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Synthesis and Cytotoxicity Activity of Novel Benzotrizolyl Methyl[1,2,4-triazolo][3,4-*b*]thiadiazinyl Substituted Chromene 2-ones

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The biological importance of the coumarins and triazolothiadiazines, prompted as to synthesize a series of novel benzotrizolyl methyl[1,2,4-triazolo][3,4-*b*]thiadiazinyl substituted chromene 2-ones applying simple experimental conditions with good yields. All the synthesized molecules were well characterized by physical, analytical and spectroscopic techniques. The novel synthesized molecules **5(a-k)** were screened for the anticancer activity and some of the molecules exhibited moderate to good activity *in vitro*.

Keywords: Substituted 3-(2-bromoacetyl)coumarins, Thiocarbohydrazide, Anticancer activity, Triazolothiadiazines.

INTRODUCTION

Synthesis of nitrogen and sulphur containing heterocyclic ring systems has been attracting the researchers over the past decade due to their vast applications. Azoles especially [1,2,4]-triazoles are one of the important and interesting therapeutically active heterocyclic compounds. Triazoles and their fused heterocyclic analogues have been extensively studied due to their diverse application in the fields of medicine, agriculture and industry. In the market many prominent drug molecules contain triazole as core functional group (*e.g.*, letrozole, voriconazole, itraconazole, flucanazole and posaconazole). The N-bridged [1,2,4]-triazoles exhibit wide range of pharmacological activities such as antimicrobial [1,2], antibacterial [3], antifungal [4], antiviral [5], antihelmenthic [6], anti-inflammatory, analgesic [7], anticancer [8,9], antitubercular [10] and anticonvulsant [11] properties.

In addition, benzopyaran-2-ones are important category of compounds widely distributed in nature. In the recent years the chemistry of coumarins has attracted interest in the field of natural products, dyes, agrochemicals, material sciences and medicine. A large number of coumarins and their derivatives have shown remarkable biological activities such as antibacterial [12,13], antifungal [14], anticoagulants [15], anti-inflammatory [16], antitumor [17,18], antioxidant [19], free radical scavenger [20] and antiviral [21,22], *etc*.

Hybridization of two biologically active pharmacophores may have significant therapeutic profile when compared to the individual pharmacophores. The use of computational studies to elicit the molecular level interactions in order to explain the biological properties of the newly synthesized molecules. Prompted by the above observations, we have synthesized series of novel benzotrizolyl methyl[1,2,4-triazolo][3,4-*b*]thiadiazinyl substituted chromene 2-ones and screened for *in silico* and their *in vitro* anticancer activity.

EXPERIMENTAL

Benzotriazole and other solvents used in the present study were procured from commercial sources and used without any further purification. The melting points of the final products were determined on Stuart melting Point apparatus (SMP-30, Stuart, Staffordshire UK) in open capillaries. The progress of the reaction was monitored by thin layer chromatography (TLC) by using commercially available silica coated Merck silica gel 60 F_{254} aluminum sheets and spots were visualized under UV-visible light. ^{1}H NMR and ^{13}C NMR were recorded on BRUKER AVANCE-III 400 MHz, 100 MHz by using DMSO-d_6 as a solvent and tetramethylsilane as internal standard. Mass spectra were recorded on WATERS-MICROMASS Quattro micro API mass spectrometer. The IR spectral data of all the compounds were recorded on Bruker FT-IR spectrometer (Model: TENSOR-27). Elemental analysis was performed on Q-Toff-2010 elemental analyzer.

General procedure for the synthesis of 5-[(1H-benzo-[d][1,2,3]triazol-1-yl)methyl]-4-amino-4H-1,2,4-triazole-3-thiol: An equimolar mixture of benzotriazole-1-yl-acetic acid (**1**) and thiocarbohydrazide (**2**) were taken in the round bottom flask. The reactants were thoroughly mixed and fused the

contents by raising the temperature to 120-130 °C. The molten reaction mixture was kept at 120-130 °C for about 15 min. Finally the resulting reaction mass temperature was reduced to room temperature and treated with 5 % aqueous sodium bicarbonate solution to remove the unreacted carboxylic acid. The separated solid was filtered, dried and recrystallized from appropriate solvent.

