Available online www.jocpr.com

Journal of Chemical and Pharmaceutical Research, 2012, 4(7):3377-3382

Research Article

ISSN : 0975-7384
CODEN(USA) : JCPRC5

Synthesis, characterization and biological screening of new spirochromanones

Nilesh N Gajera* and Mukesh C Patel

Department of chemistry, P S Science and H D Patel Arts College, S V Campus, Kadi-382 715, North Gujarat, India.

ABSTRACT

A new spirochromanones based on N-[1-(6-bromo-4-oxo-3, 4-dihydro-1'H-spiro[chromene-2,4'-piperidin]-1'-yl)-3-methyl-1-oxobutan-2-yl] benzenesulfonamides were synthesized by series of reactions such as cyclization, deprotation, coupling and condensation. Newly synthesized compounds were characterized by spectroscopic techniques like NMR, IR, Mass spectrometry and elemental analysis. The compounds were tested to emphasize their activity against the fungi and pathogenic strains of bacteria. The compounds bearing fluoro, cyano and difluoromethoxy groups at para position showed competitive antifungal activity, while the compounds carrying either halogen or methyl substituent at ortho position showed significant antibacterial activity.

Keywords: Spirochromanones, Acetyl CoA Carboxylase (ACC), Spectral Analysis, Antimicrobial Activity, Minimum inhibitory concentration (MIC).

INTRODUCTION

The synthesis of chromanones is of great interest in the field of organic chemistry because they exhibits wide variety of well documented biological activity [1-5], including antiviral [6], antimicrobial [7], antiarrhychmic [8], antidiabetic [9,10], Antioxidant and antitumor [11-13]. Recent literature reports that spirochromanone derivatives have been found to be a strong ACC inhibiting effect [14]. ACC exists in plants, parasites, bacteria and fungai, and it participates in the growth of cells, For example, aryloxyphenoxypropionic acid-type herbicides represented by diclofop, and cyclohexanedion-type herbicides represented by sethoxydim exerts their activity by inhibiting ACC in plants [15], and the aryloxyphenoxypropionic acid also exhibits a growth-inhibiting effect in parasites [16]. In addition, sorafen and moiramide B Known as ACC inhibitors exhibit an antibacterial effect and antifungal effect [17-18]. Keeping in view of the biological and medicinal importance of chromones and their derivatives; it was thought interesting to synthesize the new spirochromanones based derivatives. The present article comprises the synthesis, characterization and biological screening of new spirochromanones derivatives.

