particular, compounds **7q**, **7l** and **7r** showed the best antiproliferative profile of this compound series with IC_{50} values of 14.2, 17.9, 19.1 μM, respectively. The activity was further enhanced in the presence of an electron withdrawing group at the fourth position of the benzene ring attached to the triazole hybrid.

Additionally, compounds **5a–f** and **7a–t** were evaluated for antimycobacterial activity against *Mycobacterium bovis* strain (BCG) using a turbidometric assay. The screening



3a and 3d — **6a and 6d** — **7a-t**

3a R = CH_3, R_1 = H
3d R = H, R_1 = H

6a R = CH_3, R_1 = H
6d R = H, R_1 = H

7a R = CH_3, R_1 = H, R_2 = C_6H_5
7b R = CH_3, R1 = H, R_2 = p-Cl-C_6H_4
7c R = CH_3, R_1 = H, R_2 = m-CH_3-C_6H_4
7d R = CH_3, R_1 = H, R_2 = p-NO_2-C_6H_4
7e R = CH_3, R_1 = H, R_2 = m-Cl-C_6H_4
7f R = CH_3, R_1 = H, R_2 = 2,6-$(CH_3)_2$-C_6H_3
7g R = CH_3, R_1 = H, R_2 = p-NO_2-Benzyl
7h R = CH_3, R_1 = H, R_2 = p-F-Benzyl
7i R = CH_3, R_1 = H, R_2 = Cyclopentyl
7j R = CH_3, R_1 = H, R_2 = Hexyl
7k R = H, R_1 = H, R_2 = C_6H_5
7l R = H, R_1 = H, R_2 = p-Cl-C_6H_4
7m R = H, R_1 = H, R_2 = m-CH_3-C_6H_4
7n R = H, R_1 = H, R_2 = p-NO_2-C_6H_4
7o R = H, R_1 = H, R_2 = m-Cl-C_6H_4
7p R = H, R_1 = H, R_2 = 2,6-$(CH_3)_2$-C_6H_3
7q R = H, R_1 = H, R_2 = p-NO_2-Benzyl
7r R = H, R_1 = H, R_2 = p-F-Benzyl
7s R = H, R_1 = H, R_2 = Cyclopentyl
7t R = H, R_1 = H, R_2 = Hexyl

Scheme 2 Synthesis of new flavone/triazole/benzimidazole hybrid heterocyclic compounds

Table 1 IC_{50} μM values of tested compounds against BCG and MCF-7

Entry	Tested compounds	BCG values % inhibition	STD DEV	MCF-7 IC_{50}(μM)	SD
1	7a	12.7	0.040	NA	NA
2	7b	NA		NA	NA
3	7c	0.1	0.048	NA	NA
4	7d	4.0	0.043	30.8	0.003
5	7e	0.2	0.057	99.7	0.102
6	7f	1.1	0.050	40.2	0.039
7	7g	1.4	0.045	37.6	0.052
8	7h	11.6	0.063	NA	**NA**
9	7i	8.7	0.052	91.4	0.066
10	7j	26.2	0.012	25.1	0.472
11	7k	13.1	0.005	NA	**NA**
12	7l	13.6	0.013	**17.9**	0.001
13	7m	6.3	0.025	NA	NA
14	7n	18.4	0.011	NA	NA
15	7o	12.2	0.017	NA	NA
16	7p	2.1	0.057	42.3	0.004
17	7q	5.7	0.026	**14.2**	0.018
18	7r	0.1	0.005	**19.1**	0.083
19	7s	5.2	0.084	41.2	0.001
20	7t	27.3	0.054	31.9	0.008
21	5a	**41.7**	0.012	34.2	0.052
22	5b	1.9	0.009	45.2	0.078
23	5c	9.9	0.045	92.7	0.009
24	5d	14.4	0.064	68.7	0.472
25	5e	8.9	0.025	90.9	0.213
26	5f	9.0	0.019	NA	NA
	Doxorubicin	–	–	6.2 μM	–
	Rifampicin	97%	–	–	–

bold values indicate with moderate activity

results revealed that the series of compounds **5a–f** and **7a–t** inhibited the growth of the bacteria with a MIC range of 0.1 to > 40 mg/mL. Compound **5a** was found to be associated with moderate antimycobacterial activity with BCG% inhibition value of 41.7.

Conclusion

In summary, 2 libraries of new hybrid heterocyclic compounds, 9-alkyl-10-arylpyrano[2′,3′:5,6]chromeno[4,3-c] isoxazol-8(4*H*)-ones **5a–f** and 7-((1-alkyl/aryl-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[*d*]imidazol- 2-yl)- 2-phenyl-4*H*-chromen-4-ones **7a–t**, were synthesized by using an intramolecular cyclization and click chemistry approach. Compounds **7q**, **7l** and **7r** showed moderate antiproliferative activity against human breast cancer cell line MCF-7, and compound **5a** showed moderate antimycobacterial activity against bovis strain. From this, we were able to identify a few active molecules which are capable of inhibiting the growth of Mycobacterium bovis strain (BCG) and human breast cancer cell line MCF-7.

Experimental

All reagents were purchased from Merck and commercial sources and were used without further purification. Melting points (m.p.) were determined in open capillary tubes on a Buchi 530 melting point apparatus and are uncorrected. Thin layer chromatography (TLC) was performed to monitor progress of the reaction and assess purity of the compounds; spots were detected by their absorption under UV light. IR spectra were recorded using an IR prestige-21 (FTIR, Shimadzu). Mass spectra were recorded using a 'Hewlett–Packard' HP GS/MS 5890/5972. ^{1}H NMR spectra were recorded on a Bruker DPX operating at 400 MHz in $CDCl_3$ or DMSO − d_6 solvent, using tetramethylsilane (TMS) as an internal standard. Chemical shifts are shown as δ values (ppm); the coupling constants (J) are expressed in Hertz (Hz). Signals are represented as s (singlet), d (doublet), t (triplet), q (quartet) or m (multiplet).

General procedure for the synthesis of 8-formyl-7-hydroxyflavones (2a–f)

7-Hydroxyflavones **1a–f** (1.0 mmol) dissolved in glacial acetic acid (20 mL), hexamethylenetetramine (HMTA) (4.0 mmol) were added, and the resulting solution was stirred at 100 °C for 6 h. The solution was treated with dil. HCl (1:1) followed by further heating for 30min; it was diluted to 500 mL with cold water and left overnight in a refrigerator. Product was filtered, dried and purified by column chromatography in ethyl acetate/*n*-hexane (9:1) to get 8-formyl-7-hydroxyflavones **2a–f**.

