Synthesis and Characterization of 3,5-disubstituted Pyrazoline derivatives

4.1. Introduction

Pyrazoles are five membered heterocycles with two nitrogen atoms in adjacent position and they are also called as Azoles [1]. Recently pyrazole derivatives have been found to possess significant activities such as antimicrobial, anticancer [2], antitumor [3], analgesic [4], antitumor, antimalarial, anticonvulsant and leishmanicidal activity [5]. In addition to this some condensed Pyrazoles have wide spectrum of biological action including anti-inflammatory [6-8] and antiviral activity [9]. Compounds including 1,2-diazole nucleus and their N-substituted derivatives are known to possess corrosion inhibitor tendency [10-12]. Preparation of Pyrazoles using conventional methods was well documented [13-15].

Saleem A Basaif et al. [16] prepared novel pyrazole derivatives by the condensation of para-sulphamyl phenyl hydrazine with chalcones and also further derivatized them with benzene sulphamyl urea and thiourea. The synthesized compounds were evaluated for their antimicrobial activity against *Staphylococcus aureus* and *Escherichia coli*. The compounds (1), (2), (3) and (4) reported to possess moderate activity against the tested microbes.
Pablo Machado et al. [17] in search of new analgesics, synthesized amide based pyrazoles analogues. The newly synthesized compounds were subjected for their analgesic activity. One of each series of pyrazoles (5), (6), (7) & (8) showed significant analgesic effect in the writhing test of mice. The decrease in pain-related behavior obtained was close to that achieved with Aspirin.

Abdel Aziz et al. [18] synthesized new series of amides containing pyrazoles carbohydrazide moiety, compounds (9) and (10) showed significant effects as analgesic and anti-inflammatory compounds.
New Pyrazoline scaffold was synthesized by the condensation between diketones and hydrazine’s by Fong. T.M et al. [19] which on coupling with various hydrazine’s to get new series of amides and tested them for their biological potency. In this research one of the compound (11) showed potent CB1 cannabinoid receptor antagonist activity.

Arunkumar .S et al. [20] synthesized novel pyrazoline derivatives containing Gallic acid and studied their anti-inflammatory activity. The newly synthesized compounds were evaluated for in vivo anti-inflammatory activity by carrageenan induced paw edema test. Among the series compounds (12), (13) and (14) showed comparable activity with standard drug diclofenac sodium.
In search of novel medicinal compounds, Hassan M. Faidallah et al. [21] synthesized variety of poly substituted fused heterocyclic pyrazole rings and evaluated for their synergistic anticancer and antimicrobial activities. In this series many of them showed weak anticancer activity, in addition, they have tested the compounds for their in vitro antibacterial and antifungal activities also using agar–diffusion method. Some of the tested compounds (15), (16), (17) and (18) were indicated significant antifungal and antibacterial activities.

Mohite P.B et al. [22] synthesized new series of pyrazoles containing tetrazole nucleus. Compounds (19) and (20) inhibit the denaturation of albumin in 68.33% and 70.00% respectively when compared with control, possess potent anti-inflammatory activity. Other compounds like (21) and (22) inhibit the denaturation
of proteins by 54.16%, 65.00% and 62.50% respectively. It means these compounds showed good anti-inflammatory activity.

P V Badadhe et al. [23] synthesized novel pyrazolines containing isoxazoline units by the reaction between chalcones with hydroxylamine hydrochloride in the presence of alkali and further the newly synthesized compounds have been screened for their antimicrobial activity. Some of the compounds (23), (24), (25) and (26) showed moderate antimicrobial activity when compared to the reference drug Gentamycin, Cefixime and Ketoconazole.
Alessandro Balbi et al. [24] synthesized thirty-six novel pyrazole derivatives and studied their antiproliferative activity in human lung carcinoma A549 cells, human ovarian adenocarcinoma A2780 cells and murine P388 leukemia cells. The compound (27) showed an antiproliferative activity in all the cell lines tested only at concentrations greater than 50 µM.

V. Madhava Rao et al. [25] synthesized a new series of novel pyrazoles from commercially available 2-Hydroxyacetophenone with different cinnamic acids in pyridine solution using condensing agent POCl₃ and they got good yields. Synthesized compounds were assessed for antibacterial activity by agar cup method and filter paper disc method. Among the series of compounds chlorosubstituted pyrazole ring compounds (28) and (29) were found to be more effective than the remaining compounds were tested against Xanthomonas campestris and Aspergillus niger species.
In an effort to develop synthetic procedure for heterocyclic synthesis, Rafat M et al. [26] prepared pyrazole derivatives by the condensation of 2-Cyanoacetylhydrazine with chloroacetyl chloride in the presence of 1,4-dioxane. The antitumor activity of the novel synthesized compounds was tested against human tumor cell lines, namely breast adenocarcinoma (MCF-7), non-small cell lung cancer (NC1-H460) and CNS cancer (SF-268). Some of the compounds (30), (31) and (32) were found to exhibit higher inhibitory effects towards the tumor cells lines than the Gram positive control doxorubicin.