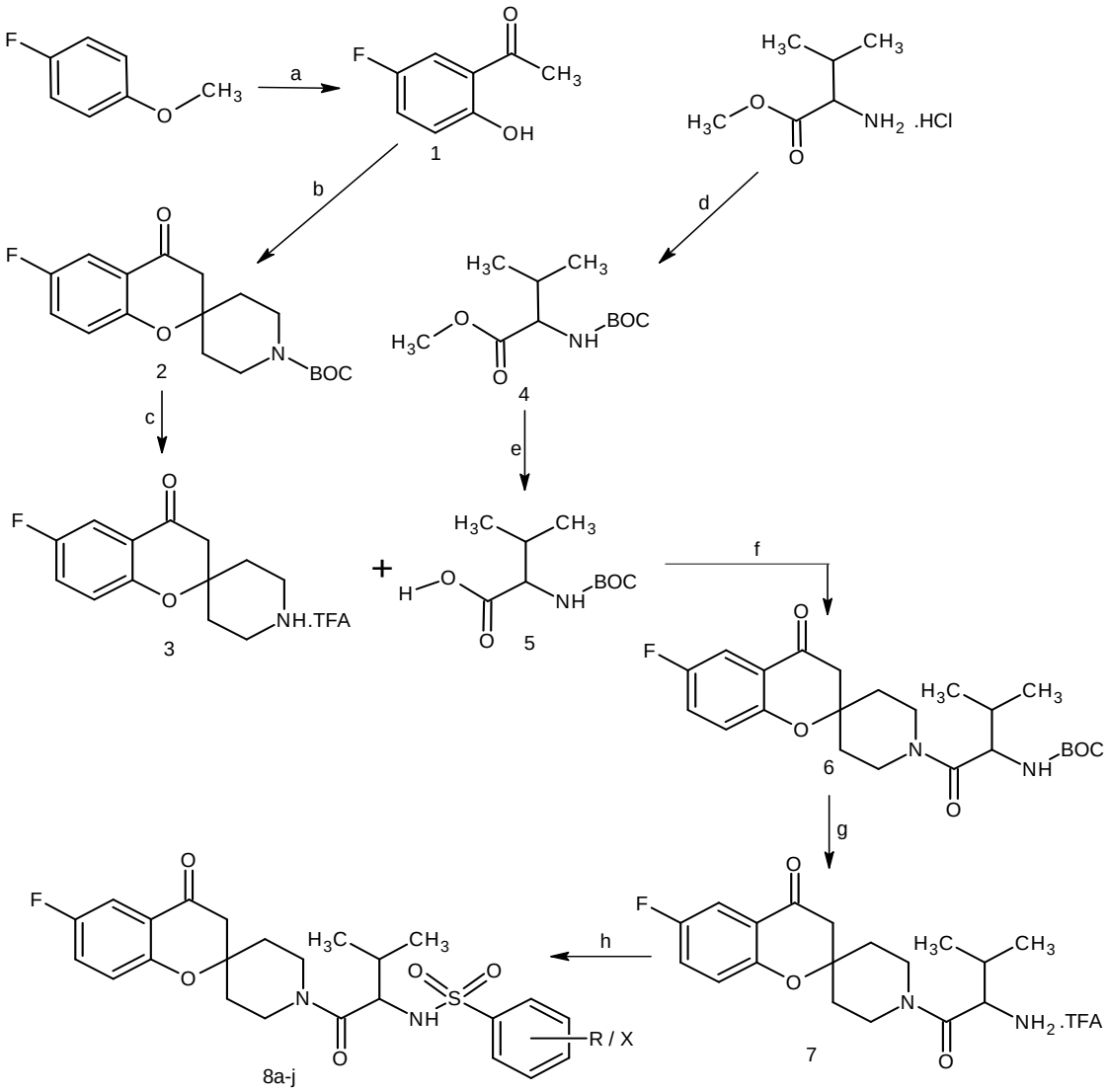
Chemistry

In the present work, compound **3** was synthesized by Fridle-craft acylation & demethylation of 4-Bromoanisol followed by cyclization with N-BOC piperazine in the presence of pyrrolidine in refluxing methanol and BOC deprotection using trifluoroacetic acid in methylene chloride at room temperature reported in literature [14].

Valine methyl ester hydrochloride salt was treated with ditert-butoxy dicarbonate (BOC-anhydride) in the presence of triethylamine at room temperature yielded N- BOC-Valine methyl ester **4**. The ester was hydrolyzed using aqueous NaOH in methanol at room temperature produced BOC-N- valine **5**.

Amide linkage between acid **5** and amine **3** was carried out using N-ethyl-N`-3-dimethylaminopropylcarbodiimide (EDC), 1-Hydroxybenzotriazole (HOBT) and triethylamine in tetrahydrofuran at room temperature produced amide **6**, which on treatment with trifluoroacetic acid in methylene chloride at room temperature yielded TFA salt of deprotected amine **7.** Amine **7** was treated with various phenyl sulfonylchloride in the presence of TEA furnished sulfonamide derivatives **8a-j**.

Reaction Scheme:



Reagents & conditions: (a) Anhy. $AlCl_3$, CH_3COCl, MDC, 50°C, 10 hr (b) N-BOC-4- piperidone, CH_3OH, 65°C, 12 hr (c) TFA, MDC, rt, 15 hr (d) di *tert*-butoxydicarbonate, TEA, MDC, rt, 8 hr (e)NaOH, 70% MeOH in water, rt, 6 hr (f) N-Ethyl-N`-dimethylaminopropylcarbodiimide, TEA, HOBT, THF, rt, 15 hr (g) TFA, MDC, rt, 12 hr (h) Ar-SO_2Cl, TEA, THF, rt, 5-10 hr.

The structures of **8a-j** were confirmed by IR, ^{1}H NMR and MS. ^{1}H NMR spectra of compound showed that -NH-SO_2- resonates near 5.9 δ as a doublet of doublet and -$(CH_3)_2$ resonates near 0.9 δ as a multiplet. Protons of spirocyclic piperidine, -$C(CH_2)_2$- and -$N(CH_2)_2$- resonates near 1.0-2.1 δ(4H, multiplet) and 2.6-4.3 δ(4H, multiplet) respectively. 2H doublet resonates near 2.6 δ due to -CO-CH_2- of chromanones ring. In IR spectra, stretching band of sulfonamide linked -NH- absorption observed near 3281 cm^{-1} and -SO_2- absorption observed near 1351 cm^{-1} and

1125 cm^{-1}. Ar-CO- , -NH-CO- absorption observed near 1693 cm^{-1} and 1634 cm^{-1} respectively. Absorption bends of ether linkage of chromanone ring observed near 1270 cm^{-1} and 1166 cm^{-1}.

