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Synthesis, characterization and biological evaluation of some new pyrazolyl bipyridyl substituted dicoumarins

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The present study describes the synthesis of pyrazolyl bipyridyl substituted dicoumarin derivatives carried out by the reaction of various 3-[3-{3-(coumarin-3-yl)-1-phenyl-1H-pyrazol-4-yl}acryloyl]coumarins (coumarin chalcones) **(4a-f)** with appropriate pyridoyl methyl pyridinium iodide salts **(5-7)** in the presence of ammonium acetate in refluxing acetic acid. All synthesized target compounds **(8a-f)**, **(9a-f)** and **(10a-f)** were characterized by IR, ^{1}H-NMR, ^{13}C-APT and representative mass spectral data. The compounds were subjected to *in vitro* antimicrobial screening against a representative panel of bacteria (*Bacillus subtilis, Staphylococcus aureus, Escherichia coli, Salmonella typhi)* and fungi *(Aspergillus niger, Candida albicans*).

Keywords: coumarins, coumarin chalcones, Krohnke reaction, antimicrobial screening.

INTRODUCTION

Coumarins are the best known aromatic lactones isolated from various natural sources. Over the past decades coumarins have attracted strong scientific interest stemming from their broad spectrum of pharmacological properties such as antimicrobial [1], anti-inflammatory [2], CNS depressant [3], antioxidant [4], antitumor [5], antiviral [6], antiasthamatic [7], antiulcer [8], cytotoxic properties [9], etc. The actual trend in the field of synthetic chemistry of coumarins is the introduction of a heterocyclic moiety as a substituent either in the lactone or benzene ring in a coumarin core structure possessing various biological activities [10-14] in which pyrazolyl-substituted coumarins are well documented in the literature and known for their various biological properties like antimicrobial [15], anticonvulsant [16], antioxidant, antihyperglycemic [17], etc. They also show fluorescent and absorption emission characteristics [18].

Bipyridines are compounds formed by coupling of two pyridine rings. Bipyridines are widely used in coordination and supramolecular chemistry [19]. They also possess effective biological properties like antibacterial [20], antifungal [21], antimycoplasmal [22], antimalarial [23], antitumor activity [24], etc. Moreover, a large number of bipyridines are used in photocatalysis [25], as chemosensors [26] and luminescent probes for biomolecular systems [27].

Thus, considering the importance of pyrazolyl-substituted coumarins and bipyridines, it was thought worthwhile to synthesize some new coumarin derivatives having both of these structural features in a single molecule. Therefore, in the present work, synthesis of various pyrazolyl-bipyridyl-substituted dicoumarins was carried out and all synthesized compounds were screened for their antimicrobial activity.

EXPERIMENTAL

All melting points are uncorrected. All reactions were performed with commercially available reagents used without further purification. Organic solvents were purified by standard methods and stored over molecular sieves. All IR spectra (KBr disc) were recorded on a Shimadzu FTIR 8400-S spectrometer. ^{1}H-NMR and ^{13}C-APT spectra were recorded on a Bruker Advance 400 spectrometer operating at 400 MHz for ^{1}H-NMR and 100 MHz for ^{13}C-APT. The chemical shift (δ) is reported in ppm using chloroform-d as a solvent and calibrated standard solvent signal. Mass spectra were recorded on a Shimadzu QP 2010 spectrometer. All compounds were routinely checked for completion of the reaction on silica gel 60 F254 TLC plates and their spots were visualized by exposure to a UV lamp, iodine vapour or $KMnO_4$ reagents. In the present work, various 3-{4'-[3'''-(coumarin-3'''-yl)-1'''-phenyl-1*H*-pyrazol-4'''-yl]-2',2''-bipyridin-6'-yl}coumarins **(8a-f)**, 3-{4'-[3'''-(coumarin-3'''-yl) -1'''-phenyl-1*H*-pyrazol-4'''-yl]-2',3''-bipyridin-6'-yl}coumarins **(9a-f)** and 3-{4'-[3'''-(coumarin-3'''-yl)-1'''-phenyl-1*H*-pyrazol-4'''-yl]-2',4''-bipyridin-6'-yl}coumarins **(10a-f)** were synthesized by the reaction of various 3-[3-{3-(coumarin-3-yl)-1-phenyl-1*H*-pyrazol-4-yl} acryloyl]coumarins (coumarin chalcones) **(4a-f)** with appropriate pyridoyl methyl pyridinium iodide salts **(5-7)** under Krohnke's reaction conditions.

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Compounds **(5-7)** were prepared according to the procedure given in [28].

General procedure for the preparation of 3-[3-{3-(coumarin-3-yl)-1-phenyl-1H-pyrazol-4-yl}acryloyl]coumarins (4a-f) (coumarin chalcones).

In a 100-mL round bottom flask, an appropriate 3-[(1-phenyl-4-formyl)pyrazol-3-yl]coumarin (0.01 mole) and an appropriate 3-acetyl coumarin (0.01 mole) were taken in 50 mL of ethanol. Catalytic amount of piperidine was added and the reaction mixture was stirred for 10 min at room temperature. The mixture was then refluxed on a water bath for 4 h. It was allowed to cool to room temperature. The solid product separated out was filtered off, washed with cold ethanol and dried. It was recrystallized from ethanol. Coumarin chalcones **4a**, **4b**, **4d** and **4e** were prepared according to procedure [29].

5,6-Benzo-3-[3-{3-(coumarin-3-yl)-1-phenyl-1H-pyrazol-4-yl}acryloyl]coumarin (4c): IR(KBr, ν_{max}, cm^{-1}): 1736(C=O stretching of δ-lactone of coumarin), 1697(α,β unsaturated carbonyl group), 1651 and 1543(aromatic C=C and C=N stretchings), 748 and 687(C-H bending vibrations of mono substituted benzene ring), 3063(aromatic C-H stretching). ^{1}H NMR(400MHz, $CDCl_3$, δ): 7.36-8.49 (18H, multiplet, fifteen aromatic protons + two olefinic proton + C_5' proton of pyrazole ring), 9.36 and 9.41(2H, two singlets, C_4 and C_4'' protons of coumarin).

5,6-Benzo-8''-methoxy-3-[3-{3-(coumarin-3-yl)-1-phenyl-1H-pyrazol-4-yl}acryloyl] coumarin (4f): IR(KBr, ν_{max}, cm^{-1}): 1705(C=O stretching of δ-lactone of coumarin), 1682(α,β unsaturated carbonyl group), 1605 and 1535(aromatic C=C and C=N stretchings), 748(C-H bending vibrations of mono substituted benzene ring), 3055(aromatic C-H stretching). ^{1}H NMR(400MHz, $CDCl_3$, δ): 4.03(3H, singlet, OCH_3), 7.18-8.48(17H, multiplet, fourteen aromatic protons + two olefinic proton + C_5' proton of pyrazole ring), 9.35 and 9.38(2H, two singlets, C_4 and C_4'' protons of coumarin).

General procedure for the synthesis of 3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',2''-bipyridin-6'-yl}coumarins (8a-f), 3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',3''-bipyridin-6'-yl}coumarins (9a-f) and 3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',4''-bipyridin-6'-yl} coumarins (10a-f).