General procedure for the synthesis of 4-oxo-2-aryl-7-(prop-2-ynyloxy)-4*H*-chromene-8-carbaldehydes (3a–f)

7-Hydroxyflavone **1a-f** (1.0 mmol) dissolved in acetic acid (20 mL), then hexamethylenetetramine (HMTA) (4.0 mmol) was added, and the resulting solution was stirred at 90 °C for1 h. The reaction was treated with cold water and filtered. The separated product was dried and recrystallized from methanol to give 4-oxo-2-aryl-7-(prop-2-ynyloxy)-4*H*-chromene-8-carbaldehydes **3a–f**.

3-Methyl-4-oxo-2-phenyl-7-(prop-2-ynyloxy) -4*H*-chromene-8-carbaldehyde (3a)

Off-white solid, yield 75%; m.p. 240–242 °C. IR (KBr, cm^{-1}): 1725 (CHO), 1690 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.24 (s, 3H, CH_3), 2.63 (s, 1H, CH), 4.97 (s, 2H, CH_2), 7.53–7.55 (m, 4H, Ar–H), 7.79–7.81 (m, 2H, Ar–H), 8.49 (d, J = 9.14 Hz, 1H, Ar–H), 10.65 (s, 1H, CHO) ppm. ESI-MS: m/z 319 $[M + H]^+$.

2-(4-Methoxyphenyl)-3-methyl-4-oxo-7-(prop-2-ynyloxy)-4*H*-chromene-8-carbaldehyde (3b)

Off-white solid, yield 72%; m.p. 239–240 °C. IR (KBr, cm^{-1}): 1729 (CHO), 1695 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.19 (s, 3H, CH_3), 2.65 (s, 1H, CH), 3.90 (s, 3H, OCH_3), 4.89 (s, 2H, CH_2), 7.50–7.53 (m, 4H, Ar–H), 7.72–7.75 (m, 2H, Ar–H), 10.55 (s, 1H, CHO) ppm. ESI-MS: m/z 349 $[M + H]^+$.

3-Methyl-2-(4-nitrophenyl)-4-oxo-7-(prop-2-ynyloxy) -4*H*-chromene-8-carbaldehyde (3c)

Off-white solid, yield 70%; m.p. 243–244 °C. IR (KBr, cm^{-1}): 1727 (CHO), 1700 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.27 (s, 3H, CH_3), 2.67 (s, 1H, CH), 5.01 (s, 2H, CH_2), 7.54–7.57 (m, 2H, Ar–H), 7.75–7.77 (m, 2H, Ar–H), 8.29 (d, J = 9.14 Hz, 2H, Ar–H), 10.63 (s, 1H, CHO) ppm. ESI-MS: m/z 364 $[M + H]^+$.

4-Oxo-2-phenyl-7-(prop-2-ynyloxy)-4*H*-chromene-8-carbaldehyde (3d)

Off-white solid, yield 75%; m.p. 235–237 °C. IR (KBr, cm^{-1}): 1729 (CHO), 1699 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.64 (s, 1H, CH), 5.01 (s, 2H, CH_2), 6.81 (s, 1H, Ar–H), 7.49–7.53 (m, 4H, Ar–H), 7.78–7.83 (m, 2H, Ar–H), 8.39 (d, J = 9.14 Hz, 1H, Ar–H), 10.52 (s, 1H, CHO) ppm. ESI-MS: m/z 305 $[M + H]^+$.

2-(4-Methoxyphenyl)-4-oxo-7-(prop-2-ynyloxy) -4*H*-chromene-8-carbaldehyde (3e)

Off-white solid, yield 71%; m.p. 238–239 °C. IR (KBr, cm^{-1}): 1739 (CHO), 1716 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.60 (s, 1H), 3.89 (s, 3H, OCH_3), 5.05 (s, 2H, CH_2), 6.84 (s, 1H, Ar–H), 7.52–7.56 (m, 4H, Ar–H), 7.80–7.84 (m, 2H, Ar–H), 10.59 (s, 1H, CHO) ppm. ESI-MS: m/z 335 $[M + H]^+$.

2-(4-Nitrophenyl)-4-oxo-7-(prop-2-ynyloxy)-4*H*-chromene-8-carbaldehyde (3f)

Off-white solid, yield 70%, m.p. 242–244 °C. IR (KBr, cm^{-1}): 1735 (CHO), 1706 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.65 (s, 1H), 4.99 (s, 2H, CH_2), 6.83 (s, 1H, Ar–H), 7.55–7.59 (m, 2H, Ar–H), 7.75–7.79 (m, 2H, Ar–H), 8.59 (d, J = 9.14 Hz, 2H, Ar–H), 10.63 (s, 1H, CHO) ppm. ESI-MS: m/z 350 $[M + H]^+$.

General procedure for the synthesis of (E)-3-alkyl-4-oxo-2-aryl-7-(prop-2-yn-1-yloxy) -4*H*-chromene-8-carbaldehyde oximes (4a–f)

To a stirred solution of 4-oxo-2-phenyl-7-(prop-2-ynyloxy)-4*H*-chromene-8-carbaldehydes **3a–f** (1.0 mmol) in EtOH was added sodium acetate (1.5 mmol) and hydroxylamine hydrochloride (2.0 mmol) at room temperature. The reaction mixture was stirred for 1 h at reflux temperature. After completion of reaction, as judged by TLC, solvent was removed and diluted with cold water. The obtained precipitate was filtered, dried and recrystallized from methanol to give the products **4a–f**.

(E)-3-Methyl-4-oxo-2-phenyl-7-(prop-2-yn-1-yloxy) -4*H*-chromene-8-carbaldehyde oxime (4a)

Off-white solid, yield 80%; m.p. 250–252 °C. IR (KBr, cm^{-1}): 1739 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.08 (s, 3H, CH_3), 3.72 (s, 1H, CH), 5.07 (s, 2H, CH_2), 7.35–7.37 (d, J = 8.71 Hz, 1H, Ar–H), 7.58–7.59 (m, 4H, Ar–H), 7.81 (s, 2H, Ar–H), 8.10–8.12 (d, J = 8.71 Hz, 1H, CH), 11.58 (s, 1H, OH) ppm. ESI-MS: m/z 334 $[M + H]^+$.