Yield: (60 %); m.p.: 180-182 °C. IR (KBr, ν_{max}, cm^{-1}): 3287 (-NH_2), 1377 (-C=S); ^{1}H NMR (400 MHz, DMSO-d_6, ppm): δ 8.07-8.09 (d, *J* = 8.3 Hz, 1H, Ar-H), 7.91-7.93 (d, *J* = 8.3 Hz, 1H, Ar-H), 7.51-7.54 (t, *J* = 7.4 Hz, 1H, Ar-H), 7.44-7.42 (t, *J* = 7.2 Hz, 1H, Ar-H), 6.25 (s, 2H, -CH_2), 3.05 (broad s, 3H, -NH_2, -SH). ^{13}C NMR (100 MHz, DMSO-d_6, ppm) δ 167.20, 160.10, 145.21, 132.91, 127.20, 127.43, 118.11, 118.30, 41.40.MS (ES *m/z*) for $C_9H_9N_7S$: 248.27 $[M + H]^+$.

General procedure for the synthesis of novel benzotrizolyl methyl [1,2,4-triazolo][3,4-*b*]thiadiazinyl substituted chromene 2-ones: A mixture of 5-[(1H-benzo[d][1,2,3]triazol-1-yl)methyl]-4-amino-4H-1,2,4-triazole-3-thiol (1 mmol) and substituted 3-(2-bromoacetyl)coumarins (1 mmol) were taken in the round bottom flask having ethanol (5 mL) as a solvent medium. The reaction mixture was stirred for about 10 min at room temperature and finally the temperature was raised to 65-70 °C. The progress of the reaction was monitored through thin layer chromatography, finally the temperature was decreased to room temperature and the isolated product was filtered washed with methanol and air dried. The products were re crystallized from suitable solvent.

3-[3-{(1H-Benzo[d][1,2,3]triazol-1-yl)methyl}-7H-[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazin-6-yl]-2H-chromen-2-one (5a): IR (KBr, ν_{max}, cm^{-1}) 1718 (C=O); 1605 (C=N); ^{1}H NMR (400 MHz, DMSO-d_6, δ ppm): 8.27 (s, 1H, coumarin 4th proton), 8.09 (d, *J* = 8.4 Hz, 1H, Ar-H), 7.93 (d, *J* = 8.4 Hz, 1H, Ar-H), 7.75-7.81 (m, 2H, Ar-H), 7.47-7.54 (m, 3H, Ar-H), 7.39-7.43 (t, 1H, Ar-H), 6.35 (s, 2H, -CH_2), 4.28 (s, 2H, -CH_2): ^{13}C NMR (100 MHz, DMSO-d_6, δ ppm): 158.47, 153.93, 153.47, 148.41, 145.06, 142.14, 133.99, 133.23, 129.80, 127.49, 124.12, 122.27, 119.12, 118.14, 116.35, 111.01, 41.47, 24.14. MS (ES *m/z*) for $C_{20}H_{13}N_7O_2S$: 416.18 $[M + H]^+$.

