SYNTHESIS AND ANTIBACTERIAL EVALUATION OF NOVEL 3,6-DISUBSTITUTED COUMARIN DERIVATIVES

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EXPERIMENTAL SECTION

Materials and instrumentation.

All the reagents and solvents were purchased from Aldrich/Merck and used without further purifications. Melting points were determined in open capillaries using Stuart SMP30 apparatus and are uncorrected. The progress of the reactions as well as purity of compounds was monitored by thin layer chromatography with F₂₅₄ silica-gel precoated sheets using hexane/ethyl acetate 8/2 as eluent; UV light and iodine vapours were used for detection. IR spectra were recorded on a Perkin-Elmer 100S spectrometer utilizing KBr pellets. ¹H NMR and ¹³C NMR spectra were obtained at 400 MHz and 100 MHz respectively on Bruker using DMSO-*d*₆ as solvent and TMS as internal standard. Elemental analyses were performed on a Carlo-Erba model EA1108 analytical unit. Mass spectra were recorded on a Jeol JMSD-300 spectrometer.

General procedure for the synthesis of ethyl-2-(3-acetyl-2-oxo-2H-chromen-6-yl)-4-methylthiazole-5-carboxylate (3)

A mixture of ethyl 2-(3-formyl-4-hydroxyphenyl)-4-methylthiazole-5-carboxylate (**1**) (0.1 mol) and ethylacetoacetate (**2**) (0.12 mol) were dissolved in ethanol and cooled at 0-5 °C. Piperidine (1 mL) was added drop wise to this mixture while stirring. The reaction mixture left overnight, resulting in the formation of yellow colored solid. Purification by recrystallization from ethanol to afford analytically pure product.

General procedure for the synthesis of 3,6-disubstituted coumarin derivatives (6a-l)

A mixture of ethyl-2-(3-acetyl-2-oxo-2H-chromen-6-yl)-4-methylthiazole-5-carboxylate (**3**, 1 mmol), thiosemicarbazide (1 mmol) and various phenacyl bromides (**5a-g**) / 3-(2-bromoacetyl)-2H-chromen-2-ones (**5h-k**) / 2-(2-bromoacetyl)-3H-benzo[*f*]chromen-3-one (**5l**) (1 mmol) were dissolved in 10 mL of absolute ethanol in the presence of catalytic amount of acetic acid and heated at refluxing temperature for 2-4 hrs. Monitored the reaction by TLC and poured contents in an ice cold water, the solid obtained was filtered, dried and recrystallised in ethanol.

Spectral data of synthesized compounds (3, 6a-6l)

Ethyl 2-(3-acetyl-2-oxo-2H-chromen-6-yl)-4-methylthiazole-5-carboxylate (3)

Yellow solid; mp. 194-196 °C; ¹H NMR (400MHz, DMSO-*d*₆): 1.32 (t, 3H, *J* = 7.2 Hz), 2.59 (s, 3H), 2.71 (s, 3H), 4.29-4.34 (m, 2H), 7.58 (d, 1H, *J* = 8.8 Hz), 8.29-8.32 (m, 1H), 8.63 (s, 1H), 8.75 (s, 1H); ¹³C NMR (100 MHz, DMSO-*d*₆): δ 14.0, 17.1, 29.9, 61.2, 117.2, 118.7, 125.4, 128.7, 131.8, 146.3, 149.4, 156.2, 156.7, 157.9, 160.4, 161.2, 195.0;

Ethyl-4-methyl-2-(2-oxo-3-(1-(2-(4-phenylthiazol-2-yl)hydrazono)ethyl)-2H-chromen-6-yl)thiazole-5-carboxylate (6a)