In a 100-mL round bottom flask equipped with a condenser, guard tube and magnetic needle, an appropriate pyridoyl methyl pyridinium iodide salt **(5)** or **(6)** or **(7)** (0.003 mole) in glacial acetic acid (15mL) was taken. To this ammonium acetate (0.03 mole) was added under stirring at room temperature. Then a solution of an appropriate 3-[3-{3-(coumarin-3-yl)-1-phenyl-1H-pyrazol-4-yl} acryloyl]coumarin (coumarin chalcone) **(4a-f)** (0.003 mole) in glacial acetic acid (15 mL) was added under stirring at room temperature and the reaction mixture was further stirred for 1 h at room temperature and then refluxed for 8 h at 140°C. It was then allowed to cool to room temperature and was poured into ice-cold water (75 mL). The crude solid obtained was extracted with chloroform (3 × 30 mL). The organic layer was washed with 5% sodium bicarbonate solution (3 × 20 mL), water (2 × 20 mL) and dried over anhydrous sodium sulphate. The removal of chloroform under reduced pressure gave a crude material which was subjected to column chromatography using silica gel and chloroform-ethylacetate (9:1) as an eluent to give compounds **(8a-f)**, **(9a-f)** and **(10a-f)**. The compounds were recrystallized from chloroform-hexane.

3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',2''-bipyridin-6'-yl} coumarin (8a): Yield: 65%, m.p.184-185˚C, IR(KBr, ν_{max}, cm^{-1}): 1729(C=O stretching of δ-lactone of coumarin), 1605 and 1478(aromatic C=C and C=N stretchings), 758 and 691(C-H bending vibrations of mono substituted benzene ring), 3062(aromatic C-H stretching). ^{1}H-NMR(400 MHz, $CDCl_3$, δ): 7.32-7.91 (15H, multiplet, aromatic protons except C_5'''-H, C_4''''-H, C_3'-H, C_5'-H, C_3''-H, C_6''-H, C_4-H), 8.27(1H, singlet, C_5'''-H), 8.52-8.58(4H, multiplet, protons at C_4'''', C_3', C_5' and C_3''), 8.66(1H, poorly resolved doublet of doublet, C_6''-H), 9.00(1H, singlet, C_4-H). ^{13}C-APT(100 MHz, $CDCl_3$, δ): 116.28(CH), 116.68(CH), 118.52(CH), 119.29(CH), 119.49(C), 119.57(C), 121.34(CH), 121.67(C), 121.98(CH), 122.38(C), 123.83(CH), 124.47(CH), 124.49(CH), 125.16(C), 127.27(CH), 127.35(CH), 128.47(CH), 128.92(CH), 129.62(CH), 131.96(CH), 132.01(CH), 136.89(CH), 139.54(C), 142.04(C), 142.46(CH), 143.50(CH), 145.81(C), 149.02(CH), 150.64(C), 153.90(C), 154.35(C), 155.73(C), 155.93(C), 159.66(CO of coumarin), 160.09(CO of coumarin). Mass (*m/z*): M^+ 586(24%), 562(18%), 456(26%), 328(12%), 91(17%), 77(100%), 56(28%). Anal.Calcd. for $C_{37}H_{22}N_4O_4$: C, 75.76; H, 3.78; N, 9.55 %. Found: C, 75.88; H, 3.87; N, 9.59 %.

8-Methoxy-3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',2''-bipyridin-6'-yl} coumarin (8b): Yield: 60%, m.p.170-172˚C, IR(KBr, ν_{max}, cm^{-1}): 1720(C=O stretching of δ-

lactone of coumarin), 1612 and 1497(aromatic C=C and C=N stretchings), 756 and 694(C-H bending vibrations of mono substituted benzene ring), 2839(aliphatic C-H stretching), 3063(aromatic C-H stretching). ^{1}H-NMR(400MHz, $CDCl_3$, δ): 4.01(3H, singlet, OCH_3), 7.09-7.89(14H, multiplet, aromatic protons except C_5'''-H, C_4''''-H, C_3'-H, C_5'-H, C_3''-H, C_6'' -H, C_4-H), 8.27(1H, singlet, C_5'''-H), 8.49-8.56(4H, multiplet, protons at C_4'''', C_3', C_5' and C_3''), 8.63(1H, poorly resolved doublet of doublet, C_6''-H), 8.94(1H, singlet, C_4-H). ^{13}C-APT(100MHz, $CDCl_3$, δ): 56.25(OCH_3), 113.80(CH), 116.05(CH), 116.78(CH), 118.60(CH), 119.25(C), 119.60(CH), 120.16(C), 120.33(CH), 121.33(CH), 121.55(C), 122.07(CH), 122.41(C), 123.88(CH), 124.32(CH), 124.58(CH), 125.27(C), 127.42(CH), 128.68(CH), 129.67(CH), 131.93(CH), 133.83(C), 136.99(CH), 139.61(C), 140.15(C), 142.32(C), 142.95(CH), 143.85(CH), 146.02(C), 146.92(C), 150.80(C), 154.33(C), 155.86(C), 158.57(CO of coumarin), 159.56(CO of coumarin). Anal.Calcd. for $C_{38}H_{24}N_4O_5$: C, 74.02; H, 3.92; N, 9.09 %. Found: C, 74.10; H, 4.00; N, 9.13 %.

5,6-Benzo3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',2''-bipyridin-6'-yl}coumarin (8c): Yield: 72%, m.p.207-209˚C, IR(KBr, ν_{max}, cm^{-1}): 1712(C=O stretching of δ-lactone of coumarin), 1604 and 1474(aromatic C=C and C=N stretchings), 763 and 686(C-H bending vibrations of mono substituted benzene ring), 3055(aromatic C-H stretching). ^{1}H-NMR(400MHz, $CDCl_3$, δ): 7.33-8.01(17H, multiplet, aromatic protons except C_5'''-H, C_4''''-H, C_3'-H, C_5'-H, C_3''-H, C_6'' -H, C_4-H), 8.26(1H, singlet, C_5'''-H), 8.47-8.63(5H, multiplet, protons at C_4'''', C_3', C_5', C_3'' and C_6''), 9.75(1H, singlet, C_4-H). ^{13}C-APT (100MHz, $CDCl_3$, δ): 113.82(C), 116.27(CH), 116.59(CH), 116.70(C), 117.15(C), 117.77(CH), 118.45(C), 119.31(CH), 119.51(CH), 120.78(CH), 121.24(CH), 121.73(C), 121.89(CH), 121.99(C), 122.48(C), 123.82(CH), 123.97(C), 124.47(CH), 126.11(CH), 127.25(CH), 127.34(C), 128.37(CH), 128.48(CH), 129.11(CH), 129.71(CH), 130.40(C), 131.96(CH), 133.48(CH), 136.93(CH), 138.13(CH), 139.60(C), 142.09(C), 143.56(CH), 149.13(CH), 151.01(C), 155.89(C), 156.10(C), 159.69(CO of coumarin), 160.16 (CO of coumarin). Anal.Calcd. for $C_{41}H_{24}N_4O_4$: C, 77.08; H, 3.80; N, 8.80 %. Found: C, 77.13; H, 3.89; N, 8.96 %.