![Chemical structures](image1)

30 31 32

In search of potent drugs for the group of diseases called leishmaniasis caused by kinetoplastid protozoan parasite, Manuel Sanchez Moreno et al. [27] synthesized pyrazole with benzophthalazine derivatives. They reported that compounds (33) and (34) were more active against two leishmania species and they are less toxic against mammalian cells than the reference drug Glucantime, further they explained that the mono substituted compounds were significantly more effective and less toxic than their disubstituted counterparts.

![Chemical structures](image2)

33 34

Adnan A Bekhit et al. [28] synthesized novel series of pyrazole derivatives and tested for their in vivo anti-malarial activity using mice infected with chloroquine sensitive P. berghei at a dose level 50 µ mol/kg. Compounds (35) and (36) showed in vitro IC50 values lower (p < 0.05) than that of standard drug chloroquine.
phosphate (0.188 ± 0.003 µM) using the RKL9 strains. In this work the pyrazole compounds (35) and (36) with carboxylic group showed potent activity.

![Chemical structures](image)

El Quali et al. [29] studied the thermodynamic and corrosion inhibitor property for C38 steel in molar hydrochloric acid of pyrazole derivatives. The newly synthesized compounds were also characterized by electro chemical technique. All the compounds were tested for their anticorrosion activity, among the tested compounds (37), (38) and (39) exhibited more than 90% anticorrosion activity at the volume 96%. This shows that, the inhibition efficiencies increases with the concentration of the inhibitors.

![Chemical structures](image)

Rigvendra Malik et al. [30] reported the synthesis of sulpha substituted pyrazoles by the condensation between diketones and sulphamyl benzene diazonium chloride. New series of compounds were tested for their anti-inflammatory activity. In this work compound (40) found to possess significant activity at the dosage of 100 and 200 mg/kg, but the effect declined at higher dose.

![Chemical structures](image)
Xiaoming Ding et al. [31] synthesized pyrazole derivatives containing 1,3,4-thiadiazole moiety under microwave irradiation and they were evaluated for their herbicidal and antifungal activities. Two compounds a phenyl group (41) and 4-tert-butylphenyl group (42) possess good herbicidal activity for dicotyledonous Brassica campestris and Raphanus sativa with the inhibition of 90% for root and 90% for stalk at 100 ppm respectively. The biological activity results showed that amide group with 1,3,4-thiadiazole ring reduces the antifungal activity but increases the herbicidal activity.

Present work

In recent year’s extensive research has been carried out on the synthesis and evaluation of pharmacological activities of amide containing Pyrazoline ring for different activities, they have been proved to be important pharmacophores. Among them many substituted amide derivatives containing Pyrazoline ring are of important class of bioactive compounds with a wide spectrum of research importance, because of their pharmacological significance. Literature survey has revealed the positive information on amide with pyrazolines and their biological significance. This prompted us to plan for the synthesis of novel series of amide derivatives bearing 3,5-disubstituted pyrazole moiety with piperazine nucleus as shown in the Scheme 4.1 (page 168) and Scheme 4.2 (page 193). Their biological activities such as antimicrobial, anthelmintic, anti-inflammatory and anticancer activity are discussed in detail in Chapter 6 of the thesis.
4.2. Materials and Methods

All the chemicals were purchased from Spectrochem, Merck India, SD Fine and Sigma–Aldrich. Chemicals and solvents used were of Laboratory Reagent grade. Completion of reaction was confirmed by thin layer chromatography using precoated TLC plates which involves separation of substances of a mixture over a thin layer of an adsorbent (silica gel or alumina) about 0.2 mm thickness is spread over a aluminum sheet of suitable size and solvent systems are dichloromethane / methanol (9:1) or petroleum ether / ethyl acetate (6:4). Further purification of compounds was done using column chromatography. Melting points were determined in one end open capillary tubes on a liquid paraffin bath and are uncorrected. Liquid chromatography mass spectra, HPLC, $^1$H and $^{13}$C NMR spectra were recorded on Agilent Mass spectrometer, Bruker model avance II (399.65 and 300.12 MHz $^1$H NMR) and Bruker model avance II (100 MHz, $^{13}$C NMR) instruments respectively. Chemical shifts were reported in $\delta$ ppm using TMS as an internal standard, elemental analysis (CHN) was performed in an Elementary Micro cube analyzer.
4.3. Experimental