3-[3-{(1H-Benzo[d][1,2,3]triazol-1-yl)methyl}-7H-[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazin-6-yl]-5,8-di-*tert*-butyl-2H-chromen-2-one (5b): IR (KBr, ν_{max}, cm^{-1}): 1723 (C=O); 1619 (C=N); ^{1}H NMR (400 MHz, DMSO-d_6, δ ppm): 8.25 (s, 1H, coumarin 4th proton), δ 8.06-8.10 (m, 1H, Ar-H), 7.93-7.96 (m, 1H, Ar-H), 7.68 (d, *J* =1.6 Hz, 1H, Ar-H), 7.61 (d, *J* = 2.0 Hz 1H, Ar-H), 7.51-7.55 (m, 1H, Ar-H), 7.41-7.47 (m, 1H, Ar-H), 6.35 (s, 2H, CH_2), 4.31 (s, 2H, CH_2), 1.48 (s, 9H, *tert*-butyl), 1.37 (s, 9H, *tert*-butyl): ^{13}C NMR (100 MHz DMSO-d_6 ppm) δ 158.20, 153.38, 150.71, 148.39, 146.90, 145.96, 145.72, 145.05, 143.86, 142.12, 136.25, 133.25, 128.61, 127.48, 126.75, 124.45, 124.31, 124.06, 120.91, 119.11, 118.21, 117.91, 111.05, 41.45, 34.65, 31.00, 29.50, MS (ES *m/z*) for $C_{28}H_{29}N_7O_2S$: 527.8 $[M + H]^+$.

3-[3-{(1H-Benzo[d][1,2,3]triazol-1-yl)methyl}-7H-[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazin-6-yl]-6-chloro-2H-chromen-2-one (5c): IR (KBr, ν_{max}, cm^{-1}) 1723 (C=O), 1619 (C=N), 1205 (C-Cl); ^{1}H NMR (400 MHz, DMSO-d_6, δ ppm): 8.31 (s, 1H, coumarin 4th proton), 8.09 (d, *J* = 8.4 Hz, 1H, Ar-H), 7.91-7.93 (m, 2H, Ar-H), 7.79-7.82 (dd, *J* = 2.4 Hz, 8.8 Hz, 1H, Ar-H), 7.58 (d, *J* = 8.8 Hz, 1H, Ar-H), 7.51-7.55 (t, *J* = 7.2 Hz, 1H, Ar-H), 7.39-7.43 (t, *J* = 7.2 Hz, 1H, Ar-H), 6.33 (s, 2H, -CH_2), 4.28 (s, 2H, -CH_2); ^{13}C NMR (100 MHz DMSO-d_6, δ ppm): 158.10, 153.35, 152.59, 148.43, 145.09, 143.29, 142.15, 133.39, 133.21, 128.83, 128.61, 127.53, 124.10, 123.54, 119.55, 119.15, 118.42, 111.04, 41.43, 24.18 MS (ES *m/z*) for $C_{20}H_{12}N_7O_2SCl$: 450.15 $[M + H]^+$, 452.15 $[M+2]^+$.

3-[3-{(1H-Benzo[d][1,2,3]triazol-1-yl)methyl}-7H-[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazin-6-yl]-6-bromo-2H-chromen-2-one (5d): IR (KBr, ν_{max}, cm^{-1}): 1726 (C=O); 1602 (C=N); 1205 (C-Br) ^{1}H NMR (400 MHz, DMSO-d_6, δ ppm): 8.30 (s, 1H, coumarin 4th proton), 7.39-7.45 (m, 1H, Ar-H), 7.50-7.54 (m, 2H, Ar-H), 7.91-793 (m, 2H, Ar-H), 8.05-8.10 (m, 2H, Ar-H), 6.33 (s, 2H), 4.28 (s, 2H): ^{13}C NMR (100 MHz DMSO-d_6, δ ppm): 158.04, 153.31, 152.99, 148.42, 145.55, 145.07, 143.21, 142.12, 136.52, 136.14; 133.20, 132.47, 131.60, 127.52, 124.07, 123.46, 120.02, 119.14, 118.64, 118.35, 116.61, 116.29, 111.03, 41.41, 24.16; MS (ES *m/z*) for $C_{20}H_{12}N_7O_2SBr$: 494.07 $[M + H]^+$, 496.01$[M+2]^+$.