8''''-Methoxy-3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',2''-bipyridin-6'-yl} coumarin (8d): Yield: 67%, m.p.230-231˚C, IR(KBr, ν_{max}, cm^{-1}): 1728(C=O stretching of δ-lactone of coumarin), 1620 and 1481(aromatic C=C and C=N stretchings), 748 and 678(C-H bending vibrations of mono substituted benzene ring), 2847(aliphatic C-H stretching), 3070(aromatic C-H stretching). ^{1}H-NMR(400MHz, $CDCl_3$, δ): 3.96(3H, singlet, OCH_3), 7.12-7.89(14H, multiplet, aromatic protons except C_5'''-H, C_4''''-H, C_3'-H, C_5'-H, C_3''-H, C_6'' -H, C_4-H), 8.26(1H, singlet, C_5'''-H), 8.47-8.56(4H, multiplet, protons at C_4'''', C_3', C_5' and C_3''), 8.63(1H, poorly resolved doublet of doublet, C_6''-H), 8.94(1H, singlet, C_4- H). ^{13}C-APT(100MHz, $CDCl_3$, δ): 56.34(OCH_3), 114.22(CH), 114.82(C), 116.23(CH), 118.70(CH), 119.52(CH), 120.09(CH), 121.50(CH), 122.09(CH), 123.88(CH), 124.32(CH), 124.54(CH), 126.99(C), 127.24(CH), 127.46(CH), 129.09(CH), 129.72(CH), 130.31(C), 132.01(CH), 135.77(C), 137.12(C), 138.25(C), 139.50(C), 141.07(CH), 142.30(CH), 142.70(C), 143.71(C), 145.75(CH), 146.03(C), 148.98(CH), 150.67(C), 151.86(C), 153.87(C), 154.53(C), 159.11(CO of coumarin), 160.11(CO of coumarin). Anal.Calcd. for $C_{38}H_{24}N_4O_5$: C, 74.02; H, 3.92; N, 9.09 %. Found: C, 74.16; H, 4.02; N, 9.19 %.

8,8''''-Dimethoxy-3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',2''-bipyridin-6'-yl}coumarin (8e): Yield: 64%, m.p.200-202˚C, IR(KBr, ν_{max}, cm^{-1}): 1713(C=O stretching of δ-lactone of coumarin), 1605 and 1474(aromatic C=C and C=N stretchings), 756 and 694(C-H bending vibrations of mono substituted benzene ring), 2908(aliphatic C-H stretching), 3063(aromatic C-H stretching). ^{1}H-NMR(400MHz, $CDCl_3$, δ): 3.96(3H, singlet, OCH_3), 3.98(3H, singlet, OCH_3), 7.10-7.88(13H, multiplet, aromatic protons except C_5'''-H, C_4''''-H, C_3'-H, C_5'-H, C_3''-H, C_6''-H, C_4-H), 8.25(1H, singlet, C_5'''-H), 8.47-8.57(4H, multiplet, protons at C_4'''', C_3', C_5' and C_3''), 8.65(1H, poorly resolved doublet of doublet, C_6''-H), 8.95(1H, singlet, C_4-H). ^{13}C-APT(100MHz, $CDCl_3$, δ): 56.23(OCH_3), 56.32(OCH_3), 113.75(CH), 114.14(CH), 118.65(CH), 119.19(CH), 119.44(CH), 119.92(CH), 120.01(C), 120.22(C), 120.40(CH), 121.35(CH), 121.78(C), 122.13(CH), 122.44(C), 122.53(C), 123.79(CH), 124.28(CH), 124.34(CH), 125.40(C), 127.23(CH), 127.41(CH), 136.87(CH), 139.56(C), 142.21(C), 142.64(CH), 143.60(CH), 144.10(C), 145.79(C), 146.80(C), 147.07(C), 149.02(CH), 150.60(C), 155.70(C), 155.97(C), 158.99(CO of coumarin), 159.52(CO of coumarin). Anal.Calcd. for $C_{39}H_{26}N_4O_6$: C, 72.44; H, 4.05; N, 8.66 %. Found: C, 72.63; H, 4.19; N, 8.73 %.

5,6-Benzo-8''''-methoxy-3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',2''-

bipyridin-6'-yl}coumarin (8f): Yield: 70%, m.p.255-256˚C, IR(KBr, ν_{max}, cm^{-1}): 1720(C=O stretching of δ-lactone of coumarin), 1604 and 1504(aromatic C=C and C=N stretchings), 771 and 671(C-H bending vibrations of mono substituted benzene ring), 2893(aliphatic C-H stretching), 3078 (aromatic C-H stretching). ^{1}H-NMR(400MHz, $CDCl_3$, δ): 3.94(3H, singlet, OCH_3), 7.37-8.36(18H, multiplet, aromatic protons except C_4''''-H, C_3'-H, C_5'-H, C_6''-H, C_4-H), 8.46(1H, poorly resolved doublet of doublet, C_6''-H), 8.65(1H, singlet, C_4''''-H), 8.80(2H, poorly resolved doublet, C_3'-H and C_5'-H), 9.65(1H, singlet, C_4-H). ^{13}C-APT(100MHz, $CDCl_3$, δ): 56.35(OCH_3), 110.07(C), 112.91(C), 113.29(C), 113.76(C), 114.37(CH), 115.72(C), 116.05(CH), 116.83(CH), 116.98(CH), 118.56(C), 119.14(C), 119.54(C), 120.65(CH), 120.80(CH), 121.43(CH), 122.56(CH), 122.75(CH), 123.03(CH), 126.35(CH), 126.48(CH), 128.44(CH), 129.06(C), 129.39(CH), 129.69(CH), 130.04(CH), 130.37(CH), 130.80(C), 131.78(C), 132.30(CH), 138.21(C), 139.46(CH), 145.32(C), 146.55(CH), 147.73(CH), 149.74(C), 154.99(C), 156.78(C), 160.02(CO of coumarin), 161.25(CO of coumarin). Anal.Calcd. for $C_{42}H_{26}N_4O_5$: C, 75.67; H, 3.93; N, 8.40 %. Found: C, 75.82; H, 4.03; N, 8.52 %.

3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',3''-bipyridin-6'-yl}coumarin (9a): Yield: 69%, m.p.184-185˚C, IR(KBr, ν_{max}, cm^{-1}): 1735(C=O stretching of δ-lactone of coumarin), 1605 and 1466(aromatic C=C and C=N stretchings), 756 and 625(C-H bending vibrations of mono substituted benzene ring), 3062(aromatic C-H stretching). ^{1}H-NMR(400 MHz, $CDCl_3$, δ): 7.25-8.31(18H, multiplet, aromatic protons + C_4''''-H except C_3'-H, C_6''-H, C_4-H and C_2''- H), 8.46(1H, doublet, J=1.2 Hz, C_3'-H), 8.58(1H, poorly resolved doublet of doublet, C_6''-H), 8.90(1H, singlet, C_4-H), 9.20(1H, poorly resolved doublet, C_2''-H). ^{13}C-APT(100MHz, $CDCl_3$, δ): 114.48(CH), 114.97(C), 116.26(CH), 116.75(CH), 117.92(CH), 119.16(C), 119.45(CH), 119.49(C), 121.16(CH), 121.40(C), 122.06(C), 123.55(CH), 124.58(CH), 124.69(CH), 127.35(CH), 128.49(CH), 129.06(CH), 129.62(CH), 132.21(CH), 134.50(CH), 134.73(C), 139.38(C), 142.29(C), 142.79(CH), 143.66(CH), 145.59(C), 148.25(CH), 149.70(CH), 151.29(C), 153.90(C), 154.28(C), 159.73(CO of coumarin), 160.11(CO of coumarin). Anal.Calcd. for $C_{37}H_{22}N_4O_4$: C, 75.76; H, 3.78; N, 9.55 %. Found: C, 75.88; H, 3.87; N, 9.59 %.