EXPERIMENTAL SECTION

The melting points of the compounds were determined by open capillary method and were uncorrected. IR Spectra of the compounds were recorded on Perkin-Elmer FT-IR spectrophotometer using KBr disc method. ^{1}H-NMR spectra were recorded on Bruker DRX-400(400 MHz FT NMR) using TMS as a reference and sample was prepared in $CDCl_3$ and DMSO-d_6. Chemical shifts are expressed in parts per million. Mass spectra were recorded on water-2996 LCMS instrument. The solvents used were of AR grade and they were used without further purification. Reaction monitoring and purity of the compounds were checked by TLC using silica gel $_{254}F_{60}$.

Synthesis of 6-bromo-1-(tert-butoxycarbonyl) Spiro (chroman-2,4`-piperidine)-4-one (6)
To a stirred solution of **5**(0.013M), N-ethyl-N`-3-dimethylaminopropylcarbodiimide (0.013 M), 1-Hydroxybenzotriazole (0.001 M) and triethylamine (0.039 M) in methylene chloride, amine **3** (0.012 M) was added at room temperature. Progress of reaction was monitored by TLC. After completion of reaction, solvent was evaporated under reduced pressure. Product was partitioned between water (200 ml) and ethyl acetate (100 ml). Ethyl acetate layer was passed through Na_2SO_4 and evaporated to dryness. The residual product was purified on a silica gel column, packed in hexane. Column was eluted with hexane: Ethyl acetate (80: 20 v/v) gave pure compound **6** as yellow semi solid mass (yield: 54 %).

6-Bromo- Spiro (chroman-2,4`-piperidine)-4-one . TFA salt (7)
To the clear solution of **6** (0.011 M) in methylene chloride (25 ml), trifluoroacetic acid (0.11 M) was added at room temperature and stirred at room temperature for 8 h. Progress of reaction was monitored by TLC. After completion of reaction, solvent was evaporated under reduced pressure. Residual mass was stirred with diethyl ether (100 ml) and separated solid was filtered off, washed with diethyl ether (10 ml) and dried under reduced pressure to gave **7** as a yellow solid (yield: 78 %).

General procedure for synthesis of *N*-[1-(6-bromo-4-oxo-3,4-dihydro-1'*H*-spiro[chromene-2,4'-piperidin]-1'-yl)-3-methyl-1-oxobutan-2-yl]benzene sulfonamide derivative (8)
To a homogeneous cooled solution of compound **7** (0.0059 M), catalytic dimethylamino pyridine and triethylamine (0.0064 M) in dry methylene chloride, aryl sulfonyl chloride (0.0065 M) was added and stirred at room temperature. Progress of reaction was monitored by TLC. After completion of reaction, solvent was evaporated under reduced pressure. An oily residue was dissolved in ethyl acetate and washed with 5 % aqueous HCl (50 ml) followed by saturated sodium bicarbonate solution (50 ml) and finally with water (50 ml). Ethyl acetate layer was passed through Na_2SO_4 and evaporated to dryness. The solid residue was purified on a silica gel column, packed in hexane. Column was eluted with hexane: ethyl acetate (80: 20 v/v) gave pure compound **8a-j** as white solid.

N-[2-(6-Fluoro-4-oxo-2-propyl-chroman-2-yl)ethyl]-2-[(2-chloro-4-fluoro-phenyl) sulfonylamino]-3-methyl-butanamide (8a): Yield: 72 %; mp:203-205˚C ; Anal. Calcd. for $C_{24}H_{25}ClF_2N_2O_5S$: C, 54.70; H, 4.78; N; 5.32 %. Found: C, 54.51; H, 4.62; N, 5.15 %; IR (KBr, cm^{-1}): 3281, 1693, 1634, 1351, 1270, 1166, 1125; 1H-NMR (400 MHz, DMSO-d6, δ / ppm): 0.94(6H, m, -C(C**H**$_3$)$_2$), 1.17 -2.10(4H, m, -C(C**H**$_2$)$_2$-), 1.86(1H, m, -C**H**-(CH$_3$)$_2$), 2.65(2H, d, -CO-C**H**$_2$-), 2.75-4.31(4H, m, -N(C**H**$_2$)$_2$-), 4.01(1H, m, -NH-C**H**-CO-$_{valine}$), 5.98(1H, dd, -N**H**-SO$_2$-), 6.93-6.96(1H, m, Ar-H), 7.12(1H, m, Ar-H), 7.22-7.24(1H, m, Ar-H), 7.26-7.30(1H, m, Ar-H), 7.52(1H, m, Ar-H), 8.03(1H, t, Ar-H); MS: *m/z* 525 (M-1).