3-[3-{(1H-Benzo[d][1,2,3]triazol-1-yl)methyl}-7H-[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazin-6-yl]-8-methoxy-2H-chromen-2-one (5e): IR (KBr, ν_{max}, cm^{-1}): 1721 (C=O); 1609 (C=N); 1276 (C-O); ^{1}H NMR (400 MHz, DMSO-d_6, δ ppm): 8.25 (s, 1H, coumarin 4th proton), 8.08 (d, *J* = 8.4 Hz, 1H, Ar-H), 7.92 (d, *J* = 8 Hz, 1H, Ar-H), 7.50-7.53 (t, *J* = 7.2 Hz, 1H, Ar-H), 7.39-7.47 (m, 3H, Ar-H), 7.33-7.35 (m, 1H, Ar-H), 6.35 (s, 2H, CH_2), 4.28 (s, 2H, CH_2), 3.95 (s, 3H, OCH_3); ^{13}C NMR (100 MHz DMSO-d_6, δ ppm): 158.19, 153.40, 148.41, 146.39, 145.05, 144.88, 143.25, 142.13, 133.21, 127.48, 125.19, 124.12, 122.37, 120.73, 119.10, 118.69, 110.99, 56.21, 41.46, 24.10; MS (ES *m/z*) for $C_{21}H_{15}N_7O_3S$: 446.15 $[M + H]^+$.

3-[3-{(1H-Benzo[d][1,2,3]triazol-1-yl)methyl}-7H-[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazin-6-yl]-6-bromo-8-methoxy-2H-chromen-2-one (5f): IR (KBr, ν_{max}, cm^{-1}): 1729 (C=O); 1603 (C=N); 1272 (C-O); ^{1}H NMR (400 MHz, DMSO-d_6, δ ppm): 8.27 (s, 1H, coumarin 4th proton), 8.08 (d, *J* = 8.4 Hz, 1H, Ar-H), 7.92 (d, *J* = 8.4 Hz, 1H, Ar-H), 7.58-7.61 (m, 2H, Ar-H), 7.50-7.54 (t, *J* = 7.6 Hz, 1H, Ar-H), 7.39-7.42 (t, *J* = 7.6 Hz, 1H, Ar-H), 6.33 (s, 2H, -CH_2), δ 4.28 (s, 2H, -CH_2), 3.98 (s, 3H, -OCH_3); ^{13}C NMR (100 MHz DMSO-d_6, δ ppm): 157.74, 153.24, 148.42, 147.32, 145.07, 143.53, 142.67, 142.11, 133.20, 127.51, 124.07, 123.58, 122.57, 119.95, 119.13, 118.57, 116.52, 111.03, 56.80, 41.41, 24.12: MS (ES *m/z*) for $C_{21}H_{14}N_7O_3SBr$: 524.01 $[M + H]^+$; 526.04 $[M+2]^+$.

3-[3-{(1H-Benzo[d][1,2,3]triazol-1-yl)methyl}-7H-[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazin-6-yl]-6-(diethylamino)-2H-chromen-2-one (5g): IR (KBr, ν_{max}, cm^{-1}): 1713 (C=O); 1616 (C=N); ^{1}H NMR (400 MHz, DMSO-d_6, δ ppm): 8.07-8.10 (m, 2H, Ar-H), 7.93 (d, *J* = 8.4 Hz, 1H, Ar-H), 7.51-7.55 (t, *J* = 7.2 Hz, 1H, Ar-H), 7.49 (d, *J* = 8.8 Hz, 1H, Ar-H), 7.41-7.44 (t, *J* = 7.6 Hz, 1H, Ar-H), 6.84-6.86 (d, *J* = 8.8 Hz 1H, Ar-H), 6.63 (s, 1H, Ar-H), 6.34 (s, 2H, -CH_2), 4.26 (s, 2H, -CH_2), 3.48-3.53 (q, *J* = 6.8 Hz, 4H, -CH_2), 1.14-1.18 (t, *J* = 6.8 Hz, 6H, -CH_3); ^{13}C NMR (100 MHz, DMSO-d_6, δ ppm): 159.49, 157.38, 153.99, 152.44, 148.10, 145.04, 144.99, 142.15, 133.22, 131.24, 127.49, 124.12, 119.11, 111.93,

110.97, 110.21, 107.40, 96.08, 44.34, 41.54, 23.90, 12.29; MS (ES *m/z*) for $C_{24}H_{22}N_8O_2S$: 487.18 $[M + H]^+$.