8-Methoxy-3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',3''-bipyridin-6'-yl} coumarin (9b): Yield: 68%, m.p.168-170˚C, IR(KBr, ν_{max}, cm^{-1}): 1718(C=O stretching of δ-lactone of coumarin), 1597 and 1474(aromatic C=C and C=N stretchings), 763 and 694(C-H bending vibrations of mono substituted benzene ring), 2962(aliphatic C-H stretching), 3055(aromatic C-H stretching). ^{1}H-NMR(400MHz, $CDCl_3$, δ): 3.97(3H, singlet, OCH_3), 7.09-8.37(17H, multiplet, aromatic protons + C_4''''-H except C_3'-H, C_6''-H, C_4-H and C_2''-H), 8.54(1H, doublet, J=1.2 Hz, C_3'-H), 8.64(1H, poorly resolved doublet of doublet, C_6''-H), 8.94(1H, singlet, C_4-H), 9.26(1H, doublet, J=1.6 Hz, C_2''-H). ^{13}C-APT(100MHz, $CDCl_3$, δ): 56.41(OCH_3), 114.23(CH), 116.31(CH), 118.55(CH), 119.51(C), 119.57(C), 119.80(CH), 119.96(CH), 121.27(CH), 121.61(C), 122.08(C), 122.24(CH), 124.56(CH), 124.64(CH), 124.92(C), 127.33(CH), 127.41(CH), 129.07(CH), 129.64(CH), 131.03(C), 132.27(CH), 139.42(C), 139.63(CH), 142.50(C), 142.94(CH), 143.84(CH), 144.02(CH), 145.65(C), 146.42(C), 147.18(C), 150.34(CH), 151.53(C), 153.98(C), 154.34(C), 160.19(CO of coumarin), 160.64(CO of coumarin). Anal.Calcd. for $C_{38}H_{24}N_4O_5$: C, 74.02; H, 3.92; N, 9.09 %. Found: C, 74.13; H, 4.01; N, 9.17 %.

5,6-Benzo3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',3''-bipyridin-6'-yl} coumarin (9c): Yield: 70%, m.p.207-209˚C, IR(KBr, ν_{max}, cm^{-1}): 1720(C=O stretching of δ-lactone of coumarin), 1603 and 1497(aromatic C=C and C=N stretchings), 748 and 687(C-H bending vibrations of mono substituted benzene ring), 3070(aromatic C-H stretching). ^{1}H-NMR(400MHz, $CDCl_3$, δ): 7.35- 8.46(20H, multiplet, aromatic protons + C_4''''-H except C_3'-H, C_6''-H, C_4-H and C_2''-H), 8.63(1H, doublet, J=0.8 Hz, C_3'-H), 8.68(1H, poorly resolved doublet of doublet, C_6''-H), 9.37(1H, poorly resolved doublet, C_2''-H), 9.77(1H, singlet, C_4-H). ^{13}C-APT(100MHz, $CDCl_3$, δ): 113.23(C), 113.78(C), 114.45(C), 114.88(CH), 115.73(C), 116.48(CH), 116.78(CH), 117.98(CH), 119.17(C), 119.56(CH), 121.27(CH), 121.42(C), 121.96(CH), 123.37(C), 123.70(C), 124.71(CH), 126.22(CH), 127.33(CH), 127.43(CH), 128.50(CH), 128.60(CH), 129.00(CH), 129.66(C), 130.32(C), 132.23(CH), 133.76(CH), 134.63(CH), 138.49(CH), 139.45(C), 142.44(C), 143.68(CH), 148.27(CH), 149.62(CH), 151.66(C), 154.30(C), 159.73(CO of coumarin), 160.01(CO of coumarin). Anal.Calcd. for $C_{41}H_{24}N_4O_4$: C, 77.08; H, 3.80; N, 8.80 %. Found: C, 77.13; H, 3.89; N, 8.96 %.

8''''-Methoxy-3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',3''-bipyridin-6'-yl} coumarin (9d): Yield: 72%, m.p.230-231˚C, IR(KBr, ν_{max}, cm^{-1}): 1713(C=O stretching of δ-lactone of coumarin), 1605 and 1458(aromatic C=C and C=N stretchings), 756 and 694(C-H bending vibrations of mono substituted benzene ring), 2977(aliphatic C-H stretching), 3055(aromatic C-H stretching). ^{1}H-NMR(400MHz, $CDCl_3$, δ): 3.97(3H, singlet, OCH_3), 7.13-8.39(17H, multiplet, aromatic protons + C_4''''-H except C_3'-H, C_6''-H, C_4-H and C_2''-H), 8.52(1H, doublet, J=1.2 Hz, C_3'-H), 8.65(1H, doublet of doublet, J=4.8 and 1.2 Hz, C_6''- H), 8.96(1H, singlet, C_4-H), 9.29(1H, doublet, J=1.6 Hz, C_2''-H). ^{13}C-APT(100 MHz, $CDCl_3$, δ): 56.32(OCH_3), 114.23(CH), 116.27(CH), 118.21(CH), 119.54(C), 119.56(CH), 119.80(C), 119.95(CH), 121.43(CH), 121.58(C), 122.23(C), 123.68(CH), 124.55(CH), 124.61(CH), 127.42(CH), 128.48(CH), 129.06(CH), 129.71(CH), 132.26(CH), 134.86(CH), 135.03(C), 139.43(C), 141.07(C), 142.49(C), 142.92(CH), 143.81(CH), 146.10(C), 147.11(C), 148.07(CH), 149.45(CH), 151.44(C), 153.99(C), 159.12(CO of coumarin), 161.00(CO of coumarin). Anal.Calcd. for $C_{38}H_{24}N_4O_5$: C, 74.02; H, 3.92; N, 9.09 %. Found: C, 74.16; H, 4.02; N, 9.19 %.