N-[2-(6-Fluoro-4-oxo-2-propyl-chroman-2-yl)ethyl]-2-[(4-bromophenyl)sulfonyl amino]-3-methyl-butanamide (8b): Yield: 80 %; mp:211-214˚C ; Anal. Calcd. for $C_{24}H_{26}BrFN_2O_5S$: C, 52.08; H, 4.74; N; 5.06 %. Found: C, 51.89; H, 4.53; N, 4.94 %; IR (KBr, cm^{-1}): 3283, 1692, 1638, 1345, 1256, 1168, 1130; 1H-NMR (400 MHz, $CDCl_3$, δ / ppm): 0.97(6H, m, -C(C**H**$_3$)$_2$), 1.19 -2.11(4H, m, -C(C**H**$_2$)$_2$-), 1.89(1H , m, -C**H**-(CH$_3$)$_2$), 2.69(2H, d, -CO-C**H**$_2$-), 2.78-4.34(4H, m, -N(C**H**$_2$)$_2$-), 3.98(1H, m, -NH-C**H**-CO-$_{valine}$), 5.94(1H, dd, -N**H**-SO$_2$-), 6.98-6.99(1H, m, Ar-H), 7.16(1H, m, Ar-H), 7.21-7.23(2H, m, Ar-H), 7.24-7.28(1H, m, Ar-H), 7.53(1H, m, Ar-H), 7.98(1H, t, Ar-H); MS: *m/z* 554 & 556 (M+1).

N-[2-(6-Fluoro-4-oxo-2-propyl-chroman-2-yl)ethyl]-3-methyl-2-(o-tolylsulfonyl amino)butanamide (8c): Yield: 68 %; mp:217-219˚C; Anal. Calcd. for $C_{25}H_{29}FN_2O_5S$: C, 61.46; H, 5.98; N; 5.73 %. Found: C, 61.68; H,

6.11; N, 5.88 %; IR (KBr, cm^{-1}): 3279, 1693, 1635, 1334, 1265, 1178, 1132; 1H-NMR (400 MHz, $CDCl_3$, δ / ppm): 0.95(6H, m, -C(C**H**$_3$)$_2$), 1.21 -2.02(4H, m, -C(C**H**$_2$)$_2$-), 1.89(1H , m, -C**H**-(CH$_3$)$_2$), 2.61(2H, d, -CO-C**H**$_2$-), 2.69 (3H, s,Ar-C**H**$_3$), 2.81-4.21(4H, m, -N(C**H**$_2$)$_2$-), 3.84(1H, m, -NH-C**H**-CO-$_{valine}$), 5.68(1H, dd, -N**H**-SO_2-), 6.90-6.95(1H, m, Ar-H), 7.19-7.24(1H, m, Ar-H), 7.28-7.33(2H, m, Ar-H), 7.47-7.53(2H, m, Ar-H), 7.93(1H, d, Ar-H); MS: *m/z* 489 (M+1).

N-[2-(6-Fluoro-4-oxo-2-propyl-chroman-2-yl)ethyl]-2-[(4-cyanophenyl)sulfonyl amino]-3-methyl-butanamide (8d) : Yield: 79 %; mp:179-182˚C; Anal. Calcd. for $C_{25}H_{26}FN_3O_5S$: C, 60.11; H, 5.25; N; 8.41 %. Found: C, 59.96; H, 5.07; N, 8.17 %; IR (KBr, cm^{-1}): 3252, 2233, 1691, 1633, 1342, 1249, 1173, 1128; 1H-NMR (400 MHz, $CDCl_3$, δ / ppm): 0.94(6H, m, -C(C**H**$_3$)$_2$), 1.17 -2.10(4H, m, -C(C**H**$_2$)$_2$-), 1.88(1H , m, -C**H**-(CH$_3$)$_2$), 2.69(2H, d, -CO-C**H**$_2$-), 2.79-4.39(4H, m, -N(C**H**$_2$)$_2$-), 4.02 (1H, m, -NH-C**H**-CO-$_{valine}$), 5.85(1H, dd, -N**H**-SO_2-), 6.82-6.87(1H, m, Ar-H), 7.23(1H, m, Ar-H), 7.52(1H, m, Ar-H), 7.68(2H, t, Ar-H), 7.73(2H, t, Ar-H); MS : *m/z* 500 (M+1).