(E)-2-(4-Methoxyphenyl)-3-methyl-4-oxo-7-(prop-2 -yn-1-yloxy)-4*H*-chromene-8-carbaldehyde oxime (4b)

Off-white solid, yield 78%; m.p. 253–255 °C. IR (KBr, cm^{-1}): 1725 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$)δ 2.06 (s, 3H, CH_3), 3.61 (s, 1H, CH), 3.91 (s, 3H, OCH_3), 5.02 (s, 2H, CH_2), 7.32–7.36 (d, J = 8.79 Hz, 2H, Ar–H), 7.55–7.57 (m, 2H, Ar–H), 7.76 (s, 2H, Ar–H), 8.08–8.10 (d, J = 8.79 Hz, 1H, CH), 11.54 (s, 1H, OH) ppm. ESI-MS: m/z 365 $[M+H]^+$.

(E)-3-Methyl-2-(4-nitrophenyl)-4-oxo-7-(prop-2 -yn-1-yloxy)-4*H*-chromene-8-carbaldehyde oxime (4c)

Off-white solid, yield 76%; m.p. 252–254 °C. IR (KBr, cm^{-1}): 1729 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.12 (s, 3H, CH_3), 3.62 (s, 1H, CH), 5.21 (s, 2H, CH_2), 7.42–7.45 (d, J = 8.71 Hz, 2H, Ar–H), 7.62–7.64 (m, 2H, Ar–H), 7.87 (s, 2H, Ar–H), 8.13–8.15 (d, J = 8.71 Hz, 1H, CH), 11.59 (s, 1H, OH) ppm. ESI-MS: m/z 379 $[M + H]^+$.

(E)-4-Oxo-2-phenyl-7-(prop-2-yn-1-yloxy)-4*H*-chromene -8-carbaldehyde oxime (4d)

Off-white solid, yield 79%; m.p. 249–250 °C. IR (KBr, cm^{-1}): 1735 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.72 (s, 1H, CH), 5.21 (s, 2H, CH_2), 6.79 (s, 1H, Ar–H), 7.29–7.31 (d, J = 8.71 Hz, 1H, Ar–H), 7.47–7.49 (m, 4H, Ar–H), 7.69 (s, 2H, Ar–H), 8.01–8.02 (d, J = 8.71 Hz, 1H, CH), 11.42 (s, 1H, OH) ppm. ESI-MS: m/z 320 $[M + H]^+$.

(E)-2-(4-Methoxyphenyl)-4-oxo-7-(prop-2-yn-1-yloxy) -4*H*-chromene-8-carbaldehyde oxime (4e)

Off-white solid, yield 74%; m.p. 254–255 °C. IR (KBr, cm^{-1}): 1719 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.63 (s, 1H, CH), 3.95 (s, 3H, OCH_3), 5.01 (d, J = 1.40 Hz, 2H, CH_2), 6.91 (s, 1H, Ar–H), 7.45–7.47 (d, J = 8.71 Hz, 2H, Ar–H), 7.48–7.49 (m, 2H, Ar–H), 7.71 (s, 2H, Ar–H), 8.00–8.02 (d, J = 8.71 Hz, 1H, CH), 11.47(s, 1H, OH) ppm. ESI-MS: m/z 350 $[M + H]^+$.

(E)-2-(4-Nitrophenyl)-4-oxo-7-(prop-2-yn-1-yloxy) -4*H*-chromene-8-carbaldehyde oxime (4f)

Off-white solid, yield 72%; m.p. 256–258 °C. IR (KBr, cm^{-1}): 1740 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$)δ 3.55 (s, 1H, CH), 5.17 (S, 2H, CH_2), 6.84 (s, 1H, Ar–H), 7.42–7.45 (d, J= 8.71 Hz, 2H, Ar–H), 7.60–7.62 (m, 2H, Ar–H), 7.86 (s, 2H, Ar–H), 8.17–8.19 (d, J = 8.71 Hz, 1H, CH), 11.49 (s, 1H, OH) ppm. ESI-MS: m/z364 $[M + H]^+$.

General procedure for the synthesis of 9-alkyl-10-arylpyrano[2′,3′:5,6]chromeno[4,3-c]isoxazol-8(4*H*)-ones (5a–f)

Compounds **4a–f** (1.0 mmol) were dissolved in acetonitrile and then the ceric ammonium nitrate (2.0 mmol) at 0–5 °C was added; the resulting mixture was stirred at 5 °C for 30 min. The reaction mixture was extracted with chloroform and wash with brine solution. The crude material was purified by column chromatography in chloroform/methanol (9:1) to give the products **5a–f**.

9-Methyl-10-phenylpyrano[2′,3′:5,6]chromeno[4,3-c]isoxazol-8(4*H*)-one (5a)

Light brown color solid, yield 70%; m.p. 298–300 °C. IR (KBr, cm^{-1}): 1623 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$)δ 2.28 (s, 3H, CH_3), 5.39 (s, 2H, O-CH_2), 7.08 (d, J = 8.70 Hz, 1H, Ar–H), 7.54 (m, 5H, Ar–H), 8.26 (m, 2H, Ar–H) ppm. ^{13}C NMR(100 MHz, DMSO − d_6) δ 11.8, 62.2, 102.7, 110.3, 115.8, 116.6, 116.8, 128.4, 128.6, 129.1, 129.3, 130.5, 132.3, 150.2, 152.5, 159.0, 160.0, 176.3 ppm. ESI-MS: m/z 332 $[M + H]^+$. Anal. calcd. for $C_{20}H_{13}NO_4$: C, 72.50; H, 3.95; N, 4.23%. Found: C, 72.02; H, 4.12; N, 4.32%.

10-(4-Methoxyphenyl)-9-methylpyrano[2′,3′:5,6]chromeno[4,3-c]isoxazol-8(4*H*)-one (5b)

Light brown color solid, yield 68%; m.p. 299–300 °C. IR (KBr, cm^{-1}): 1628 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.19 (s, 3H, CH_3), 3.73 (s, 3H, OCH_3), 5.24 (s, 2H, OCH_2), 7.06 (d, J = 8.70 Hz, 1H, Ar–H), 7.55 (m, 4H, Ar–H), 8.28 (m, 2H, Ar–H) ppm. ^{13}**C** NMR (100 MHz, DMSO − d_6) δ 12.1, 56.5, 62.8, 102.7, 110.3, 115.8, 116.6, 116.8, 128.4, 128.6, 129.0, 129.4, 130.1, 132.5, 150.0, 152.7, 159.9, 160.2, 176.8 ppm. ESI-MS: m/z 362 $[M+1]^+$. Anal. calcd. for $C_{21}H_{15}NO_5$: C, 69.80; H, 4.18; N, 3.88%. Found: C, 70.01; H, 3.98; N, 3.70%.