8,8''''-Dimethoxy-3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',3''-bipyridin-6'-yl} coumarin (9e): Yield: 64%, m.p.198-200˚C, IR(KBr, ν_{max}, cm^{-1}): 1720(C=O stretching of δ-lactone of coumarin), 1504 and 1478(aromatic C=C and C=N stretchings), 771 and 694(C-H bending vibrations of mono substituted benzene ring), 2962(aliphatic C-H stretching), 3063(aromatic C-H stretching). ^{1}H-NMR(400MHz, $CDCl_3$, δ): 3.95 and 3.97(6H, two singlets, 2 x OCH_3), 7.09-8.57(18H, multiplet, aromatic protons + C_4''''-H except C_4-H and C_2''-H), 8.65(1H, singlet, C_4-H), 8.93(1H, poorly resolved doublet, C_2''-H). ^{13}C-APT(100MHz, $CDCl_3$, δ): 56.25(OCH_3), 56.34(OCH_3), 114.17(CH), 114.88(C), 118.71(CH), 119.21(C), 119.54(CH), 119.92(C), 120.05(CH), 120.27(C), 120.42(CH), 121.41(CH), 121.79(C), 122.21(CH), 122.51(C), 122.60(CH), 123.06(C), 123.81(CH), 123.94(C), 124.23(CH), 124.34(C), 127.26(CH), 127.44(CH), 129.74(CH), 137.02(CH), 139.08(CH), 139.66(C), 142.64(C), 143.47(CH), 144.13(CH), 144.96(C), 147.02(C), 149.17(CH), 150.74(C), 159.01(C), 161.82(CO of coumarin), 163.97(CO of coumarin). Anal.Calcd. for $C_{39}H_{26}N_4O_6$: C, 72.44; H, 4.05; N, 8.66 %. Found: C, 72.63; H, 4.19; N, 8.73 %.

5,6-Benzo-8''''-methoxy-3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',3''-bipyridin-6'-yl}coumarin (9f): Yield: 68%, m.p.255-256˚C, IR(KBr, ν_{max}, cm^{-1}): 1728(C=O stretching of δ-lactone of coumarin), 1605 and 1474(aromatic C=C and C=N stretchings), 779 and 686(C-H bending vibrations of mono substituted benzene ring), 2932(aliphatic C-H stretching), 3062(aromatic C-H stretching). ^{1}H-NMR(400MHz, $CDCl_3$, δ): 3.97(3H, singlet, OCH_3), 7.13-8.52 (19H, multiplet, aromatic protons + C_4''''-H except C_3'-H, C_6''-H, C_4-H and C_2''- H), 8.65(1H, poorly resolved doublet, C_3'-H), 8.71(1H, doublet, poorly resolved doublet of doublet, C_6''-H), 9.41(1H, poorly resolved doublet, C_2''-H), 9.83(1H, singlet, C_4-H). ^{13}C-APT(100MHz, $CDCl_3$, δ): 56.32(OCH_3), 115.61(C), 115.98(CH), 116.04(CH), 117.59(C), 118.44(C), 119.06(C), 119.48(C), 120.76(CH), 121.01(CH), 121.31(CH), 122.34(CH), 123.22(CH), 126.51(CH), 128.09(CH), 128.57(CH), 128.94(C), 129.70(CH), 129.80(CH), 130.09(CH), 130.51(CH), 130.87(C), 131.69(CH), 131.74(C), 137.87(C), 140.04(CH), 141.77(CH), 143.13(C), 143.90(CH), 144.22(C), 144.53(CH), 145.24(C), 145.88(CH), 146.70(C), 148.13(CH), 148.59(C), 152.39(C), 155.63(C), 161.09(CO of coumarin), 161.52(CO of coumarin). Anal.Calcd. for $C_{42}H_{26}N_4O_5$: C, 75.67; H, 3.93; N, 8.40 %. Found: C, 75.82; H, 4.03; N, 8.52 %.

3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',4''-bipyridin-6'-yl} coumarin (10a): Yield: 63%, m.p.184-185˚C, IR(KBr, ν_{max}, cm^{-1}): 1713(C=O stretching of δ-lactone of coumarin), 1604 and 1466(aromatic C=C and C=N stretchings), 748 and 686(C-H bending vibrations of mono substituted benzene ring), 3063(aromatic C-H stretching). ^{1}H-NMR(400MHz, $CDCl_3$, δ): 7.34-7.96 (16H, multiplet, aromatic protons except C_5'''-H, C_4''''-H, C_3'-H, C_2''-H, C_6''-H, C_4-H), 8.27(1H, singlet, C_5'''-H), 8.40(1H, singlet, C_4''''-H), 8.59(1H, poorly resolved doublet, C_3'-H), 8.74(2H, doublet, J=6.0Hz, C_2''-H and C_6''-H), 8.98 (1H, singlet, C_4-H). ^{13}C-APT(100MHz, $CDCl_3$, δ): 107.79(C), 110.30(C), 113.13(C), 115.97(CH), 116.89(CH), 117.11(CH), 118.08(CH), 118.40(C), 118.80(C), 119.53(CH), 120.70(CH), 126.07(CH), 126.30(CH), 126.77(CH), 129.18(CH), 129.42(CH), 130.13(CH), 134.15(CH), 136.45(CH), 138.18(C), 143.14(CH), 145.28(C), 147.62(CH), 149.45(CH), 149.94(C), 150.41(C), 153.71(C), 154.08(C), 154.39(C), 159.80(CO of coumarin), 160.03(CO of coumarin). Anal.Calcd. for $C_{37}H_{22}N_4O_4$: C,

75.76; H, 3.78; N, 9.55 %. Found: C, 75.88; H, 3.87; N, 9.59 %.

8-Methoxy-3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',4''-bipyridin-6'-yl} coumarin (10b): Yield: 66%, m.p.170-172˚C, IR(KBr, ν_{max}, cm^{-1}): 1728(C=O stretching of δ-lactone of coumarin), 1605 and 1504(aromatic C=C and C=N stretchings), 756 and 694(C-H bending vibrations of mono substituted benzene ring), 2970(aliphatic C-H stretching), 3055(aromatic C-H stretching). ^{1}H-NMR(400MHz, $CDCl_3$, δ): 4.00(3H, singlet, OCH_3), 7.13-7.96(15H, multiplet, aromatic protons except C_5'''-H, C_4''''-H, C_3'-H, C_2''-H, C_6''-H, C_4-H), 8.27(1H, singlet, C_5'''-H), 8.40(1H, singlet, C_4''''-H), 8.59(1H, poorly resolved doublet, C_3'-H), 8.74(2H, poorly resolved doublet, C_2''-H and C_6''-H), 8.96(1H, singlet, C_4-H). ^{13}C-APT(100MHz, $CDCl_3$, δ): 56.51(OCH_3), 110.13(C), 112.96(C), 115.79(C), 116.93(CH), 117.54(C), 118.19(C), 118.62(C), 118.78(C), 119.04(C), 120.66(CH), 121.16(CH), 122.11(CH), 122.35(C), 123.43(C), 123.98(CH), 124.13(CH), 124.88(CH), 124.99(CH), 125.36(CH), 125.85(CH), 126.00(CH), 126.19(CH), 126.48(C), 129.20(CH), 129.94(CH), 130.39(CH), 134.04(CH), 136.07(C), 138.24(C), 142.64(CH), 153.71(C), 159.83(CO of coumarin), 160.28(CO of coumarin). Anal.Calcd. for $C_{38}H_{24}N_4O_5$: C, 74.02; H, 3.92; N, 9.09 %. Found: C, 74.14; H, 4.02; N, 9.18 %.