3-[3-{(1H-Benzo[d][1,2,3]triazol-1-yl)methyl}-7H-[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazin-6-yl]-6,8-dinitro-2H-chromen-2-one (5h): IR (KBr, ν_{max}, cm^{-1}): 1745 (C=O); 1619 (C=N); ^{1}H NMR (400 MHz, DMSO-d_6, δ ppm): 8.77 (s, 1H, coumarin 4th proton), 8.51-8.55 (m, 1H, Ar-H), 8.09 (d, *J* = 8.4 Hz, 1H, Ar-H), 7.94 (d, *J* = 8.4 Hz, 1H, Ar-H), 7.76 (d, *J* = 9.2 Hz, 1H, Ar-H), 7.51-7.55 (m, 1H, Ar-H), 7.40-7.43 (m, 1H, Ar-H), 6.34 (s, 2H, CH_2), 4.31 (s, 2H, CH_2); ^{13}C NMR (100 MHz DMSO-d_6, δ ppm): 157.65, 157.37, 152.99, 148.47, 145.08, 143.88, 143.41, 142.07, 133.22, 128.07, 127.56, 125.46, 124.24, 124.11, 119.15, 118.54, 117.93, 111.07, 41.37, 24.09; MS (ES *m/z*) for $C_{20}H_{11}N_9O_6S$: 506.48 $[M + H]^+$.

3-[3-{(1H-Benzo[d][1,2,3]triazol-1-yl)methyl}-7H-[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazin-6-yl]-6,8-dibromo-2H-chromen-2-one (5i): IR (KBr, ν_{max}, cm^{-1}): 1722 (C=O); 1611 (C=N); ^{1}H NMR (400 MHz, DMSO-d_6, δ ppm): 8.31 (s, 2H, Ar-H), 8.07-8.09 (m, 2H, Ar-H), 7.92 (d, *J* = 7.6, 1H; Ar-H), 7.53 (m, 1H, Ar-H), 7.41 (m, 1H, Ar-H), 6.32 (s, 2H, CH_2), 4.29 (s, 2H, CH_2); ^{13}C NMR (100 MHz, DMSO-d_6, δ ppm): 164.52, 157.47, 152.96, 149.89, 148.43, 145.09, 142.91, 142.09, 138.02, 133.20, 131.28, 127.56, 127.36, 124.08, 123.90, 121.05, 119.14, 116.66, 111.04, 110.76, 110.32, 48.40, 41.40, 24.11; MS (ES *m/z*): 574 $[M + H]^+$.

2-[3-{(1H-Benzo[d][1,2,3]triazol-1-yl)methyl}-7H-[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazin-6-yl]-3H-benzo[f]-chromen-3-one (5j): IR (KBr, ν_{max}, cm^{-1}): 1725 (C=O); 1624 (C=N); ^{1}H NMR (400 MHz, DMSO-d_6, δ ppm): 9.17 (s, 1H, coumarin 4th proton), 8.59 (d, *J* = 8.4 Hz, 1H, Ar-H), 8.35 (d, *J* = 9.2 Hz, 1H, Ar-H), 8.13 (d, *J* = 8.4 Hz, 1H, Ar-H), 8.08 (d, *J* = 8 Hz, 1H, Ar-H), 7.95 (d, *J* = 8.4 Hz, 1H, Ar-H), 7.86-7.89 (t, *J* = 7.6 Hz, 1H, Ar-H), 7.67-7.73 (m, 2H, Ar-H), 7.43-7.46 (t, *J* = 8.0 Hz, 1H, Ar-H), 7.33-7.37 (t, *J* = 7.6 Hz, 1H, Ar-H), 6.43 (s, 2H, CH_2), 4.36 (s, 2H, CH_2):^{13}C NMR (100 MHz DMSOD-d_6, δ ppm): 158.53, 154.57, 153.23, 148.45, 145.12, 142.23, 140.88, 135.68, 133.15, 130.00, 129.12, 128.99, 127.44, 126.64, 124.13, 122.37, 120.96, 119.12, 116.51, 112.66, 110.94, 41.59, 23.99; MS (ES *m/z*) for $C_{24}H_{15}N_7O_2S$: 466.18 $[M + H]^+$.