Reagents and conditions: a) $\text{H}_2\text{SO}_4$, EtOH, 80 °C; b) NaH, THF, 35 °C; c) EtOH, cat AcOH, 80 °C; d) TEA, $\text{T}_3\text{P}^{\oplus}$, EDC, RT; e) TFA, rt; f) TEA, $\text{T}_3\text{P}^{\oplus}$, EDC, RT

Scheme 4.1
Procedure for the preparation of Ethyl-3-Methylbutanoate (2):
3-Methylbutanoic acid (1) (0.245 mol) was taken in 250 mL of ethanol and added catalytic amount of Con. H₂SO₄. The reaction mixture was refluxed for 6 h under inert atmosphere at 80 °C, contents were cooled to RT and neutralized with dilute alkali; ester was extracted with ethyl acetate, which was dried over anhydrous MgSO₄ and concentrated under fractional distillation to get the title product (2).

Procedure for the preparation of 2-methyl-4,6-dioxo-6-phenylhexane-1-ylium(4):
NaH (0.216 mol) was taken in dry THF (200 mL) to this added Ethyl-3-Methylbutanoate (2) (0.10 mol) taken in 50 ml dry THF at 15 °C, stirred the reaction mixture for 1 h, then added 2-Fluroacetophenone (0.144 mol) and stirred the reaction mixture for 18 h at 35 °C. After completion of the reaction, THF was removed and reaction mixture was quenched with saturated solution of NH₄Cl (100 ml), organic compound was extracted with chloroform (200 ml) and distilled off. The crude mass was purified by column chromatography with neutral alumina by using pet ether ethyl acetate (7:3) as eluent to get compound (4).

IR: (KBr, cm⁻¹): 1649.8 (CO) (Fig 4.1); ¹H-NMR (CDCl₃) δ ppm: 16.22 (s, 1H, -OH), 7.97-7.91 (m, 1H, Ar-H), 7.48-7.43 (m, 1H, Ar-H), 7.28-7.22 (m, 1H, Ar-H), 7.16-7.09 (m, 1H, Ar-H), 6.26 (s, 1H, enol H), 2.31 (d, J = 1.38 Hz, 2H, CH₂), 2.21-2.11(m, 1H, junction H), 1.01-0.99 (d, J = 6.8 Hz, 6H, (CH₃)₂) (Fig 4.2); LCMS: 223.4 (M+1) (Fig 4.3); HPLC: Purity 94.06% (Fig 4.4); MP:199-200°C; Yield:60%.

Procedure for the preparation of 2-(5-(2-fluorophenyl)-3-isobutyl-1H-pyrazol-1-yl)benzoic acid (6) :
Compound (4) (0.98mol) in ethanol (150 ml) was added 2-Hydrazinobenzoicacid (0.98 mol) and catalytic amount of acetic acid (2-3 drops) contents were stirred for 8 h at 80 °C, after completion of the reaction, solvent was removed, the crude mass was purified by column chromatography. IR: (KBr cm⁻¹): 1694.16 (CO) (Fig 4.5); ¹H-NMR (CDCl₃) δ ppm: 8.10 (d, J =7.5 Hz, 1H, Ar-H), 7.45-7.42 (m, 2H, Ar-H), 7.39-7.27 (m, 1H, Ar-H), 7.13-7.01 (m, 1H, Ar-H), 6.95 (d, J = 7.6 Hz, 1H, =CH), 6.47 (s, 1H, pyrazole-H), 2.64 (d, J = 7.1 Hz, 1H, junction H), 2.10-2.01 (m, 1H, junction-H), 1.02 (d, J = 6.6 Hz, 6H, (CH₃)₂) (Fig 4.6); LCMS: 339.2 (M+1) (Fig
4.7); **HPLC**: Purity 94.06% ([Fig 4.8]; **CHN** analysis complies with the calculated percentage ([Fig 4.9]); **MP**: 211-213°C; **Yield**: 66%.
Fig 4.1: IR spectrum of compound 4
Fig 4.2: $^1$H NMR spectrum of compound 4 (Enol form)
Fig 4.3: LCMS of compound 4
Fig 4.4: HPLC spectrum of compound 4
Fig 4.5: IR spectrum of compound 6
Fig 4.6: $^1$H NMR spectrum of compound 6
Fig 4.7: LCMS of compound 6
Fig 4.8: HPLC spectrum of compound 6
Fig 4.9: CHN Analysis of compound 6