N-[2-(6-Fluoro-4-oxo-2-propyl-chroman-2-yl)ethyl]-3-methyl-2-[(3,4,5-trifluoro phenyl) sulfonylamino] butanamide (8e): Yield: 87 %; mp:188-191˚C; Anal. Calcd. for $C_{24}H_{24}F_4N_2O_5S$: C, 54.54; H, 4.58; N; 5.30 %. Found: C, 54.50; H, 4.52; N, 5.26 %; IR (KBr, cm^{-1}): 3268, 1694, 1638, 1344, 1250, 1165, 1132; 1H-NMR (400 MHz, DMSO-d6, δ / ppm): 0.88(6H, m, -C(C**H**$_3$)$_2$), 1.15 -2.15(4H, m, -C(C**H**$_2$)$_2$-), 1.85(1H , m, -C**H**-(CH$_3$)$_2$), 2.7(2H, d, -CO-C**H**$_2$-), 2.75-4.25(4H, m, -N(C**H**$_2$)$_2$-), 3.97(1H ,m, -NH-C**H**-CO-$_{valine}$), 7.05(1H, m, Ar-H), 7.42-7.57(2H, m, Ar-H), 7.71(1H, t, Ar-H), 7.77(1H, t, Ar-H), 8.16-8.28(1H, dd, -N**H**-SO_2-); MS : *m/z* 529 (M+1).

N-[2-(6-Fluoro-4-oxo-2-propyl-chroman-2-yl)ethyl]-3-methyl-2-[[4-(trifluoro methoxy)phenyl]sulfonylamino]butanamide (8f) : Yield: 71 %; mp:160-162˚C; Anal. Calcd. for $C_{25}H_{26}F_4N_2O_6S$: C, 53.76; H, 4.69; N; 5.02 %. Found: C, 53.62; H, 4.72; N, 5.11%; IR (KBr, cm^{-1}): 3281, 1693, 1634, 1348, 1258, 1175, 1133; 1H-NMR (400 MHz, $CDCl_3$, δ / ppm): 0.96(6H, m, -C(C**H**$_3$)$_2$), 1.11-2.04(4H, m, -C(C**H**$_2$)$_2$-), 1.86(1H , m, -C**H**-(CH$_3$)$_2$), 2.65(2H, d, -CO-C**H**$_2$-), 2.75-4.26(4H, m, -N(C**H**$_2$)$_2$-), 4.02 (1H, m, -NH-C**H**-CO-$_{valine}$), 6.02(1H, dd, -N**H**-SO_2-), 6.93-6.98(1H, m, Ar-H), 7.01-7.13(1H, m, Ar-H), 7.53(1H, m, Ar-H), 7.39(2H, t, Ar-H), 7.73(2H, t, Ar-H); MS: *m/z* 558 (M+1).

N-[2-(6-Fluoro-4-oxo-2-propyl-chroman-2-yl)ethyl]-2-[(4-isopropylphenyl)sulfonyl amino]-3-methyl-butanamide (8g) : Yield: 62 %; mp:184-186˚C; Anal. Calcd. for $C_{27}H_{33}FN_2O_5S$: C, 62.77; H, 6.44; N; 5.42 %. Found: C, 62.48; H, 6.26; N, 5.17 %; IR (KBr, cm^{-1}): 3275, 1695, 1636, 1349, 1254, 1149, 1137; 1H-NMR (400 MHz, $CDCl_3$, δ / ppm): 0.94 (6H, m, -C(C**H**$_3$)$_2$), 1.09 -1.99(4H, m, -C(C**H**$_2$)$_2$-), 1.31(6H, m, Ar-CH-(C**H**$_3$)$_2$), 1.90(1H , m, -C**H**-(CH$_3$)$_2$), 2.61(2H, d, -CO-C**H**$_2$-), 2.89-4.17(4H, m, -N(C**H**$_2$)$_2$-), 2.99(1H, m, Ar-C**H**-(CH$_3$)$_3$), 3.90(1H, m, -NH-C**H**-CO-$_{valine}$), 5.72(1H, d, -N**H**-SO_2-), 6.89(1H, m, Ar-H), 7.24(1H, m, Ar-H), 7.35(2H, d, Ar-H), 7.53(1H, m, Ar-H), 7.73(2H, t, Ar-H); MS : *m/z* 517 (M+1).