5,6-Benzo3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',4''-bipyridin-6'-yl} coumarin (10c): Yield: 70%, m.p.205-207˚C, IR(KBr, ν_{max}, cm^{-1}): 1735(C=O stretching of δ-lactone of coumarin), 1612 and 1496(aromatic C=C and C=N stretchings), 779 and 678(C-H bending vibrations of mono substituted benzene ring), 3047(aromatic C-H stretching). ^{1}H-NMR(400MHz, $CDCl_3$, δ): 7.12-7.96(18H, multiplet, aromatic protons except C_5'''-H, C_4''''-H, C_3'-H, C_2''-H, C_6''-H, C_4-H), 8.26(1H, singlet, C_5'''-H), 8.40(1H, singlet, C_4''''-H), 8.58(1H, poorly resolved doublet, C_3'-H), 8.73(2H, poorly resolved doublet, C_2''-H and C_6''-H), 9.28(1H, singlet, C_4-H). ^{13}C-APT(100MHz, $CDCl_3$, δ): 109.93(C), 112.75(C), 113.87(C), 115.58(C), 116.83(CH), 117.08(C), 118.41(CH), 118.57(C), 118.67(C), 118.73(C), 118.95(CH), 120.98(CH), 121.52(CH), 122.93(CH), 123.63(CH), 126.55(CH), 127.18(CH), 128.53(CH), 129.34(CH), 129.76(CH), 130.09(CH), 131.65(CH), 131.72(C), 134.51(CH), 137.81(C), 143.51(C), 144.60(C), 146.19(CH), 147.07(CH), 147.76(CH), 147.98(C), 149.69(CH), 152.78(C), 153.54(C), 160.98(CO of coumarin), 162.27(CO of coumarin). Anal.Calcd. for $C_{41}H_{24}N_4O_4$: C, 77.08; H, 3.80; N, 8.80 %. Found: C, 77.13; H, 3.89; N, 8.96 %.

8''''-Methoxy-3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',4''-bipyridin-6'-yl} coumarin (10d): Yield: 67%, m.p.230-231˚C, IR(KBr, ν_{max}, cm^{-1}): 1720(C=O stretching of δ-lactone of coumarin), 1597 and 1458(aromatic C=C and C=N stretchings), 748 and 694(C-H bending vibrations of mono substituted benzene ring), 2970(aliphatic C-H stretching), 3063(aromatic C-H stretching). ^{1}H-NMR(400MHz, $CDCl_3$, δ): 3.97(3H, singlet, OCH_3), 7.16-7.94(15H, multiplet, aromatic protons except C_5'''-H, C_4''''-H, C_3'-H, C_2''-H, C_6''-H, C_4-H), 8.23(1H, singlet, C_5'''-H), 8.38(1H, singlet, C_4''''-H), 8.54(1H, poorly resolved doublet, C_3'-H), 8.72(2H, doublet, J=4.4 Hz, C_2''-H and C_6''-H), 8.94(1H, singlet, C_4-H). ^{13}C-APT(100MHz, $CDCl_3$, δ): 56.24(OCH_3), 116.71(C), 118.57(CH), 119.28(C), 119.51(CH), 120.22(CH), 120.36(C), 121.29(CH), 121.62(CH), 122.05(C), 122.43(C), 123.80(CH), 124.28(CH), 124.47(CH), 125.33(C), 127.24(CH), 128.45(CH), 129.60(CH), 131.92(CH), 136.84(CH), 139.56(C), 140.09(C), 142.14(C), 142.60(CH), 143.46(CH), 145.82(CH), 146.85(C), 149.03(CH), 150.59(C), 154.34(C), 155.70(C), 155.92(C), 159.48(CO of coumarin), 159.60(CO of coumarin). Anal.Calcd. for $C_{38}H_{24}N_4O_5$: C, 74.02; H, 3.92; N, 9.09 %. Found: C, 74.16; H, 4.02; N, 9.19 %.

8,8''''-Dimethoxy-3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',4''-bipyridin-6'-yl} coumarin (10e): Yield: 69%, m.p.200-202˚C, IR(KBr, ν_{max}, cm^{-1}): 1713(C=O stretching of δ-lactone of coumarin), 1612 and 1497(aromatic C=C and C=N stretchings), 771 and 686(C-H bending vibrations of mono substituted benzene ring), 2962(aliphatic C-H stretching), 3070(aromatic C-H stretching). ^{1}H-NMR(400MHz, $CDCl_3$, δ): 3.97(3H, singlet, OCH_3), 4.00(3H, singlet, OCH_3), 7.13-7.96(14H, multiplet, aromatic protons except C_5'''-H, C_4''''-H, C_3'-H, C_2''-H, C_6''-H, C_4-H), 8.24(1H, singlet, C_5'''-H), 8.38(1H, singlet, C_4''''-H), 8.57(1H, poorly resolved doublet, C_3'-H), 8.73(2H, doublet, J=5.2 Hz, C_2''-H and C_6''-H), 8.95(1H, singlet, C_4-H). ^{13}C-APT(100MHz, $CDCl_3$, δ): 56.32(OCH_3), 56.41(OCH_3), 113.78(C), 116.07(C), 116.80(C), 118.62(CH), 119.25(C), 119.60(CH), 120.15(C), 120.35(CH), 121.33(CH), 121.55(C), 122.03(CH), 122.41(C), 123.88(CH), 124.32(CH), 124.57(CH), 125.28(CH), 127.43(CH), 128.69(CH), 129.67(CH), 131.92(CH), 133.83(C), 136.96(CH), 139.60(C), 140.18(C), 142.35(C), 142.93(CH), 143.85(CH), 146.04(C), 146.92(C), 150.81(C), 154.35(C), 160.06(CO of coumarin), 161.17(CO

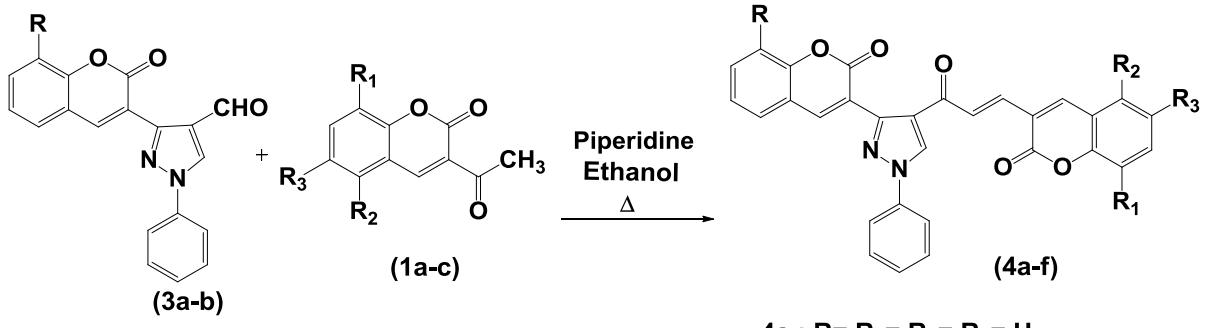
of coumarin). Anal.Calcd. for $C_{39}H_{26}N_4O_6$: C, 72.44; H, 4.05; N, 8.66 %. Found: C, 72.63; H, 4.19; N, 8.73 %.