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General Procedure for the preparation tert-butyl 4-(2-(5-(2-fluorophenyl)-3-isobutyl-1H-pyrazol-1-yl)benzoyl)piperazine-1-carboxylate (8) :

Compound (6) (0.05 mol), N-Boc piperazine (7) (0.065 mol), T3P® (0.14 mol), TEA (0.17 mol) were stirred in dry EDC (160 ml) under inert atmosphere at RT for 12 h. The reaction mixture was washed with brine, water, dried over anhydrous Na2SO4, and distilled off. Crude mass was purified by column chromatography to get title compound (8) in good yield. 1H-NMR (CDCl3) δ ppm: 8.10 (d, J = 7.5 Hz, 1H, Ar-H), 7.45-7.42 (m, 2H, Ar-H), 7.39-7.27 (m, 1H, Ar-H), 7.13-7.01 (m, 1H, Ar-H), 6.95 (d, J = 7.6 Hz, 1H, =CH), 6.47 (s, 1H, pyrazole-H), 3.68 (t, J = 5.3 Hz, 4H, CH2NCH2), 3.50 (t, J = 5.2 Hz, 4H, CH2NCH2), 2.64 (d, J = 7.1 Hz, 1H, junction H), 2.10-2.01 (m, 1H, junction-H), 1.60 (s, 9H, (CH3)3), 1.02 (d, J = 6.6 Hz, 6H, (CH3)2). LCMS: 315.12 (M+1). MP: 241-242 °C. Yield: 83 %.

Procedure for the preparation of (2-(5-(2-fluorophenyl)-3-isobutyl-1H-pyrazol-1-yl)phenyl)(piperazine-1-yl)methanone (9) :

A mixture of compound (8) (0.039 mol) and TFA (80 ml) was taken in MDC and stirred the reaction mixture at room temperature. MDC was distilled off, the residue obtained was neutralized with NaHCO3 and finally extracted with MDC. Obtained solid was tritirated with pet ether to get scaffold (9). 1H-NMR (CDCl3) δ ppm: 9.75(s, 1H, NH), 8.10 (d, J = 7.5 Hz, 1H, Ar-H), 7.45-7.42 (m, 2H, Ar-H), 7.39-7.27 (m, 1H, Ar-H), 7.13-7.01 (m, 1H, Ar-H), 6.95 (d, J = 7.6 Hz, 1H, =CH), 6.47 (s, 1H, pyrazole-H), 3.68 (t, J = 5.3 Hz, 4H, CH2NCH2), 3.50 (t, J = 5.2 Hz, 4H, CH2NCH2), 2.64 (d, J = 7.1 Hz, 1H, junction H), 2.10-2.01 (m, 1H, junction-H), 1.02 (d, J = 6.6 Hz, 6H, (CH3)2)LCMS: 315.12 (M+1). MP: 221-222 °C. Yield: 84 %.

General Procedure for the preparation of (6-(4-chloro-2-(trifluoromethyl)phenyl)-2-methylpyrimidin-4-yl)(4-(substituted)piperazin-1-yl)methanone (11a-j) :

Scaffold (9) (0.002 mol), different substituted acids (10a-j) (0.0036 mol), T3P® (0.14 mol), TEA (0.17 mol) were taken in dry EDC, stirred under inert atmosphere at RT for 12 h. Contents were washed with water, 10% NaHCO3, brine and dried over Na2SO4. Crude was purified by column chromatography to get title compounds (11a-j) in good yield. Physical data of all the final compounds were entered in Table 4.1.
Table 4.1: Physical data of final compounds (11a-j)

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<th>Compound</th>
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<th>Yield (%</th>
<th>MP (°C)</th>
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<td>C</td>
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<td>11a</td>
<td>C_{31}H_{32}F_{13}N_{4}O_{2}</td>
<td>55</td>
<td>145</td>
<td>68.12 (68.10)</td>
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<td>C_{32}H_{17}F_{11}N_{4}O_{4}</td>
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<td>178</td>
<td>69.84 (69.83)</td>
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<td>11c</td>
<td>C_{32}H_{39}F_{11}N_{4}O_{4}</td>
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<td>143</td>
<td>64.64 (64.63)</td>
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<td>11d</td>
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<td>167</td>
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<td>11e</td>
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<td>182</td>
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<td>187</td>
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<td>11i</td>
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<td>197</td>
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<td>68</td>
<td>158</td>
<td>72.92 (72.91)</td>
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Spectral interpretation for the final compounds (11a-j)