Pale yellow solid; mp. 224-226 °C; IR (KBr, cm⁻¹) $\nu_{\max}$: 3404 (NH), 1741(C=O), 1718 (C=O), 1611 (C=N), 1565 (C=C); ¹H NMR (400MHz, DMSO-*d*₆): δ 1.31 (t, 3H, *J* = 7.2 Hz), 2.29 (s, 3H), 2.71 (s, 3H), 4.28-4.33 (m, 2H), 7.29-7.44 (m, 2H), 7.55 (d, 1H, *J* = 8.8 Hz), 7.89 (d, 2H, *J* = 7.2 Hz), 8.19-8.22 (m, 1H), 8.30 (s, 1H), 8.56-8.62 (m, 2H), 8.74 (s, 1H), 11.45 (s, 1H); ¹³C NMR (100 MHz, DMSO-*d*₆): δ 14.0, 16.0, 17.1, 61.2, 104.4, 116.9, 119.5, 121.5, 125.5, 127.2, 127.4, 128.5, 129.7, 134.5, 139.9, 144.4, 154.7, 158.5, 160.2, 161.2, 167.3, 169.2; MS (ESI) *m/z*: 531 [M+1]⁺; Anal. Calcd. for C₂₇H₂₂N₄O₄S₂: C, 61.12; H, 4.18; N, 10.56. Found: C, 61.24; H, 4.06; N, 10.61.

Ethyl-2-(3-(1-(2-(4-(4-chlorophenyl)thiazol-2-yl)hydrazono)ethyl)-2-oxo-2H-chromen-6-yl)-4-methylthiazole-5-carboxylate (6b)

Yellow solid; mp. 188-190 °C; IR (KBr, cm⁻¹) $\nu_{\max}$: 3404 (NH), 1742 (C=O), 1718 (C=O), 1611 (C=N) 1565 (C=C), 759 (C-Cl); ¹H NMR (400MHz, DMSO-*d*₆): δ 1.31 (t, 3H, *J* = 7.2 Hz), 2.59 (s, 3H), 2.70 (s, 3H), 4.28-4.34 (m, 2H), 7.45-7.59 (m, 2H), 7.90 (d, 1H, *J* = 8.4 Hz), 8.20 (d, 1H, 8.8 Hz), 8.29 (t, 2H, *J* = 7.2 Hz), 8.55 (s, 1H), 8.62 (s, 1H), 8.74 (s, 1H), 11.47 (s, 1H); ¹³C NMR (100 MHz, DMSO-*d*₆): δ 14.0, 17.1, 29.9, 61.2, 117.2, 118.7, 121.5, 121.7, 125.3, 127.2, 128.5, 128.7, 131.8, 146.3, 156.0, 157.8, 160.2, 161.2, 194.8; MS (ESI) *m/z*: 567 [M+2]⁺; Anal. Calcd. for C₂₇H₂₁ClN₄O₄S₂: C, 57.39; H, 3.75; N, 9.92. Found: C, 57.47; H, 3.62; N, 9.83.

Ethyl-2-(3-(1-(2-(4-(4-bromophenyl)thiazol-2-yl)hydrazono)ethyl)-2-oxo-2H-chromen-6-yl)-4-methylthiazole-5-carboxylate (6c)

Yellow solid; mp. 239-241 °C; IR (KBr, cm⁻¹) $\nu_{\max}$: 3415 (NH), 1718 (C=O), 1610 (C=N), 1569 (C=C), 718 (C-Br); ¹H NMR (400MHz, DMSO-*d*₆): δ 1.31 (t, 3H, *J* = 7.2 Hz), 2.28 (s, 3H), 2.71 (s, 3H), 4.28-4.33 (m, 2H), 7.46 (s, 1H), 7.54 (d, 1H, *J* = 8.4 Hz), 7.61 (d, 2H, *J* = 8.8 Hz), 7.83 (d, 2H, *J* = 8.4 Hz), 8.19-8.22 (m, 1H), 8.29 (s, 1H), 8.56 (s, 1H), 11.44 (s, 1H); ¹³C NMR (100

MHz, DMSO-*d*₆): δ 14.0, 16.0, 17.1, 61.2, 116.9, 121.3, 126.6, 127.3, 127.5, 128.5, 129.7, 131.2, 131.4, 131.8, 132.6, 140.2, 154.7, 158.4, 160.2, 161.2, 167.3, 168.2; MS (ESI) *m/z*: 610 [M+1]⁺; Anal. Calcd. for C₂₇H₂₁BrN₄O₄S₂: C, 53.20; H, 3.47; N, 9.19. Found: C, 53.41; H, 3.33; N, 9.24.