N-[2-(6-Fluoro-4-oxo-2-propyl-chroman-2-yl)ethyl]-2-[(4-fluoro-2-methyl-phenyl) sulfonylamino]-3-methyl-butanamide (8h) : Yield: 77 %; mp:185-187˚C; Anal. Calcd. for $C_{25}H_{28}F_2N_2O_5S$: C, 59.28; H, 5.57; N; 5.53 %. Found: C, 59.09; H, 5.43; N, 5.31 %; IR (KBr, cm^{-1}): 3277, 1694, 1635, 1354, 1268, 1142, 1129; 1H-NMR (400 MHz, CDCl3, δ / ppm): 0.95(6H, m, -C(C**H**$_3$)$_2$), 1.20 -1.99(4H, m, -C(C**H**$_2$)$_2$-), 1.90(1H, m, -C**H**-(CH$_3$)$_2$), 2.68(2H, d, -CO-C**H**$_2$-), 2.72(3H, s, Ar-C**H**$_3$), 2.79-4.24(4H, m, -N(C**H**$_2$)$_2$-), 3.91(1H, m, -NH-C**H**-CO-$_{valine}$), 5.68(1H, dd, -N**H**-SO_2-), 6.90-6.94(1H, m, Ar-H), 7.20-7.24(1H, m, Ar-H), 7.28-7.33(1H, m, Ar-H), 7.38-7.41(1H, m, Ar-H), 7.47-7.53(1H, m, Ar-H), 7.93(1H, m, Ar-H); MS: *m/z* 507 (M+1).

N-[2-(6-Fluoro-4-oxo-2-propyl-chroman-2-yl)ethyl]-2-[(3,5-dimethylphenyl) sulfonylamino]-3-methyl-butanamide (8i) : Yield: 69%; mp:183-185˚C; Anal. Calcd. for $C_{26}H_{31}FN_2O_5S$: C, 62.13; H, 6.22; N; 5.57 %. Found: C, 61.99; H, 6.01; N, 5.27 %; IR (KBr, cm^{-1}): 3271, 1693, 1637, 1368, 1272, 1154, 1130; 1H-NMR (400 MHz, $CDCl_3$, δ/ppm): 0.95(6H, m, -C(C**H**$_3$)$_2$), 1.11-2.07(4H, m, -C(C**H**$_2$)$_2$-), 1.88(1H, m, -C**H**-(CH$_3$)$_2$), 2.59(6H, s, Ar-(C**H**$_3$)$_2$), 2.69(2H, d, -CO-C**H**$_2$-), 2.77-4.19(4H, m, -N(C**H**$_2$)$_2$-), 3.95(1H, m, -NH-C**H**-CO-$_{valine}$), 5.77(1H, d, -N**H**-SO_2-), 6.96-7.11(2H, m, Ar-H), 7.24-7.29(1H, m, Ar-H), 7.32(2H, d, Ar-H), 7.47-7.52(1H, m, Ar-H); MS: *m/z* 503 (M+1).

N-[2-(6-Fluoro-4-oxo-2-propyl-chroman-2-yl)ethyl]-2-[(3-cyano-4-fluoro-phenyl) sulfonylamino]-3-methyl-butanamide (8j) : Yield: 82 %; mp:125-127˚C; Anal. Calcd. for $C_{25}H_{25}F_2N_3O_5S$: C, 58.02; H, 4.87; N; 8.12 %. Found: C, 57.79; H, 4.53; N, 7.91 %; IR (KBr, cm^{-1}): 3253, 1694, 1634, 1372, 1269, 1152, 1129; 1H-NMR (400 MHz, $CDCl_3$, δ/ppm): 0.98(6H, m, -C(C**H**$_3$)$_2$), 1.13-2.13(4H, m, -C(C**H**$_2$)$_2$-), 1.89(1H, m, -C**H**-(CH$_3$)$_2$), 2.71(2H, d, -

CO-C**H**$_2$-), 2.78-4.31(4H, m, -N(C**H**$_2$)$_2$-), 3.97(1H, m, -NH-C**H**-CO-$_{valine}$), 5.74(1H, d, -N**H**-SO$_2$-), 6.90-6.94(1H, m, Ar-H), 7.20-7.24(1H, m, Ar-H), 7.38-7.41(1H, m, Ar-H), 7.47-7.53(1H, m, Ar-H), 7.68-7.71(1H, m, Ar-H), 7.93 (1H, m, Ar-H); MS: *m/z* 516 (M-1).