(4-(2,4-difluorobenzoyl)piperazin-1-yl)(2-(5-(2-fluorophenyl)-3-isobutyl-1H-pyrazol-1-yl)phenyl)methanone (11a):
IR (KBr, cm⁻¹): 2859.92 (CH Stretching), 1639.24 (C=O stretching), 1475.28 (C=N stretching), 1166.72 (CF bending) (Fig 4.10); ¹H NMR (399.6 MHz, CDCl₃) δ ppm: 7.92 (d, J = 1.9 Hz, 1H, Ar-H), 7.65-7.33 (m, 9H, Ar-H), 6.95 (d, J = 6.9 Hz, 1H, Ar-H), 6.46 (s, 1H, pyrazole-H), 3.66 (t, J = 4.8 Hz, 4H, CH₂NCH₂), 3.17 (t, J = 5.2 Hz, 4H, CH₂NCH₂), 2.64 (d, J = 7.1 Hz, 2H, CH₂), 2.20-2.09 (m, 1H, junction H), 1.02 (d, J = 6.6 Hz, 6H, (CH₃)₂) (Fig 4.11); ¹³C NMR (100 MHz, CDCl₃) δ ppm: 167.77, 165.81 (C=O), 165.41 (C=O), 160.49, 136.11, 135.36, 132.51, 132.159 (2C), 130.11 (2C), 129.80 (2C), 129.48, 127.16 (2C), 127.10, 127.05 (2C), 127.00, 124.28, 121.55, 116.76, 46.80, 46.25, 45.75, 42.04 (2C), 26.08 (junction C), 25.71 (2C) (Fig 4.12). CHN analysis complies with the calculated percentage (Fig 4.13).

(4-(2,4-dimethoxy-6-methylbenzoyl)piperazin-1-yl)(2-(5-(2-fluorophenyl)-3-isobutyl-1H-pyrazol-1-yl)phenyl)methanone (11b):
IR (KBr, cm⁻¹): 2866.67 (CH Stretching), 1682.59 (C=O stretching), 1453.10 (C=N stretching), 1266.04 (CF bending) (Fig 4.14); ¹H NMR (399.6 MHz, CDCl₃) δ ppm: 7.99 (d, J = 1.9 Hz, 1H, Ar-H), 7.63-7.38 (m, 8H, Ar-H), 6.98 (d, J = 8.7 Hz, 1H, Ar-H), 6.47 (s, 1H, pyrazole-H), 3.87 (s, 3H, OCH₃), 3.80 (s, 3H, OCH₃), 3.66 (t, J = 4.8 Hz, 4H, CH₂NCH₂), 3.17 (t, J = 5.2 Hz, 4H, CH₂NCH₂), 2.77 (s, 3H, CH₃), 2.63 (d, J = 6.9 Hz, 2H, CH₂), 2.10-2.06 (m, 1H, junction H), 1.02 (d, J = 6.2 Hz, 6H, (CH₃)₂) (Fig 4.15); ¹³C NMR (100 MHz, CDCl₃) δ ppm: 167.79, 165.68 (C=O), 165.40 (C=O), 160.62, 153.07, 150.88, 136.07, 135.36, 132.51, 132.14, 130.11, 129.76, 129.45, 127.07, 127.01, 126.46, 124.28, 121.54, 120.55, 116.59, 116.02, 114.01 (2C), 56.59, 55.96, 47.04 (2C), 46.05 (2C), 45.60, 42.29, 25.06, 16.71 (2C) (Fig 4.16). CHN analysis complies with the calculated percentage (Fig 4.17).
Fig 4.10: IR spectrum of compound 11a
Fig 4.11: $^1$H NMR spectrum of compound 11a
Fig 4.12: $^{13}$C NMR spectrum of compound 11a
Fig 4.13: CHN Analysis of compound 11a

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Fig 4.14: IR spectrum of compound 11b
Fig 4.15: $^1$H NMR spectrum of compound 11b
Fig 4.16: $^{13}$C NMR spectrum of compound 11b
Fig 4.17: CHN Analysis of compound 11b

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(4-(2-(5-(2-fluorophenyl)-3-isobutyl-1H-pyrazol-1-yl)benzoyl)piperazin-1-yl)(3-(trifluoromethoxy)phenyl)methanone (11c):