9-Methyl-10-(4-nitrophenyl)pyrano[2′,3′:5,6]chromeno[4,3-c]isoxazol-8(4*H*)-one (5c)

Light brown color solid, yield 65%; m.p. 295–296 °C. IR (KBr, cm^{-1}): 1630 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) 2.31 (s, 3H, CH_3), 5.42 (s, 2H, O-CH_2), 7.11 (d, J= 8.70 Hz, 1H, Ar–H), 7.56 (m, 4H, Ar–H), 8.29 (m, 2H, Ar–H) ppm. ^{13}**C** NMR (100 MHz, DMSO-d_6) δ 12.1, 63.4, 102.7, 110.3, 115.8, 116.6, 116.8, 128.4, 128.6, 129.0, 129.4, 130.1, 132.5, 148.9, 152.7, 159.9, 160.2, 177.5 ppm. ESI-MS: m/z 377 $[M + H]^+$. Anal. calcd. for $C_{20}H_{12}N_2O_6$: C, 63.83; H, 3.21; N, 7.44%. Found: C, 64.01; H, 3.31; N, 7.24%.

10-Phenylpyrano[2′,3′:5,6]chromeno[4,3-c]isoxazol-8(4*H*)-one (5d)

Light brown color solid, yield 69%; m.p. 297–298 °C. IR (KBr, cm^{-1}): 1633 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$)δ 5.39 (s, 2H, O-CH_2), 6.71 (s, 1H, Ar–H), 7.07 (d, J = 8.70 Hz, 1H, Ar–H), 7.57 (m, 5H, Ar–H), 8.30 (m, 2H, Ar–H) ppm. ^{13}C NMR (100 MHz, DMSO-d_6) δ 65.2, 102.7, 110.3, 115.8, 116.6, 116.8, 128.4, 128.6, 129.0, 129.4, 130.1, 132.5, 148.9, 152.7, 159.9, 160.2, 176.3 ppm. ESI-MS: m/z 318 $[M + H]^+$. Anal. calcd. for $C_{19}H_{11}NO_4$: C, 71.92; H, 3.49; N, 4.41%. Found: C, 72.32; H, 3.38; N, 4.30%.

10-(4-Methoxyphenyl)pyrano[2′,3′:5,6]chromeno[4,3-c]isoxazol-8(*4H*)-one (5e)

Light brown color solid, yield 62%; m.p. 293–295 °C. IR (KBr, cm^{-1}): 1627 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.89 (s, 3H, OCH_3), 5.45 (s, 2H, O − CH_2), 7.05 (d, J = 8.70 Hz, 1H, Ar–H), 7.56 (m, 5H, Ar–H), 8.18 (m, 2H, Ar–H) ppm. ^{13}**C** NMR (100 MHz, DMSO-d_6) δ 56.5, 64.5, 102.7, 110.3, 115.8, 116.6, 116.8, 128.4, 128.6, 129.0, 129.4, 130.1, 132.5, 148.9, 152.7, 159.9, 160.2, 176.3 ppm. ESI-MS: m/z 348 $[M + H]^+$. Anal. calcd. for $C_{20}H_{13}NO_5$: C, 69.16; H, 3.77; N, 4.03%. Found: C, 69.80; H, 4.18; N, 3.88%.

10-(4-Nitrophenyl)pyrano[2′,3′:5,6]chromeno[4,3-c]isoxazol-8(4*H*)-one (5f)

Light brown color solid, yield 65%; m.p. 297–299 °C. IR (KBr, cm^{-1}): 1635 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.39 (s, 2H, O − CH_2), 7.08 (d, J = 8.70 Hz, 1H, Ar–H), 7.54 (m, 5H, Ar–H), 8.26 (m, 2H, Ar–H) ppm. ^{13}**C** NMR (100 MHz, DMSO − d_6)δ 63.4, 102.7, 110.3, 115.8, 116.6, 116.8, 128.4, 128.6, 129.0, 129.4, 130.1, 132.5, 148.9, 152.7, 159.9, 160.2, 177.5 ppm. ESI-MS: m/z 363 $[M + H]^+$. Anal. calcd. for $C_{19}H_{10}N_2O_6$: C, 62.99; H, 2.78; N, 7.73%. Found: C, 63.83; H, 3.21; N, 7.44%.

General procedure for the synthesis of 8-(1*H*-benzo[*d*]imidazol-2-yl)-2-phenyl-7-(prop-2-ynyloxy)-4*H*-chromen-4-one (6a and 6d)

4-Oxo-2-phenyl-7-(prop-2-ynyloxy)-4*H*-chromene-8-carbaldehydes **3a** and **3d** (1.0 mmol) were dissolved in 50 mL of ethanol, orthophenylenediamine (OPDA) (1.0 mmol) was added, and 4–5 drops of acetic acid were added and the resulting solution was stirred at 80 °C for 4 h. The solvent was evaporated, and ice-cold water was added. The obtained precipitate was filtered and purified by column chromatography using ethyl acetate/hexane (9:2) to get 8-(1*H*-benzo[*d*]imidazol-2-yl)-2-phenyl-7-(prop-2-ynyloxy)-4*H*-chromen-4-ones **6a** and **6d**.

8-(1*H*-Benzo[d]imidazol-2-yl)-3-methyl-2-phenyl-7-(prop-2-ynyloxy)-4*H*-chromen-4-one (6a)

Light brown color solid, yield 79%; m.p. 260–261 °C. IR (KBr, cm^{-1}): 3086 (NH), 1717 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.16 (s, 3H, CH_3), 2.57 (s, 1H, CH), 4.84 (s, 2H, CH_2), 7.15–7.17 (d, J = 9.03 Hz, 1H, Ar–H), 7.28–7.37 (m, 5H, Ar–H), 7.48 (s, 1H, Ar–H), 7.65–7.67 (m, 2H, Ar–H), 7.82 (s, 1H, Ar–H), 8.25 (d, J = 8.70 Hz, 1H, Ar–H), 10.52 (s, 1H, NH) ppm. ESI-MS: m/z 407 $[M + H]^+$.