Ethyl-4-methyl-2-(3-(1-(2-(4-(4-nitrophenyl)thiazol-2-yl)hydrazono)ethyl)-2-oxo-2H-chromen-6-yl)thiazole-5-carboxylate (6d)

Yellow solid; mp. 196-198 °C; IR (KBr, cm⁻¹) $\nu_{\max}$: 3416 (NH), 1735 (C=O), 1598 (C=N), 1340 (NO₂); ¹H NMR (400MHz, DMSO-*d*₆): δ 1.32 (t, 3H, *J* = 7.2 Hz), 2.30 (s, 3H), 2.72 (s, 3H), 4.29-4.34 m, 2H), 7.56-7.61 (m, 1H), 7.79 (s, 1H), 8.15 (d, 1H, *J* = 8.8 Hz), 8.22-8.33 (m, 3H), 8.59 (s, 1H), 8.66 (s, 1H), 8.76 (s, 1H), 11.60 (s, 1H); ¹³C NMR (100 MHz, DMSO-*d*₆): δ 14.1, 17.0, 61.2, 117.2, 118.1, 121.4, 123.0, 125.3, 127.2, 128.2, 129.5, 132.2, 146.1, 156.0, 158.8, 160.2, 162.3; MS (ESI) *m/z*: 576 [M+1]⁺; Anal. Calcd. for C₂₇H₂₁N₅O₆S₂: C, 56.34; H, 3.68; N, 12.17. Found: C, 56.44; H, 3.53; N, 12.23.

Ethyl-4-methyl-2-(2-oxo-3-(1-(2-(4-(*p*-tolyl)thiazol-2-yl)hydrazono)ethyl)-2H-chromen-6-yl)thiazole-5-carboxylate (6e)

Yellow solid; mp. 235-237 °C; IR (KBr, cm⁻¹) $\nu_{\max}$: 3412 (NH), 1718 (C=O), 1622 (C=N), 1578 (C=C); ¹H NMR (400MHz, DMSO-*d*₆): δ 1.31 (t, 3H, *J* = 7.2 Hz), 2.28 (s, 3H), 2.32 (s, 3H), 2.71 (s, 3H), 4.28-4.33 (m, 2H), 7.22 (d, 2H, *J* = 8.0 Hz), 7.29 (s, 1H), 7.54 (d, 1H, *J* = 8.8 Hz), 7.77 (d, 2H, *J* = 8.0 Hz), 8.19-8.22 (m, 1H), 8.29 (s, 1H), 8.56 (s, 1H), 11.45 (s, 1H); ¹³C NMR (100 MHz, DMSO-*d*₆): δ 14.0, 16.0, 17.1, 20.7, 61.1, 103.4, 116.9, 119.5, 121.5, 125.4, 127.1, 128.4, 129.1, 129.6, 131.8, 136.7, 139.9, 144.3, 154.7, 158.5, 160.20, 161.2, 167.3, 169.1; MS (ESI) *m/z*: 545 [M+1]⁺; Anal. Calcd. for C₂₈H₂₄N₄O₄S₂: C, 61.75; H, 4.44; N, 10.29. Found: C, 61.87; H, 4.21; N, 10.12.