IR (KBr, cm⁻¹): 2974.12 (CH Stretching), 1646.18 (C=O stretching), 1474.12 (C=N stretching), 1165.22 (CF₃ bending); ¹H NMR (399.6 MHz, CDCl₃) δ ppm: 8.06 (d, J = 7.4 Hz, 1H, Ar-H), 7.46-7.02 (m, 10H, Ar-H), 6.95 (d, J = 7.6 Hz, 1H, Ar-H), 6.47 (s, 1H, pyrazole-H), 3.65 (t, J = 9.1 Hz, 4H, CH₂NCH₂), 3.42 (t, J = 9.2 Hz, 4H, CH₂NCH₂), 2.64 (d, J = 7.11 Hz, 2H, -CH₂-), 2.10-2.01 (m, 1H, junction H), 1.02 (d, J = 6.61 Hz, 6H, (CH₃)₂); ¹³C NMR (100 MHz, CDCl₃) δ ppm: 167.78, 165.61 (C=O), 165.31 (C=O), 160.26, 153.11, 149.98, 136.12, 135.26, 133.12, 132.15, 132.10, 130.11, 129.71 (CF₃), 129.61, 127.07, 127.02, 126.64, 124.28, 121.45, 120.55, 116.56, 116.01, 113.1(2C), 53.49, 46.52, 45.82, 45.53, 45.31, 26.11 (junction C), 16.72 (2C); LC-MS: (m/z) Calculated (Found) 594.60 (595.80) (M+1) complies.

2-(benzo[d][1,3]dioxol-4-yl)-1-(4-(2-(5-(2-fluorophenyl)-3-isobutyl-1H-pyrazol-1-yl)benzoyl)piperazin-1-yl)ethanone (11d):

IR (KBr, cm⁻¹): 2910.12 (CH Stretching), 1641.71 (C=O stretching), 1447.61 (C=N stretching), 1112.11 (CF bending); ¹H NMR (399.6 MHz, CDCl₃) δ ppm: 8.06 (d, J = 7.4 Hz, 1H, Ar-H), 7.48-7.10 (m, 9H, Ar-H), 6.94 (d, J = 7.6 Hz, 1H, Ar-H), 6.48 (s, 1H, pyrazole-H), 5.91 (s, 2H, OCH₂O), 3.65 (t, J = 9.1 Hz, 4H, CH₂NCH₂), 3.44 (t, J = 9.2 Hz, 4H, CH₂NCH₂), 2.64 (d, J = 7.1 Hz, 2H, -CH₂-), 2.10-2.01 (m, 1H, junction H), 1.02 (d, J = 6.6 Hz, 6H, (CH₃)₂); ¹³C NMR (100 MHz, CDCl₃) δ ppm: 167.76, 165.60 (C=O), 165.52 (C=O), 160.12, 154.11, 149.98, 136.12, 135.26, 133.21, 132.15, 132.08, 130.10, 129.71 (CF), 129.60, 128.06, 127.01, 126.63, 124.82, 121.54, 120.51, 116.65, 116.01, 113.12, 101.15 (OCO), 53.49, 46.52, 45.82, 45.53, 45.30, 26.11 (junction C), 16.71 (2C).

(4-(2,4-dimethoxybenzoyl)piperazin-1-yl)(2-(5-(2-fluorophenyl)-3-isobutyl-1H-pyrazol-1-yl)phenyl)methanone (11e):

IR (KBr, cm⁻¹): 2832.91 (CH Stretching), 1623.54 (C=O stretching), 1412.64 (C=N stretching), 1156.91 (CF bending); ¹H NMR (399.6 MHz, CDCl₃) δ ppm: 7.78 (d, J = 1.9 Hz, 1H, Ar-H), 7.41-7.01 (m, 9H, Ar-H), 6.91 (d, J = 7.6 Hz, 1H, Ar-H), 6.47 (s, 1H, pyrazole-H), 3.89 (s, 3H, OCH₃), 3.84 (s, 3H, OCH₃), 3.65 (t, J = 9.9 Hz, 4H, CH₂NCH₂), 3.42 (t, J = 9.8 Hz, 4H, CH₂NCH₂), 2.64 (d, J = 7.11 Hz, 2H, -CH₂-), 2.10-2.06 (m, 1H, junction H), 1.02 (d, J = 6.51 Hz, 6H, (CH₃)₂); ¹³C NMR (100 MHz, CDCl₃) δ ppm: 167.79, 165.61 (C=O), 165.30 (C=O), 160.59, 153.05, 150.86, 136.06, 135.24, 132.15, 132.41, 129.25, 129.41, 126.01, 125.81, 122.12,
121.52, 120.45, 116.02, 115.12, 114.14, 112.12, 110.12, 109.14, 53.12, 46.25, 46.14, 45.55, 45.50, 26.11 (junction C), 16.17 (2C); LC-MS: (m/z) Calculated (Found) 570.65 (571.85) (M+1) complies.