8-(1*H*-Benzo[d]imidazol-2-yl)-2-phenyl-7-(prop-2-ynyloxy)-4*H*-chromen-4-one (6d)

Light brown color solid, yield 80%; m.p. 259–261 °C. IR (KBr, cm^{-1}): 3079 (NH), 1719 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.58 (s, 1H, CH), 4.85 (s, 2H, CH_2), 6.84 (s, 1H, Ar–H), 7.14–7.16 (d, J = 9.03 Hz, 1H, Ar–H), 7.31–7.39 (m, 5H, Ar–H), 7.51 (s, 1H, Ar–H), 7.62–7.64 (m, 2H, Ar–H), 7.84 (s, 1H, Ar–H), 8.30 (d, J = 8.70 Hz, 1H, Ar–H), 10.61 (s, 1H, NH) ppm. ESI-MS: m/z 393 $[M + H]^+$

General procedure for the synthesis of 7-((1-alkyl/aryl-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[*d*]imidazol-2-yl)-2-phenyl-4*H*-chromen-4-ones (7a–t)

8-(1*H*-Benzo[*d*]imidazol-2-yl)-2-phenyl-7-(prop-2-ynyloxy)-4*H*-chromen-4-ones **6a** and **6d** (1 mmol) and the corresponding alkyl or aryl azides (1 mmol) were dissolved in 8 mL of DMF. The reaction mixture was stirred at room temperature for 10 min, and then $CuSO_4 \cdot 5H_2O$ (1 mmol) and sodium ascorbate (0.2 mmol) were added. The reaction mixture was stirred at room temperature until the starting material was consumed as judged by TLC analysis. The precipitate was filtered, dried and purified by column chromatography to give the products 7-((1-alkyl/aryl-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[*d*]imidazol-2-yl)-2-phenyl- 4*H*-chromen-4-ones **7a–t** as light brown solids with 60–80% yield.

7-((1-Phenyl-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-2-phenyl-4*H*-chromen-4-one (7a)

Light brown color solid, yield 80%; m.p. 240–242 °C. IR (KBr, cm^{-1}): 3086 (NH), 1739 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.07 (s, 3H, CH_3), 5.50 (s, 2H, CH_2), 7.50–7.79 (m, 17H, Ar–H), 8.87 (s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO-d_6) δ 13.8, 53.2, 102.5, 109.5, 110.5, 115.8, 116.4, 116.8, 117.8, 128.0, 128.4, 128.6, 128.8, 129.1, 129.3, 129.9, 130.5, 131.8, 132.3, 152.5, 152.8, 160.0, 177.1 ppm. ESI-MS: m/z 526 $[M + H]^+$. Anal. calcd. for $C_{32}H_{23}N_5 = O_3$: C, 73.13; H, 4.41; N, 13.33%. Found: C, 72.79; H, 4.49; N, 13.67%.

7-((1-(4-Chlorophenyl)-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-3-methyl-2-phenyl-4*H*-chromen-4-one (7b)

Light brown color solid, yield 75%; m.p. 239–240 °C. IR (KBr, cm^{-1}): 3084 (NH), 1740 (C=O). ^{1}H NMR(400 MHz, $CDCl_3$) δ 2.07(s, 3H, CH_3), 5.36 (s, 2H, CH_2), 7.19 (s, 1H, Ar–H), 7.53–7.79 (m, 15H, Ar–H), 8.87(s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO-d_6) δ 13.8, 52.7, 104.3, 111.1, 115.3, 116.9, 123.0, 126.4, 128.0, 128.7, 128.9, 130.4, 131.3, 131.6, 134.3, 138.9, 144.5, 152.9, 154.2, 160.4, 163.9, 177.8 ppm. ESI-MS: m/z 560 $[M + H]^+$. Anal. calcd. for $C_{32}H_{22}N_5O_3Cl$: C, 68.63; H, 3.96; N, 12.51%. Found: C, 69.83; H, 3.36; N, 12.93%.

7-((1-m-Toulyl-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-3-methyl-2-phenyl-4*H*-chromen-4-one (7c)

Light brown color solid, yield 79%; m.p. 236–237 °C. IR (KBr, cm^{-1}): 3086(NH), 1739 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.09 (m, 6H, CH_3), 5.52 (s, 2H, CH_2), 7.31–7.71(m, 16H, Ar–H), 8.80 (s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO-d_6) δ 12.0, 24.3, 72.6, 104.3, 109.5, 111.1, 115.3, 116.9, 123.0, 124.3, 126.4, 126.9, 128.0, 128.4, 128.7, 130.4, 131.6, 138.4, 138.9, 144.5, 152.9, 154.2, 160.2, 163.9, 177.6 ppm. ESI-MS: m/z 540 $[M + H]^+$. Anal. calcd. for $C_{33}H_{25}N_5O_3$: C, 73.43; H, 4.67; N, 12.98%. Found: C, 74.63; H, 4.45; N, 12.32%.

7-((1-(3-Nitrophenyl)-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-3-methyl-2-phenyl-4*H*-chromen-4-one (7d)

Light brown color solid, yield 78%; m.p. 226–228 °C. IR (KBr, cm^{-1}): 3097 (NH), 1623 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.09 (s, 3H, CH_3), 5.67 (s, 2H, CH_2), 7.19–7.95 (m, 12H, Ar–H), 8.32 (m, 3H, Ar–H), 8.65 (s, 1H, Ar–H), 9.10 (s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO $-$ d_6)δ 11.8, 72.4, 104.3, 109.5, 111.1, 115.3, 116.9, 121.0, 123.4, 124.0, 126.4, 128.0, 128.7, 129.6, 130.5, 131.6, 136.0, 138.9, 144.5, 148.0, 152.9, 154.2, 160.2, 163.9, 177.6 ppm. ESI-MS: m/z571 $[M + H]^+$. Anal. calcd. for $C_{32}H_{22}N_6O_5$: C, 67.36; H, 3.89; N, 14.73. Found: C, 67.19; H, 3.93; N, 14.23.

7-((1-(3-Chlorophenyl)-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-3-methyl-2-phenyl-4*H*-chromen-4-one (7e)

Light brown color solid, yield 75%; m.p. 238–239 °C. IR (KBr, cm^{-1}): 3084 (NH), 1740 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.07 (s, 3H, CH_3), 5.36 (s, 2H, CH_2), 7.19 (s, 1H, Ar–H), 7.53–7.79 (m, 15H, Ar–H), 8.87 (s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO $-$ d_6)δ 13.8, 72.3, 104.3, 109.5, 111.1, 115.3, 116.9, 123.0, 126.4, 128.0, 128.7, 128.9, 129.9, 130.2, 130.4, 131.6, 134.3, 138.8, 144.5, 152.9, 154.2, 160.2, 163.9, 177.6 ppm. ESI-MS: m/z 560 $[M + H]^+$. Anal. calcd. for $C_{32}H_{22}N_5O_3Cl$: C, 68.63; H, 3.96; N, 12.51%. Found: C, 69.83; H, 3.36; N, 12.93%.