5,6-Benzo-8''''-methoxy-3-{4'-[3'''-(coumarin-3''''-yl)-1'''-phenyl-1H-pyrazol-4'''-yl]-2',4''-bipyridin-6'-yl}coumarin (10f): Yield: 70%, m.p.255-256˚C, IR(KBr, ν_{max}, cm^{-1}): 1728(C=O stretching of δ-lactone of coumarin), 1604 and 1488 (aromatic C=C and C=N stretchings), 779 and 663(C-H bending vibrations of mono substituted benzene ring), 2977(aliphatic C-H stretching), 3062(aromatic C-H stretching). ^{1}H-NMR(400MHz, $CDCl_3$, δ): 3.98(3H, singlet, OCH_3), 7.14-7.96(17H, multiplet, aromatic protons except C_5'''-H, C_4''''-H, C_3'-H, C_2''-H, C_6''-H, C_4-H), 8.24(1H, singlet, C_5'''-H), 8.39(1H, singlet, C_4''''-H), 8.55(1H, doublet, J=1.2 Hz C_3'-H), 8.72(2H, doublet, J=5.2 Hz, C_2''-H and C_6''-H), 9.60(1H, singlet, C_4-H). ^{13}C-APT(100MHz, $CDCl_3$, δ): 56.38(OCH_3), 111.93(C), 116.12(C), 117.58(CH), 117.73(C), 119.85(C), 121.09(CH), 121.19(CH), 122.21(C), 122.31(C), 123.06(CH), 123.16(CH), 123.21(CH), 123.26(C), 123.42(CH), 124.00(CH), 124.09(CH), 125.11(CH), 126.25(CH), 126.37(CH), 127.38(CH), 127.44(CH), 129.11(CH), 129.31(C), 130.40(C), 131.98(CH), 133.96(CH), 137.03(C), 138.13(C), 139.63(CH), 142.09(C), 143.56(CH), 147.11(C), 149.00(C), 152.89(C), 153.10(C), 159.71(CO of coumarin), 160.19(CO of coumarin). Anal.Calcd. for $C_{42}H_{26}N_4O_5$: C, 75.67; H, 3.93; N, 8.40 %. Found: C, 75.82; H, 4.03; N, 8.52 %.

RESULTS AND DISCUSSION

CHEMISTRY

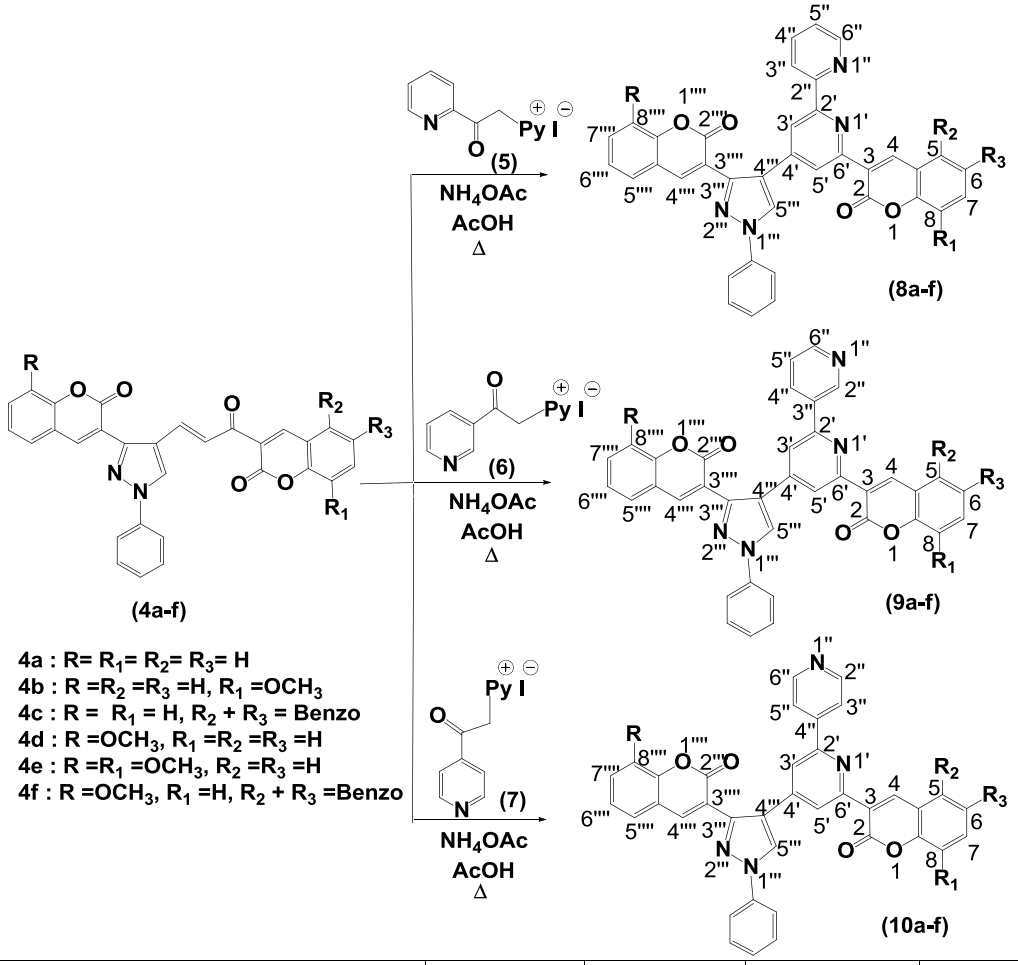
The synthetic pathway adopted to obtain target compounds **(8a-f)**, **(9a-f)** and **(10a-f)** which were synthesized by reacting various 3-[3-{3-(coumarin-3-yl)-1-phenyl-1*H*-pyrazol-4-yl}acryloyl]coumarins (coumarin chalcones) **(4a-f)** with appropriate pyridoyl methyl pyridinium iodide salt **(5-7)** in the presence of ammonium acetate in refluxing acetic acid, is shown in Scheme 1. The structures of all synthesized compounds, supported by IR, ^{1}H-NMR, ^{13}C-APT and representative mass spectral data are shown in the experimental section.



3a : R= H
3b : R= OCH_3

1a : R_1= R_2= R_3= H
1b : R_1= OCH_3, R_2= R_3= H
1c : R_1= H, R_2+ R_3= benzo

4a : R= R_1= R_2= R_3= H
4b : R= R_2= R_3= H, R_1= OCH_3
4c : R= R_1= H, R_2+ R_3= Benzo
4d: R= OCH_3, R_1= R_2= R_3= H
4e : R= R_1= OCH_3,R_2= R_3= H
4f : R= OCH_3, R_1= H, R_2+ R_3= Benzo



Compounds			R	R_1	R_2	R_3
8a:	9a:	10a:	H	H	H	H
8b:	9b:	10b:	H	OCH_3	H	H
8c:	9c:	10c:	H	H	-Benzo-	
8d:	9d:	10d:	OCH_3	H	H	H
8e:	9e:	10e:	OCH_3	OCH_3	H	H
8f:	9f:	10f:	OCH_3	H	-Benzo-	

Scheme 1. Synthetic pathway for the synthesis of the target compounds **(8a-f)**, **(9a-f)** and **(10a-f)**