RESULTS AND DISCUSSION

Antimicrobial Activity

The MICs of synthesized compounds were carried out by broth micro dilution method as described by rattan [19]. Antibacterial activity was screened against two gram positive bacteria (S. aureus, B. subtilis) and two gram negative bacteria (E. coli, P. aeruginosa). Ciprofloxacine was used as standard antibacterial agent. Antifungal activity was screened against fungi C, albicans. Fluconazole was used as a standard antifungal agent. All cultures were collected from institute of microbial technology, Chandigarh and tested against above mentioned known drugs. Mueller hinton broth was used as nutrient medium to grow and dilute the drug suspension for the test. Inoculum size for test strain was adjusted to 108 CFU (Colony Forming Unit) per milliliter by comparing the turbidity. DMSO was used as diluents to get desired concentration of drugs to test upon standard bacterial strains. Serial dilutions were prepared in primary and secondary screening. The control tube containing no antibiotic was immediately sub cultured (before inoculation) by spreading a loopful evently over a quarter of plate of medium suitable for the growth of the test organism and put for incubation at 37 C overnight. The tubes were then incubated overnight. The MIC of the control organism was read to check the accuracy of the drug concentrations. The lowest concentration inhibiting growth of the organism was recorded as the MIC. All the tubes showing visible growth (in the same manner as control tube described above) was sub cultured and incubated overnight at 37 C The amount of growth from the control tube before incubation (which represents the original inoculums) was compared. Subcultures might show: similar number of colonies indicating bacteriostatic; a reduced number of colonies indicating a partial or low bactericidal activity and no growth if the whole inoculums has been killed. The test must include a second set of the same dilutions inoculated with an organism of known sensitivity. Each synthesized drug was diluted containing 6400µg/ml concentration, as a stock solution. In primary sreening1280µg/ml, 640µg/ml, 320µg/ml concentrations of the synthesized drugs was taken. The active synthesized drugs found in the primary screening were similarly diluted to obtain 160µg/ml, 80µg/ml, 40µg/ml and 20µg/ml concentrations. The highest dilution showing at least 99% inhibition is taken as MIC.

Table-2: Biological evaluation of the compounds VIIIa-j

S No.	Compound (R/X)	MIC, µg/ml				
		Bacterial strain				Fungi
		P.aeruginosa	*E.coli*	*S.aureas*	*B.subtilis*	*C.albicans*
1	2-Cl,4-F	640	160	640	160	160
2	4-Br	1280	320	320	320	320
3	2-CH3	1280	80	1280	80	320
4	4-CN	640	640	640	160	80
5	3,4,5-F	>1280	320	320	80	320
6	4-OCF3	>1280	640	640	160	160
7	4-CH(CH3)2	>1280	640	1280	160	640
8	4-F, 2-CH3	640	1280	1280	320	640
9	3,5-CH3	1280	1280	1280	80	320
10	3-CN, 4-F	>1280	320	1280	320	320
Control	*CIPROFLOXACIN*	10	20	10	5	-
Control	*FLUCANAZOLE*	-	-	-	-	10

Minimal bactericidal concentration showed that, some of the newly synthesized compound showed little improved bactericidal activity. All compounds displayed moderate activity against all bacterial strains compared to standard drug *ciprofloxacine.* Compounds **8a** (2-chloro-4-fluorophenyl) and **8d** (4-cyanophenyl) showed overall good antibacterial activity amongst all compounds. It was observed that **8c** (o-methyl) displayed very good activity against *E. Coli & B. subtilis* but poor activity against *P. auriginosa* & *S. aureas.* **8c** (2-methylphenyl) displayed best activity (80 µg/ml) against E. coli.**8e** (3,4,5-trifluorophenyl) and **8j** (3-cyano-4-fluorophenyl) were moderately active against *E. coli* but remaining all showed poor activity. **8b** (4-bromo) and **8e** (3,4,5-trifluoro) showed moderate activity (320 µg/ml) against *S. aureas* while all other derivatives showed poor activity. **8c** (2-methylphenyl), **8e** (3,4,5-trifluorophenyl) and **8i** (3,5-dimethylphenyl) displayed good activity (80 µg/ml) against *B. Subtilis*, **8a** (2-methylphenyl), **8d** (4-cyanophenyl), **8f** (4-trifluoromethoxyphenyl) and **8g** (4-isopropyl) possessed moderate activity while remaining showed poor activity against *B. Subtilis*.