7-((1-(2,6-Dimethylphenyl)-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-3-methyl-2-phenyl-4*H*-chromen-4-one (7f)

Light brown color solid, yield 68%; m.p. 235–237 °C. IR (KBr, cm^{-1}): 3086 (NH), 1739 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.32 (m, 9H, CH_3), 5.52 (s, 2H, CH_2), 7.31–7.71 (m, 15H, Ar–H), 8.80 (s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO-d_6) δ 12.1, 15.9, 72.5, 104.3, 109.5, 111.1, 115.1, 116.9, 123.0, 126.1, 128.0, 128.6, 128.7, 129.7, 130.4, 131.6, 133.8, 138.9, 144.5, 152.9, 154.2, 160.2, 163.9, 177.6 ppm. ESI-MS: m/z 554 $[M + H]^+$. Anal. calcd. for $C_{34}H_{27}N_5O_3$: C, 73.76; H, 4.92; N, 12.65%. Found: C, 74.04; H, 4.98; N, 12.40%.

7-((1-(4-Nitrobenzyl)-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-3-methyl-2-phenyl-4*H*-chromen-4-one (7g)

Light brown color solid, yield 72%; m.p. 216–218 °C. IR (KBr, cm^{-1}): 3097 (NH), 1623 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.09 (s, 3H, CH_3), 5.00 (s, 2H, CH_2), 5.87 (s, 2H, CH_2), 7.19 (s, 2H, Ar–H), 7.67 (m, 5H, Ar–H), 7.95 (d, J = 8.92 Hz, 3H, Ar–H), 8.32 (m, 6H, Ar–H), 9.09 (s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO − d_6) δ 12.1, 57.0, 72.2, 104.3, 109.5, 111.1, 115.3, 116.8, 120.9, 121.0, 123.0, 126.4, 128.0, 128.7, 130.0, 130.4, 131.6, 138.9, 142.4, 145.2, 152.9, 154.3, 160.2, 163.9, 177.8 ppm. ESI-MS: m/z 585 $[M + H]^+$. Anal. calcd. for $C_{33}H_{24}N_6O_5$: C, 67.80; H, 4.14; N, 14.38%. Found: C, 67.60; H, 4.40; N, 14.58%.

7-((1-(4-Fluorobenzyl)-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(4,5-dihydro-1*H*-benzo[d]imidazol-2-yl)-3-methyl-2-phenyl-4*H*-chromen-4-one (7h)

Light brown color solid, yield 71%; m.p. 213–215 °C. IR (KBr, cm^{-1}): 3097 (NH), 1623 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.39 (s, 3H, CH_3), 5.28 (s, 2H, CH_2), 5.68 (s, 2H, CH_2), 7.29 (m, 4H, Ar–H), 7.68 (m, 2H, Ar–H), 7.82 (m, 5H, Ar–H), 8.01(m, 3H, Ar–H), 8.29 (s, 2H, Ar–H), 9.15 (s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO-d_6) δ 11.8, 57.0, 72.2, 104.3, 109.5, 111.1, 115.4, 115.6, 116.8, 120.9, 123.0, 126.4, 128.0, 128.7, 130.5, 130.7, 131.6, 138.9, 142.4, 152.9, 154.2, 159.9, 163.9, 177.6 ppm. ESI-MS: m/z 559 $[M+H]^+$. Anal. calcd. for $C_{33}H_{24}N_5O_3F$: C, 71.09; H, 4.34; N, 12.58%. Found: C, 71.45; H, 4.25; N, 12.09%.

7-((1-Cyclopentyl-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-3-methyl-2-phenyl-4*H*-chromen-4-one (7i)

Light brown color solid, yield 65%; m.p. 219–221 °C. IR (KBr, cm^{-1}): 3097 (NH), 1623 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.46–2.39 (m, 11H, Aliphatic-H), 3.71 (m, 1H, CH), 5.08 (s, 2H, CH_2), 6.85 (s, 2H, Ar–H), 7.29–7.95 (m, 6H, Ar–H), 8.29 (m, 3H, Ar–H), 8.68 (s, 1H, Ar–H), 9.12 (s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO−d_6) δ 12.5, 25.7, 32.1, 62.8, 72.4, 104.3, 109.5, 111.1, 115.3, 116.8, 120.9, 123.0, 126.4, 128.0, 128.7, 130.1, 131.6, 138.9, 142.4, 152.9, 154.2, 160.2, 163.9, 178.2 ppm. ESI-MS: m/z 518 $[M+H]^+$. Anal. calcd. for $C_{31}H_{27}N_5O_3$: C, 71.94; H, 5.26; N, 13.53%. Found: C, 72.03; H, 5.86; N, 13.12%.

7-((1-Hexyl-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-3-methyl-2-phenyl-4*H*-chromen-4-one (7j)

Light brown color solid, yield 65%; m.p. 214–216 °C. IR (KBr, cm^{-1}): 3082 (NH), 1738 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.46–2.39 (m, 14H, Aliphatic CH_2), 3.71 (t, J = 1.42 Hz, 2H, CH_2), 5.08 (s, 2H, CH_2), 6.92 (s, 2H, Ar–H), 7.29–7.95 (m, 6H, Ar–H), 8.29 (m, 3H, Ar–H), 8.68 (s, 1H, Ar–H), 9.12 (s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO-d_6) δ 12.5, 14.1, 22.8, 26.9, 28.4, 31.6, 52.5, 72.3, 104.3, 109.5, 111.1, 115.3, 116.8, 120.9, 123.0, 126.4, 128.0, 128.7, 130.4, 131.6, 138.9, 142.4, 152.9, 154.2, 160.2, 163.9, 178.1 ppm. ESI-MS: m/z 535 $[M + H]^+$. Anal. calcd. for $C_{32}H_{31}N_5O_3$: C, 72.03; H, 5.86; N, 13.13%. Found: C, 71.94; H, 5.2 6; N, 13.52%.