BIOLOGICAL RESULTS

Antimicrobial Activity

Tthe synthesized target compounds **(8a-f)**, **(9a-f)** and **(10a-f)** were evaluated for their *in vitro* antibacterial activity against two Gram positive bacteria *Staphylococcus aureus* (MTCC 96) and *Bacillus subtilis* (MTCC 441) and two Gram negative bacteria *Escherichia coli* (MTCC 443) and *Salmonella typhi* (MTCC 98). They were also evaluated for their *in vitro* antifungal activity against *Candida albicans* (MTCC 227) and *Aspergillus niger* (MTCC 282) as fungal strains. Broth dilution method was used for the determination of the antibacterial and antifungal activity as recommended by NCCLS [30]. Ampicillin, chloramphenicol, ciprofloxacin, norfloxacin and gentamycin were used as standard antibacterial drugs whereas griseofulvin and nystatin were used as standard antifungal drugs. All MTCC cultures were collected from the Institute of Microbial Technology, Chandigarh and tested against the above mentioned drugs. Mueller-Hinton broth was used as the nutrient medium for the test bacteria and Sabouraud dextrose broth was used for the test fungi. Inoculum size for the test strains was adjusted to 10^8 CFU (Colony Forming Units) per millilitre by comparing the turbidity. Each synthesized compound was diluted with DMSO so as to have a stock solution of 2000 μg/mL concentration. The results were recorded in the form of primary and secondary screening. The synthesized compounds **(8a-f)**, **(9a-f)** and **(10a-f)** were screened for their antibacterial and antifungal activity at the concentration of 1000, 500 and 250 μg/mL for the primary screening. The synthesized compound showing activity against microbes in the

primary screening were further screened in a second set of dilutions at concentrations of 200, 100, 62.5, 50 and 25 µg/mL. The suspension of 10 µL from each well was further incubated and growth was noted at 37°C after 24 h for bacteria and 48 h for fungi. The lowest concentration which showed no visible growth (turbidity) after spot subculture was considered as the minimum inhibitory concentration (MIC) for each compound.

The investigation of the data summarized in Table 1 reveals that many compounds were found to be active against Gram positive bacteria while some of the compounds were found to be active against Gram-negative bacterial and fungal species as compared to that of the standard antimicrobial drugs.

Antimicrobial Results

The final compounds **(8a-f)**, **(9a-f)** and **(10a-f)** were screened for their *in vitro* antibacterial and antifungal evaluation against various bacterial and fungal pathogens by the broth dilution method. Ampicillin, chloramphenicol, norfloxacin, ciprofloxacin, gentamycin, griseofulvin and nystatin were used as standard drugs. The values of MIC are summarized in Table 1.

Table-1 Antimicrobial activity of compounds (8a-f), (9a-f) and (10a-f)

Compound	Minimum Inhibitory Concentration (MIC, µgmL^{-1})							
	Gram positive bacteria			Gram negative bacteria			Fungi	
	B.s.	*S.a.*		*E.c.*	*S.t.*		*A.n.*	*C.a.*
8a	500	500		100	100		500	>1000
8b	500	500		125	100		500	>1000
8c	250	250		200	250		>1000	1000
8d	250	200		200	250		500	1000
8e	500	500		250	500		500	250
8f	100	200		62.5	200		>1000	250
9a	200	200		120	100		1000	250
9b	500	500		200	250		1000	500
9c	100	250		250	200		>1000	1000
9d	500	500		500	100		1000	250
9e	125	125		250	100		500	1000
9f	100	250		125	100		500	250
10a	500	250		500	500		>1000	500
10b	100	250		200	100		1000	500
10c	250	125		250	100		>1000	500
10d	200	100		200	250		500	1000
10e	100	100		250	125		1000	250
10f	250	250		100	200		500	1000
Ampicillin	250	250		100	100		-	-
Chloramphenicol	50	50		50	50		-	-
Ciprofloxacin	50	50		25	25		-	-
Norfloxacin	100	10		10	10		-	-
Gentamycin	1	0.25		0.05	5		-	-
Griseofulvin	-	-		-	-		100	500
Nystatin	-	-		-	-		100	100

B.s.: Bacillus subtilis, S.a.: Staphylococcus aureus, E.c.: Escherichia coli, S.t.: Salmonella typhi, A.n.: Aspergillus niger, C.a.: Candida albicans

The assessment of antimicrobial screening data reveals that compounds **8f**, **9c**, **9f**, **10b** and **10e** (MIC=100 µg/mL) exerted excellent inhibitory activity against Gram positive bacteria *Bacillus subtilis* as compared to standard drug ampicillin (MIC=250 µg/mL) and equal activity to norfloxacin (MIC=100 µg/mL). Compound **9e** (MIC=125

μg/mL) and compounds **9a** and **10d** (MIC=200 μg/mL) showed good activity compared to ampicillin (MIC=250 μg/mL) against *Bacillus subtilis*. Compounds **8c**, **8d**, **10c** and **10f** (MIC=250 μg/mL) were found to be equipotent to ampicillin (MIC=250 μg/mL) against *Bacillus subtilis*. Compounds **10d** and **10e** (MIC=100 μg/mL) exhibited excellent activity compared to ampicillin (MIC=250μg/mL) against *Staphylococcus aureus*. Compounds **9e**, **10c** (MIC=125 μg/mL) and compounds **8d**, **8f** and **9a** (MIC=200 μg/mL) showed good activity compared to ampicillin (MIC=250 μg/mL) against *Staphylococcus aureus*. Compounds **8c**, **9c**, **9f**, **10a**, **10b** and **10f** (MIC=250 μg/mL) were found comparable to ampicillin (MIC=250 μg/mL) against *Staphylococcus aureus*. Compound **8f** (MIC=62.5 μg/mL) showed excellent activity and compounds **8a** and **10f** (MIC=100μg/mL) showed equipotent activity against Gram negative bacteria *Escherichia coli* compared to ampicillin (MIC=100 μg/mL). The compounds **8a**, **8b**, **9a**, **9d**, **9e**, **9f**, **10b** and **10c** (MIC=100μg/mL) were found to be equipotent to ampicillin (MIC=100μg/mL) against Gram negative bacteria *Salmonella typhi*. Compounds **8e**, **8f**, **9a**, **9d**, **9f** and **10e** (MIC=250 μg/mL) were found to be more active than griseofulvin (MIC=500 μg/mL) whereas, compounds **9b**, **10a**, **10b** and **10c** were found to be equipotent to griseofulvin (MIC=500 μg/mL) against *Candida albicans*. None of the tested compounds showed better activity against *Aspergillus Niger* than standard drugs. Upon examining the antimicrobial data it is apparent that some of the compounds exhibit good or equal potency to standard drugs against Gram positive bacterial strains.

CONCLUSION

In summary, we have synthesized pyrazolyl-bipyridyl- substituted dicoumarin derivatives and screened for their *in vitro* antimicrobial evaluation. The present synthetic compounds have the potential to exhibit antimicrobial activity. In particular, they have shown promising antibacterial and antifungal activity against several bacterial and fungal pathogens as compared to standard drugs. These compounds can be considered as lead molecules for future investigations.

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