(4-(2-chlorobenzoyl)piperazin-1-yl)(2-(5-(2-fluorophenyl)-3-isobutyl-1H-pyrazol-1-yl)phenyl)methanone (11f):
IR (KBr, cm⁻¹): 2792.19 (CH Stretching), 1671.61 (C=O stretching), 1412.46 (C=N stretching), 1165.19 (CF bending); ¹H NMR (399.6 MHz, CDCl₃) δ ppm: 7.81 (d, J = 1.8 Hz, 1H, Ar-H), 7.62-7.12 (m, 10H, Ar-H), 6.92 (d, J = 7.7 Hz, 1H, Ar-H), 6.49 (s, 1H, pyrazole-H), 3.61 (t, J = 9.1 Hz, 4H, CH₂NCH₂), 3.42 (t, J = 9.6 Hz, 4H, CH₂NCH₂), 2.65 (d, J = 7.2 Hz, 2H, -CH₂), 2.10-2.08 (m, 1H, junction H), 1.03 (d, J = 6.6 Hz, 6H, (CH₃)₂); ¹³C NMR (100 MHz, CDCl₃) δ ppm: 167.12, 165.59 (C=O), 165.40 (C=O), 164.01, 153.02, 150.85, 136.05, 135.42, 131.14, 129.25, 129.14, 126.10, 125.18, 124.64, 123.23, 122.12, 121.52, 116.01, 115.21, 114.41, 112.21, 110.21, 52.92, 46.44, 46.42, 45.24, 45.12, 26.10 (junction C), 16.11 (2C).

(4-(2-fluorobenzoyl)piperazin-1-yl)(2-(5-(2-fluorophenyl)-3-isobutyl-1H-pyrazol-1-yl)phenyl)methanone (11g):
IR (KBr, cm⁻¹): 2816.19 (C_H Stretching), 1693.14 (C=O stretching), 1462.12 (C=N stretching), 1163.91 (CF bending); ¹H NMR (399.6 MHz, CDCl₃) δ ppm: 7.91 (d, J = 1.7 Hz, 1H, Ar-H), 7.71-7.22 (m, 10H, Ar-H), 6.94 (d, J = 7.6 Hz, 1H, Ar-H), 6.48 (s, 1H, pyrazole-H), 3.65 (t, J = 8.7 Hz, 4H, CH₂NCH₂), 3.46 (t, J = 9.1 Hz, 4H, CH₂NCH₂), 2.63 (d, J = 7.2 Hz, 2H, -CH₂), 2.11-2.09 (m, 1H, junction H), 1.04 (d, J = 6.6 Hz, 6H, (CH₃)₂); ¹³C NMR (100 MHz, CDCl₃) δ ppm: 167.51, 165.64 (C=O), 165.22 (C=O), 160.21, 153.12, 150.86, 136.16, 135.42, 132.15, 132.14, 129.24, 129.12, 126.01 (CF), 125.18, 122.21, 121.25, 120.42, 115.02, 114.12, 114.01, 112.12, 110.12, 109.21, 53.12, 46.52, 46.41, 45.12, 45.10, 26.01 (junction C), 16.20 (2C).

(4-(4-fluorobenzoyl)piperazin-1-yl)(2-(5-(2-fluorophenyl)-3-isobutyl-1H-pyrazol-1-yl)phenyl)methanone (11h):
IR (KBr, cm⁻¹): 2891.61 (CH Stretching), 1667.12 (C=O stretching), 1422.68 (C=N stretching), 1165.19 (CF bending); ¹H NMR (399.6 MHz, CDCl₃) δ ppm: 7.94 (d, J = 1.6 Hz, 1H, Ar-H), 7.78-7.26 (m, 10H, Ar-H), 6.96 (d, J = 7.9 Hz, 1H, Ar-H), 6.46 (s, 1H, pyrazole-H), 3.61 (t, J = 8.9 Hz, 4H, CH₂NCH₂), 3.48 (t, J = 9.1 Hz, 4H, CH₂NCH₂), 2.63 (d, J = 7.2 Hz, 2H, -CH₂), 2.12-2.01 (m, 1H, junction H), 1.02 (d, J = 6.2 Hz, 6H, (CH₃)₂); ¹³C NMR (100 MHz, CDCl₃) δ ppm: 167.91, 165.46
(C=O), 165.12 (C=O), 160.21, 153.21, 150.82, 136.16, 134.24, 132.51, 132.41, 129.24, 129.12, 126.02, 125.08, 122.12, 121.52, 120.24, 116.02, 114.21, 113.02, 112.42, 110.21, 109.11, 53.12, 46.12, 46.44, 45.61, 45.10, 26.02 (junction C), 16.12 (2C).