7-((1-Phenyl-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-2-phenyl-4*H*-chromen-4-one (7k)

Light brown color solid, yield 79%; m.p. 240–242 °C. IR (KBr, cm^{-1}): 3086 (NH), 1739 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.50 (s, 2H, CH_2), 6.71 (s, 1H, Ar–H), 7.50–7.79 (m, 17H, Ar–H), 8.87 (s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO-d_6) δ 72.4, 104.3, 109.5, 111.1, 115.3, 116.8, 120.9, 123.0, 126.4, 128.0, 128.7, 130.4, 131.6, 138.9, 142.4, 152.9, 154.2, 160.2, 163.9, 177.6 ppm.ESI-MS: m/z 512 $[M+H]^+$. Anal. calcd. for $C_{31}H_{21}N_5O_3$: C, 72.79; H, 4.14; N, 13.69%. Found: C, 72.81; H, 4.49; N, 13.42%.

7-((1-(4-Chlorophenyl)-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-2-phenyl-4*H*-chromen-4-one (7l)

Light brown color solid, yield 76%; m.p. 225–227 °C. IR (KBr, cm^{-1}): 3084 (NH), 1740(C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.55 (s, 2H, CH_2), 6.67 (s, 1H, Ar–H), 7.19–7.82 (m, 15H, Ar–H), 8.24 (s, 1H, Ar–H), 8.97 (s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO-d_6) δ 72.6, 104.5, 111.1, 115.3, 116.8, 116.9, 123.0, 126.4, 128.0, 128.5, 128.7, 130.4, 131.6, 138.9, 144.5, 152.9, 154.2, 163.7, 163.9, 176.9 ppm. ESI-MS: m/z 547 $[M+H]^+$. Anal. calcd. for $C_{31}H_{20}N_5O_3Cl$:

C, 68.20; H, 3.69; N, 12.83%. Found: C, 68.96; H, 3.48; N, 12.61%.

7-((1-m-Toulyl-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-2-phenyl-4*H*-chromen-4-one (7m)

Light brown color solid, yield 78%; m.p. 231–232 °C. IR (KBr, cm^{-1}): 3086(NH), 1739(C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.09 (m, 3H, CH_3), 5.52 (s, 2H, CH_2), 6.67 (s, 1H, Ar–H), 7.31–7.71 (m, 16H, Ar–H), 8.80 (s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO-d_6) δ 24.3, 72.8, 104.5, 109.5, 111.1, 115.3, 116.8, 116.9, 123.0, 124.3, 126.4, 126.9, 128.0, 128.4, 128.7, 129.1, 130.4, 131.6, 138.1, 138.9, 144.5, 152.9, 154.2, 163.7, 163.9, 178.7 ppm. ESI-MS: m/z 526 $[M+H]^+$. Anal. calcd. for $C_{32}H_{23}N_5O_3$: C, 73.13; H, 4.41; N, 13.33%. Found: C, 73.91; H, 3.63; N, 13.48%.

7-((1-(3-Nitrophenyl)-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-2-phenyl-4*H*-chromen-4-one (7n)

Light brown color solid, yield 77%; m.p. 226–228 °C. IR (KBr, cm^{-1}): 3097 (NH), 1623 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.67 (s, 2H, CH_2), 7.19–7.95 (m, 10H, Ar–H), 8.12 (m, 6H, Ar–H), 8.32 (s, 1H, Ar–H), 8.92 (s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO-d_6) δ 71.9, 104.5, 109.5, 111.1, 115.3, 116.8, 116.9, 121.1, 123.0, 124.0, 126.4, 128.0, 128.7, 129.4, 129.7, 130.4, 131.6, 138.9, 144.5, 148.0, 152.9, 154.2, 163.9, 178.2 ppm. ESI-MS: m/z 557 $[M + H]^+$. Anal. calcd. for $C_{31}H_{20}N_6O_5$: C, 66.90; H, 3.62; N, 15.13%. Found: C, 66.32; H, 3.45; N, 15.50%.

7-((1-(3-Chlorophenyl)-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-2-phenyl-4*H*-chromen-4-one (7o)

Light brown color solid, yield 76%; m.p. 223–225 °C. IR (KBr, cm^{-1}): 3084 (NH), 1740(C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.36 (s, 2H, CH_2), 6.94 (s, 2H, Ar–H), 7.49 (m, 6H, Ar–H), 7.92 (m, 5H, Ar–H), 8.32 (m, 3H, Ar–H), 8.87(s, 1H, Ar–H), 8.94 (s, 1H, NH) ppm.^{13}C NMR (100 MHz, DMSO-d_6) δ 72.0, 104.5, 109.5, 111.1, 115.3, 116.8, 116.9, 123.0, 125.4, 126.4, 128.0, 128.7, 128.9, 129.9, 130.4, 131.6, 134.3, 138.9, 144.5, 152.9, 154.2, 163.9, 177.8 ppm.ESI-MS: m/z547 $[M+H]^+$. Anal. calcd. for $C_{31}H_{20}N_5O_3Cl$: C, 68.20; H, 3.69; N, 12.83. Found: C, 68.96; H, 3.48; N, 12.61.

7-((1-(2,6-Dimethylphenyl)-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-2-phenyl-4*H*-chromen-4-one (7p)

Light brown color solid, yield 65%; m.p. 212–213 °C. IR (KBr, cm^{-1}): 3086 (NH), 1739 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.32 (m, 6H, CH_3), 5.52 (s, 2H, CH_2), 6.69 (s, 1H, Ar–H), 7.31–7.71 (m, 15H, Ar–H), 8.80 (s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO-d_6) δ 15.9, 72.7, 104.5, 109.5, 111.1, 115.3, 116.8, 116.9, 123.0, 126.1, 126.4, 128.0, 128.6, 128.7, 129.7, 130.4, 131.6, 133.8, 138.9, 144.5, 152.9, 154.2, 163.7, 163.9, 177.7 ppm. ESI-MS: m/z 540 $[M+H]^+$. Anal. calcd. for $C_{33}H_{25}N_5O_3$: C, 73.46; H, 4.67; N, 12.98%. Found: C, 74.04; H, 4.98; N, 12.40%.

7-((1-(4-Nitrobenzyl)-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-2-phenyl-4*H*-chromen-4-one (7q)

Light brown color solid, yield 70%; m.p. 210–212 °C. IR (KBr, cm^{-1}): 3097 (NH), 1623 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.00 (s, 2H, CH_2), 5.65 (s, 2H, CH_2), 6.71 (s, 1H, Ar–H), 7.19–7.97 (m, 12H, Ar–H), 8.32 (m, 2H, Ar–H), 8.65 (s, 2H, Ar–H), 9.10 (s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO-d_6) δ 57.0, 71.8, 104.6, 109.5, 111.1, 115.3, 116.6, 120.9, 121.0, 123.0, 126.4, 128.0, 128.7, 130.0, 131.6, 138.9, 142.4, 145.4, 152.9, 154.2, 163.7, 163.9, 177.6 ppm. ESI-MS: m/zm/z 571 $[M + H]^+$. Anal. calcd. for $C_{32}H_{22}N_6O_5$: C, 67.36; H, 3.89; N, 14.73%. Found: C, 67.45; H, 3.88; N, 14.53%.