Minimal fungicidal activity showed that **8d** (4-cyanophenyl) showed good activity against *C. albicans*, **8a** (2-chloro-4-fluorophenyl) showed little lower activity (160 µg/ml) then **8d**, while remaining all possessed moderate activity (in between 320-640 µg/ml) against *C. albicans.*

CONCLUSION

The antimicrobial test results revealed that some of the compounds were moderately active. However, the activities of the tested compounds were lesser than those of antimicrobial and antifungal agents used as a reference. The compound bearing fluoro, cyano and difluoromethoxy groups at para position showed competitive antifungal activity, while the compound carrying either halogen or methyl substituent at ortho position showed significant antibacterial activity.

Acknowledgement
The authors are thankful to the DNC Pharma. Ltd. and Vaibhav Analytical Services for providing laboratory facilities and analytical support and to the central diagnostic laboratory, Surat, Gujarat, India for biological screening facility. Authors are also thankful to the UGC, New Delhi for research funding under research project [No. 39-715/2010(SR)].

REFERENCES

[1] X. Zheng, Y. Hu, j. Liu and K. Ouyang, *Bioorg. Med. Chem. lett.*, **2005**, 15, 4531.
[2] G. Toker, E. Kuepeli, M. Memisoglu, E. Yesilada, *J. Ethnopharmaacol*, **2004**, 95, 393.
[3] S.S. Costa, A. Jossang, B. Bodo, M. I. M. Souza and V. L. G. Moraes, *J. Net. Prod*., **1994**, 57, 1503.
[4] G. P. Ellis and I. M. Lockhart, *Chromans and Tocopherols*; Wily: New York, **1981**.
[5] J. B. Harborne, H. J. Marby, and H. Marby, *The Flevonoids*; Academic Press; New York, **1975**.
[6] M. T. Flevin, J. D. Rizzo, A. Khilevich, A. Kucherenko, A. K. Sheinkman, V. Vilaychack, L. Lin, W. Chen, E. M. Greenwood, T. Pengsuperp, J. M. Pezzuto, S. M. Hughes, T. M. Flevin, M. Cibulski, W. A. Boulanger, R. L. Shone and Z. Xu, *J. Med. Chem.*, **1996**, 39, 1303.
[7] D. Tasdemir, M. Kaiser, R. Brun, V. Yardley, T. J. Schmidt, F. Tosun and P Ruedi, *Antimicrob. Agents Chemother*., **2006**, 50, 1352.
[8] R. Mestre, D. Escande and I. Cavero, *Euro. J. Pharmacol*.,**1990**, 183, 1239.
[9] H. Matsuda, T. Morikawa and M. Yoshikawa, *Pure Appl. Chem.*, **2002**, 74,1301.
[10] A. P. Jorge, H. Horst, E. De Souza, M. G. Pizzolatti and F. R. M. B. Silva, *Chem.-Biol. Interect.*, **2004**, 149, 89.
[11] L. C. Chang and A. D. Kinghorn, InTringali, C., Ed.; Bioactive compounds from Natural sources: Isolation, characterization and Biological properties; Tayler & Francis; London, **2001**, Cheptor-5.
[12] J. B. Harborne, *The Flevanoids*, Chapman and Hall; London, **1994**.
[13] F. M. Dean, Naturally Occurring Oxygen Ring Compounds;Butterworth: London, **1963**.
[14] T. Lino, H. Jona, J. Shibata, T. Shimamura, T. Yamakawa and L. Yang. MERCK AND CO. (Pat. no: **US 20080171767 A1**).
[15] J.W. Gronwald, *Biochem. Soc. Trans.*, **1994**, 22(3), 616.
[16] J. Jelenska, A. Sirikhachornkit, R. Haselkorn and P. Gornicki., *J. Bio. Chem.*, **2002**, 277 (26), 23208.
[17] H. f. Vahlensieck, L. Pridzun, H. Reichenbach and A. Hinnen, *Current Genetics*, **1994**, 25(2), 95.
[18] C. Freiberg, N. A. Brunner, G. Schiffer, T. Lampe, J. Pohlmann, M. Brands, M. Raabe, D. Habich and K. Ziegelbauer., *J. Bio. Chem.*, **2004,** 279 (25), 26066.
[19] A. Rattan, In: Antimicrobials in laboratory medicine, New Delhi: Churchill BI, Livingstone, **2000**, 85.