Ethyl-2-(3-(1-(2-(4-(4-methoxyphenyl)thiazol-2-yl)hydrazono)ethyl)-2-oxo-2H-chromen-6-yl)-4-methylthiazole-5-carboxylate (6f)

Red solid; mp. 216-218 °C; IR (KBr, cm⁻¹) $\nu_{\max}$: 3418 (NH), 1713 (C=O), 1612 (C=N), 1186 (C-O-C); ¹H NMR (400MHz, DMSO-*d*₆): δ 1.33 (t, 3H, *J* = 7.2 Hz), 2.31 (s, 3H), 2.73 (s, 3H), 3.69 (s, 3H), 4.30-4.36 (m, 2H), 7.30 (d, 2H, *J* = 7.6 Hz), 7.57 (d, 1H, *J* = 8.4 Hz), 7.92 (d, 1H, *J* = 7.2 Hz), 8.20-8.24 (m, 1H), 8.33 (s, 1H), 8.55-8.60 (m, 2H), 8.76 (s, 1H), 11.39 (s, 1H); ¹³C NMR (100 MHz, DMSO-*d*₆): δ 14.6, 16.2, 18.0, 55.0, 61.5, 104.6, 117.1, 119.0, 121.5, 125.9, 127.3, 128.5, 129.2, 129.9, 132.3, 136.8, 139.9, 144.3, 155.9, 158.6, 160.3, 162.5, 168.3, 169.4;

MS (ESI) m/z : 561 $[M+1]^+$; Anal. Calcd. for $C_{28}H_{24}N_4O_5S_2$: C, 59.98; H, 4.31; N, 9.99. Found: C, 60.14; H, 4.19; N, 10.13.

Ethyl-2-(3-(1-(2-(4-([1,1'-biphenyl]-4-yl)thiazol-2-yl)hydrazono)ethyl)-2-oxo-2H-chromen-6-yl)-4-methylthiazole-5-carboxylate (6g)

Pale yellow solid; mp. 209-211 °C; IR (KBr, cm^{-1}) ν_{max} : 3427 (NH), 1728 (C=O), 1613 (C=N), 1577 (C=C); 1H NMR (400MHz, DMSO- d_6): δ 1.32 (t, 3H, J = 6.8 Hz), 2.30 (s, 3H), 2.71 (s, 3H), 4.28-4.34 (m, 2H), 7.37 (t, 1H, J = 7.2 Hz), 7.45-7.50 (m, 3H), 7.55 (d, 1H, J = 8.8 Hz), 7.71-7.74 (m, 4H), 7.98 (d, 2H, J = 8.4 Hz), 8.20-8.22 (m, 1H), 8.30 (s, 1H), 8.56 (s, 1H), 11.48 (s, 1H); MS (ESI) m/z : 606 $[M]^+$; Anal. Calcd. for $C_{33}H_{26}N_4O_4S_2$: C, 65.33; H, 4.32; N, 9.23. Found: C, 65.51; H, 4.24; N, 9.14.

Ethyl-4-methyl-2-(2-oxo-3-(1-(2-(4-(2-oxo-2H-chromen-3-yl)thiazol-2-yl)hydrazono)ethyl)-2H-chromen-6-yl)thiazole-5-carboxylate (6h)

Yellow solid; mp. 220-222 °C; IR (KBr, cm^{-1}) ν_{max} : 3416 (NH), 1717 (C=O), 1608 (C=N), 1563 (C=C); 1H NMR (400MHz, DMSO- d_6): δ 1.32 (t, 3H, J = 7.2 Hz), 2.30 (s, 3H), 2.71 (s, 3H), 4.29-4.34 (m, 2H), 7.41 (d, 1H, J = 8.0 Hz), 7.46 (d, 1H, J = 8.0 Hz), 7.56 (d, 1H, J = 9.2 Hz), 7.63 (d, 1H, J = 7.2 Hz), 7.82 (t, 2H, J = 7.2 Hz), 8.21-8.23 (m, 2H), 8.32 (s, 1H), 8.59 (s, 1H), 11.52 (s, 1H); ^{13}C NMR (100 MHz, DMSO- d_6): δ 14.4, 17.2, 19.0, 104.4, 116.9, 120.1, 120.9, 123.9, 124.2, 127.2, 128.6, 128.9, 132.0, 133.1, 138.6, 148.7, 158.6, 160.2, 161.4, 167.4, 167.9, 168.8; MS (ESI) m/z : 598 $[M]^+$; Anal. Calcd. for $C_{30}H_{22}N_4O_6S_2$: C, 60.19; H, 3.70; N, 9.36. Found: C, 60.32; H, 3.86; N, 9.19.