7-((1-(4-Fluorobenzyl)-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-2-phenyl-4*H*-chromen-4-one (7r)

Light brown color solid, yield 69%; m.p. 213–215 °C. IR (KBr, cm^{-1}): 3083 (NH), 1645 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.08 (s, 2H, CH_2), 5.68 (s, 2H, CH_2), 6.79 (s, 1H, Ar–H), 7.29–7.95 (m, 12H, Ar–H), 8.29 (m, 2H, Ar–H), 8.68 (s, 2H, Ar–H), 9.12 (s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO$-d_6$) δ 57.0, 72.0, 104.3, 109.5, 111.1, 115.3, 115.4, 116.8, 120.9, 123.0, 126.4, 128.0, 128.7, 130.4, 130.7, 131.6, 138.9, 142.4, 152.9, 154.3, 159.9, 163.9, 178.6 ppm. ESI-MS: m/z 544 $[M + H]^+$. Anal. calcd. for $C_{32}H_{22}N_5O_3F$: C, 70.71; H, 4.08; N, 12.88%. Found: C, 70.95; H, 4.88; N, 12.38%.

7-((1-Cyclopentyl-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-2-phenyl-4*H*-chromen-4-one (7s)

Light brown color solid, yield 65%; m.p. 213–214 °C. IR (KBr, cm^{-1}): 3097 (NH), 1623 (C=O). ^{1}H NMR(400 MHz, $CDCl_3$) δ 1.46–2.39 (m, 8H, Aliphatic CH_2), 3.71 (m, 1H, CH), 5.08 (s, 2H, CH_2), 6.87 (s, 2H, Ar–H), 7.29–7.95 (m, 6H, Ar–H), 8.29 (m, 3H, Ar–H), 8.68 (s, 2H, Ar–H), 9.12 (s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO-d_6) δ 25.7, 32.1, 62.8, 72.1, 104.5, 109.5, 111.1, 115.3, 116.8, 120.9, 123.0, 126.4, 128.0, 128.7, 130.4, 131.6, 138.9, 142.4, 152.9, 154.2, 163.7, 163.9, 178.5 ppm. ESI-MS: m/z 504 $[M+H]^+$. Anal. calcd. for $C_{30}H_{25}N_5O_3$: C, 71.56; H, 5.00; N, 13.91%. Found: C, 71.01; H, 5.02; N, 14.12%.

7-((1-Hexyl-1*H*-1,2,3-triazol-4-yl)methoxy)-8-(1*H*-benzo[d]imidazol-2-yl)-2-phenyl-4*H*-chromen-4-one (7t)

Light brown color solid, yield 64%; m.p. 209–210 °C. IR (KBr, cm^{-1}): 3082 (NH), 1738 (C=O). ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.46–2.39 (m, 11H, Aliphatic CH_2), 3.75 (t, J = 1.42 Hz, 2H, CH_2), 5.08 (s, 2H, CH_2), 6.87 (s, 2H, Ar–H), 7.29–7.95 (m, 6H, Ar–H), 8.29 (m, 3H, Ar–H), 8.68 (s, 2H, Ar–H), 9.12 (s, 1H, NH) ppm. ^{13}C NMR (100 MHz, DMSO-d_6) δ 14.1, 22.8, 26.9, 28.4, 31.6, 52.5, 72.3, 104.5, 109.5, 111.1, 115.3, 116.5, 120.9, 123.0, 126.4, 128.0, 128.7, 130.4, 131.6, 138.9, 142.4, 152.9, 154.2, 163.7, 163.9, 178.9 ppm. ESI-MS: m/z 535 $[M + H]^+$. Anal. calcd. for $C_{31}H_{29}N_5O_3$: C, 71.66; H, 5.63; N, 13.48%. Found: C, 71.16; H, 5.41; N, 13.98%.

Biological activity

Antimycobacterial activity

The newly synthesized compounds were analyzed for their antimycobacterial effect on

*Mycobacterium bovis*strain (BCG) using a turbidometric assay. 20 mM stock solutions were prepared using 100% (v/v) DMSO. The inoculum for the assay maintained in Middlebrook 7H9 broth supplemented with 0.1% Tween 80 and 0.2% glycerol. At the time of inoculation 10% OADC was added to the media and the culture was incubated in a shaker incubator at 37 °C and 200 rpm. The assay was conducted in a 96-well microtiter plate; a 2 µL aliquot of the 1.5 mM dilution of compound was added to each well in triplicate, to which 98 µL of inoculum dilution was added to make final concentration of the compound to 30 µM. To each plate a set of controls was added to better ascertain the activity of the compounds. These included DMSO which was taken as a growth control, media control (Blank) as well as Rifampicin which was taken as positive control of inhibition. After the completion of the incubation period, the absorbance of the inoculum in wells was measured at 600 nm using a MultiMode Reader (Perkin Elmer). Percentage inhibition was determined against DMSO.

Antiproliferative activity

The cytotoxicity of the compound was tested by carrying out a Sulforhodamine B (SRB) Assay. The MCF-7 cell line was seeded in to a clear flat bottom 96-well plate (5000 cells/100 µL) in a medium supplemented with 10% serum and allowed to incubate for 18–20 h. To ensure the proper adherence of the cells to the surface bottom of the wells, incubator was continuously supplied with 5% CO_2. Variable dilutions of the compounds were prepared, and 2 µL aliquot was added to the each well to make final concentration of compound 10 µM. Each compound was tested in triplicate. DMSO and doxorubicin (as standard control anti cancer drug) were used as vehicle and positive controls, respectively. After 48 h, the cells were fixed with 10% trichloroacetic acid solution and incubated the plates at 4 °C for 1 h after which the plate was rinsed carefully with MQ water and air-dried followed by the addition of 0.057% SRB solution and kept for approx. 30 min before it was washed off with 1% acetic acid. The plates were then air-dried, and the absorbance was measured at 510 nm using a PerkinElmer Multimode Reader. To measure the absorbance 100 µL of 10 mM unbuffered tris base solution was added to each well to dissolve the SRB. The measure of absorbance is proportional to cell growth; the absorption data are used to obtain IC_{50} values.

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