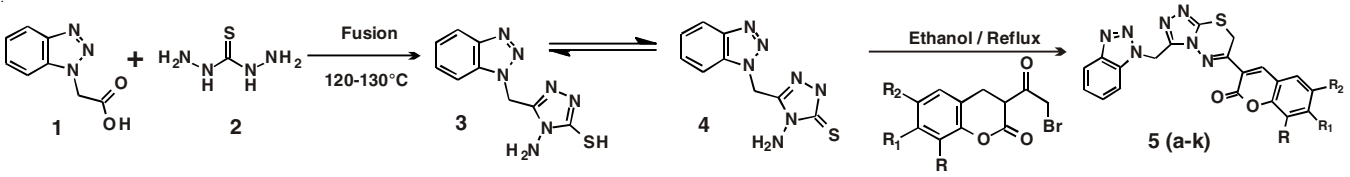
3-[3-{(1H-Benzo[d][1,2,3]triazol-1-yl)methyl}-7H-[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazin-6-yl]-7-hydroxy-2H-chromen-2-one (5k): IR (KBr, ν_{max}, cm^{-1}): 3401 (-OH); 1707 (C=O); 1614 (C=N); ^{1}H NMR (400 MHz, DMSO-d_6, ppm): 11.12 (broad s, 1H, OH), 8.18 (s, 1H, coumarin 4th proton), 8.09 (d, *J* = 8.4 Hz, 1H, Ar-H), 7.92 (d, *J* = 8.4, 1H, Ar-H), 7.63 (d, *J* = 8.4 Hz, 1H, Ar-H), 7.50-7.54 (t, *J* = 8 Hz, 1H, Ar-H), 7.39-7.43 (t, *J* = 7.6 Hz, 1H, Ar-H), 6.91-6.93 (dd, *J* = 1.6 Hz, 8 Hz, 1H, Ar-H), 6.82 (d, *J* = 2 Hz, 1H, Ar-H), 6.33 (s, 2H, CH_2), 4.25 (s, 2H, CH_2); ^{13}C NMR (100 MHz DMSO-d_6, δ ppm): 163.78, 159.43, 156.84, 154.23, 148.81, 145.75, 145.51, 142.71, 133.71, 132.10, 128.06, 124.73, 119.61, 117.37, 114.78, 111.44, 102.48, 41.98, 24.52; MS (ES *m/z*) for $C_{20}H_{13}N_7O_3S$: 432.16 $[M + H]^+$.

Cell proliferation assay by MTT method: Briefly, cells were plated at a density of 1000 to 10,000 cells/well in 96 well plates and incubated overnight at 37 °C in 5 % CO_2 incubator. The following day, the cells were treated with the test compounds and proto drug Erlotinib. Cells were allowed to incubate for different time points up to 96 h. MTT reagent was then added to the treated cells for an additional 4 h and then processed according to the standard protocol [23].