Ethyl-2-(3-(1-(2-(4-(6-chloro-2-oxo-2H-chromen-3-yl)thiazol-2-yl)hydrazono)ethyl)-2-oxo-2H-chromen-6-yl)-4-methylthiazole-5-carboxylate (6i)

Shiny bright yellow; mp. 215-217 °C; IR (KBr, cm^{-1}) ν_{max} : 3417 (NH), 1736 (C=O), 1718 (C=O), 1619 (C=N), 1565 (C=C), 757(C-Cl); 1H NMR (400MHz, DMSO- d_6): δ 1.32 (t, 3H, J = 7.6), 2.30 (s, 3H), 2.71 (s, 3H), 4.29-4.34 (m, 2H), 7.50 (d, 1H, J = 8.8 Hz), 7.54-7.67 (m, 1H), 7.85 (s, 1H), 7.97 (d, 1H, J = 7.2 Hz), 8.22 (d, 1H, J = 7.2 Hz), 8.31 (t, 1H, J = 6.4 Hz), 8.51 (s, 1H), 8.58 (s, 1H), 8.65 (d, 1H, J = 6.0 Hz), 8.76 (s, 1H), 11.55 (s, 1H); MS (ESI) m/z : 634 $[M+1]^+$; Anal. Calcd. for $C_{30}H_{21}ClN_4O_6S_2$: C, 56.91; H, 3.34; N, 8.85. Found: C, 56.84; H, 3.48; N, 8.62.

Ethyl-2-(3-(1-(2-(4-(6-bromo-2-oxo-2H-chromen-3-yl)thiazol-2-yl)hydrazono)ethyl)-2-oxo-2H-chromen-6-yl)-4-methylthiazole-5-carboxylate (6j)

Yellow solid; mp. 228-230 °C; IR (KBr, cm^{-1}) ν_{max} : 3419 (NH), 1735 (C=O), 1717 (C=O), 1606 (C=N), 777 (C-Br); ^1H NMR (400MHz, $\text{DMSO}-d_6$): δ 1.29 (t, 3H, $J = 7.6$), 2.28 (s, 3H), 2.78 (s, 3H), 4.27-4.32 (m, 2H), 7.47 (d, 2H, $J = 8.4$ Hz), 7.52-7.63 (m, 1H), 7.75 (s, 1H), 7.88 (d, 1H, $J = 7.6$ Hz), 8.20 (d, 1H, $J = 7.6$ Hz), 8.29 (t, 1H, $J = 6.8$ Hz), 8.47 (s, 1H), 8.51 (d, 1H, $J = 6.0$ Hz), 8.65 (s, 1H), 8.72 (s, 1H), 11.51. MS (ESI) m/z : 678 $[\text{M}+1]^+$; Anal. Calcd. for $\text{C}_{30}\text{H}_{21}\text{BrN}_4\text{O}_6\text{S}_2$: C, 53.18; H, 3.12; N, 8.27. Found: C, 53.27; H, 3.01; N, 8.41.

Ethyl-2-(3-(1-(2-(4-(6,8-dibromo-2-oxo-2H-chromen-3-yl)thiazol-2-yl)hydrazono)ethyl)-2-oxo-2H-chromen-6-yl)-4-methylthiazole-5-carboxylate (6k)

Yellow solid; mp. 232-234 °C; IR (KBr, cm^{-1}) ν_{max} : 3417 (NH), 1736 (C=O), 1715 (C=O), 1619 (C=N), 1567 (C=C), 778 (C-Br); ^1H NMR (400MHz, $\text{DMSO}-d_6$): δ 1.33 (t, 3H, $J = 7.6$ Hz), 2.30 (s, 3H), 2.75 (s, 3H), 4.31-4.35 (m, 2H), 7.20 (d, 2H, $J = 8.4$ Hz), 7.28 (s, 1H), 7.56 (d, 1H, $J = 8.4$ Hz), 7.75 (s, 1H), 8.16 (s, 1H), 8.26 (s, 1H), 8.54 (s, 1H), 11.39 (s, 1H); MS (ESI) m/z : 757 $[\text{M}+1]^+$; Anal. Calcd. for $\text{C}_{30}\text{H}_{20}\text{Br}_2\text{N}_4\text{O}_6\text{S}_2$: C, 47.63; H, 2.66; N, 7.41. Found: C, 47.47; H, 2.76; N, 7.32.

Ethyl-4-methyl-2-(2-oxo-3-(1-(2-(4-(3-oxo-3H-benzo[f]chromen-2-yl)thiazol-2-yl)hydrazono)ethyl)-2H-chromen-6-yl)thiazole-5-carboxylate (6l)

Bright yellow solid; mp. 242-244 °C; IR (KBr, cm^{-1}) ν_{max} : 3416 (NH), 1719 (C=O), 1615 (C=N), 1561 (C=C); ^1H NMR (400MHz, $\text{DMSO}-d_6$): δ 1.31 (t, 3H, $J = 7.6$ Hz), 2.31 (s, 3H), 2.72 (s, 3H), 4.27-4.33 (m, 2H), 7.56 (d, 2H, $J = 8.8$ Hz), 7.64-7.68 (m, 4H), 7.85 (d, 2H, $J = 8.0$ Hz), 8.11 (d, 1H, $J = 7.6$ Hz), 8.23 (t, 1H, $J = 8.4$ Hz), 8.34 (s, 1H), 8.60 (s, 1H), 11.48 (s, 1H); MS (ESI) m/z : 649 $[\text{M}+1]^+$; Anal. Calcd. for $\text{C}_{34}\text{H}_{24}\text{N}_4\text{O}_6\text{S}_2$: C, 62.95; H, 3.73; N, 8.64. Found: C, 63.05; H, 3.59; N, 8.74.

Spectra of synthesized compounds

^1H NMR (400 MHz, $\text{DMSO}-d_6$) spectrum of compound 3

^{13}C NMR (100 MHz, $\text{DMSO}-d_6$) spectrum of compound 3

IR (KBr) spectrum of compound 6a

¹H NMR (400 MHz, DMSO-*d*₆) spectrum of compound 6a

^{13}C NMR (100 MHz, DMSO- d_6) spectrum of compound 6a

Mass spectrum of compound 6a

IR (KBr) spectrum of compound 6b

¹H NMR (400 MHz, DMSO-d₆) spectrum of compound 6b

^{13}C NMR (100 MHz, $\text{DMSO}-d_6$) spectrum of compound 6b

^1H NMR (400 MHz, $\text{DMSO}-d_6$) spectrum of compound 6c

^{13}C NMR (100 MHz, $\text{DMSO}-d_6$) spectrum of compound 6c

IR (KBr) spectrum of compound 6d

^1H NMR (400 MHz, $\text{DMSO}-d_6$) spectrum of compound 6d

^{13}C NMR (100 MHz, $\text{DMSO}-d_6$) spectrum of compound 6d

¹H NMR (400 MHz, DMSO-*d*₆) spectrum of compound 6e

¹³C NMR (100 MHz, DMSO-*d*₆) spectrum of compound 6e

Mass spectrum of compound 6e

^{13}C NMR (100 MHz, DMSO- d_6) spectrum of compound 6f

IR (KBr) spectrum of compound 6f

¹H NMR (400 MHz, DMSO-d₆) spectrum of compound 6g

IR (KBr) spectrum of compound 6h

¹H NMR (400 MHz, DMSO-d₆) spectrum of compound 6h

^{13}C NMR (100 MHz, $\text{DMSO}-d_6$) spectrum of compound 6h

Mass spectrum of compound 6h

IR (KBr) spectrum of compound 6i

¹H NMR (400 MHz, DMSO-d₆) spectrum of compound 6i

IR (KBr) spectrum of compound 6j

IR (KBr) spectrum of compound 6k

IR (KBr) spectrum of compound 6l