RESULTS AND DISCUSSION

The synthetic route for the preparation of novel benzotrizolyl methyl[1,2,4-triazolo][3,4-b]thiadiazinyl substituted chromene 2-ones is depicted in **Scheme-I**. The precursors like benzotriazole-1-yl-acetic acid, thiocarbohydrazide and substituted 3-(2-bromoacetyl) coumarins needed for the synthesis of the target molecules were synthesized according to the reported literature procedures [24-26]. The expected target molecules were synthesized in two steps. In the first step, benzotriazole-1-yl-acetic acid is fused with thiocarbohydrazide to give the intermediate 5-((1H-benzo[d][1,2,3]triazol-1-yl)-methyl)-4-amino-4H-1,2,4-triazole-3-thiol (**3**) with good yield. The second step is condensation of the intermediate with substituted 3-(2-bromoacetyl) coumarins to give the target molecules (**5a-j**) in good yields (Table-1). The structures of the synthesized molecules were confirmed on the basis of analytical data like IR, NMR, Mass and elemental analysis. The IR spectrum of the compound **5a** ((1H-benzo[d][1,2,3]-triazol-1-yl)methyl)-4-amino-4H-1,2,4-triazole-3-thiol (**3**) has shown characteristic stretching frequencies at 3287, 1372 cm^{-1} indicating the presence of amino and thiol functional groups. The IR spectra of the final compounds showed absorption bands at 1723-1713 and 1624-1602 cm^{-1}, respectively. Absence of the absorption stretching frequencies of corresponding NH_2, SH and C=O of the starting materials clearly indicates the formation of the final product. ^{1}H NMR of the compounds (**5a-k**) exhibited characteristic peaks at δ 4.28 ppm due to methylene protons, peaks at δ 8.27 due to C_4-proton of coumarin. All the other remaining aromatic protons were appeared at the expected chemical shift values. ^{13}C NMR of the final products showed characteristic peaks at δ 24.14 which is due to methylene carbon and peaks at δ 158.47 due to 4th carbon of coumarin.

Biological activity: To evaluate the biological activity, all the synthesized compounds were screened for their *in vitro* anticancer activity against human lung cancer (HCC-827) cell line. The anticancer activity of each individual compound was compared with Erlotinib as FDA approval drug standard and the results were summarized in Table-2. The compound **5j** and compound **5e** showed promising good anticancer activity.



Scheme-I

TABLE-1
SYNTHESIS OF NOVEL SUBSTITUTED COUMARINYL TRIAZOLOTHIADIAZENES

Product	R	R_1	R_2	m.p. (°C)	Yield (%)
5a	H	H	H	230-232	79
5b	*t*-Butyl	H	*t*-Butyl	198-200	69
5c	H	H	Cl	252-254	82
5d	H	H	Br	224-226	86
5e	OCH_3	H	H	215-217	80
5f	OCH_3	H	Br	169-171	89
5g	H	H	$N(C_2H_5)_2$	201-203	72
5h	NO_2	H	NO_2	240-242	77
5i	Br	H	Br	235-237	91
5j		5,6-Benzo		212-214	86
5k	H	OH	H	260-262	80

TABLE-2
ANTI-CANCER ACTIVITY OF NOVEL BENZOTRIZOLYL METHYL[1,2,4-TRIAZOLO][3,4-*b*]THIADIAZINYL SUBSTITUTED CHROMENE 2-ONES (**5a-k**)

Type of cell line	Standard/reference drugs	
	Standard/Test compound	IC_{50} values (μM)
Lung cancer (HCC-827)	**5a**	> 10
	5b	> 10
	5c	> 10
	5d	> 10
	5e	8.693
	5f	> 10
	5g	> 10
	5h	> 10
	5i	> 10
	5j	0.821
	5k	8.491
	Erlotinib	0.007 nM

From this anticancer activity data it is evident that the presence of electron rich species like naphthyl and methoxy group on coumarin ring are necessary for the cytotoxic activity.

Conclusion

In conclusion, we synthesized novel, efficient benzotrizolyl methyl [1,2,4-triazolo][3,4-*b*]thiadiazinyl substituted chromene 2-ones and screened for anticancer activity by molecular docking studies. Among all the anticancer screened compounds, compounds **5j** and **5k** exhibited significant anticancer activity.

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