2-(4-dichlorophenyl)-1-(4-(2-(5-(2-fluorophenyl)-3-isobutyl-1H-pyrazol-1-yl)benzoyl)piperazin-1-yl)ethanone (11i): IR (KBr, cm⁻¹): 2791.12 (C-H stretching), 1676.21 (C=O stretching), 1462.12 (C=N stretching), 1191.16 (CF bending); ¹H NMR (399.6 MHz, CDCl₃) δ ppm: 7.92 (d, J = 1.7 Hz, 1H, Ar-H), 7.82-7.22 (m, 9H, Ar-H), 6.91 (d, J = 6.8 Hz, 1H, Ar-H), 6.44 (s, 1H, pyrazole-H), 3.82 (s, 2H, CH₂), 3.62 (t, J = 8.2 Hz, 4H, CH₂NCH₂), 3.46 (t, J = 8.9 Hz, 4H, CH₂NCH₂), 2.61 (d, J = 7.1 Hz, 2H, -CH₂-), 2.10-2.01 (m, 1H, junction H), 1.02 (d, J = 6.1 Hz, 6H, (CH₃)₂); ¹³C NMR (100 MHz, CDCl₃) δ ppm: 167.66, 165.64 (C=O), 165.10 (C=O), 160.21, 153.21, 150.80, 140.14, 136.61, 135.44, 132.15, 132.10, 128.42, 128.20, 126.20, 125.81, 122.22, 121.10, 120.42, 116.20, 114.44, 113.22, 112.28, 111.04, 109.44, 53.49, 49.22, 46.22, 46.02, 45.22, 45.01, 26.01 (junction C), 16.44 (2C).

2-(4-benzoylpiperazin-1-yl)(2-(5-(2-fluorophenyl)-3-isobutyl-1H-pyrazol-1-yl)phenyl)methanone (11j): IR (KBr, cm⁻¹): 2796.14 (CH stretching), 1667.16 (C=O stretching), 1491.21 (C=N stretching), 1161.12 (CF bending); ¹H NMR (399.6 MHz, CDCl₃) δ ppm: 7.81 (d, J = 1.7 Hz, 1H, Ar-H), 7.70-7.20 (m, 10H, Ar-H), 6.92 (d, J = 7.1 Hz, 1H, Ar-H), 6.44 (s, 1H, pyrazole-H), 3.61 (t, J = 8.8 Hz, 4H, CH₂NCH₂), 3.42 (t, J = 9.2 Hz, 4H, CH₂NCH₂), 2.61 (d, J = 6.4 Hz, 2H, -CH₂-), 2.10-2.02 (m, 1H, junction H), 1.01 (d, J = 6.4 Hz, 6H, (CH₃)₂); ¹³C NMR (100 MHz, CDCl₃) δ ppm: 167.45, 165.21 (C=O), 165.10 (C=O), 160.44, 153.44, 150.28, 136.61, 134.42, 132.15, 132.01, 129.42, 129.22, 126.01, 125.06, 122.10, 121.25, 120.24, 116.01, 114.12, 113.14, 112.24, 110.21, 109.22, 53.12, 46.34, 46.24, 45.22, 45.12, 26.02, (junction C), 16.01(2C).
Due to the extensive biological importance of pyridine containing pyrazole derivatives, we have synthesized a series of pyridine containing pyrazoline derivatives.

Reagents and conditions: a) NaH, THF, -15 °C, b) EtOH, 85 °C, c) TEA, T₃P®, EDC, 60 °C d) TFA, EDC, e) TEA, T₃P®, EDC, 60 °C

Scheme 4.2

Procedure for the synthesis of 1-(6-chloropyridin-3-yl)-4,4,4-trifluorobutane-1,3-dione (3).

NaH (0.015 mol) was taken in dry THF (15 ml) to this added Trifluoroacetone (0.010 mol) taken in 4 mL dry THF at -15 °C, stirred the reaction mixture for 1 h, then added ethyl-6-chloronicotinate (0.01 mol) and stirred the reaction mixture for 18 h at 35 °C. Reaction was monitored by TLC, after completion of the reaction, solvent was removed and reaction mixture was quenched with saturated solution of
\(\text{NH}_4\text{Cl (40 ml). The reaction mixture was extracted with ethyl acetate (80 ml), washed with water followed by brine and concentrated under reduced pressure. The crude mass was purified by column chromatography with neutral alumina by using pet ether ethyl acetate (7:3) as eluent to get compound (3) as a yellow solid. Yield: 52%.

\text{IR (KBr, cm}^{-1}): 3091–2957 (aromatic C-H stretching), 1314–1221 (CF\textsubscript{3} stretching), 1618 and 1431(C=C stretching).

\text{\textsuperscript{1}H NMR (CDCl\textsubscript{3}) \delta ppm}: 8.47 (s, 1H, Ar-H), 7.70 (d, \(J = 7.99\) Hz, 1H, Ar-H), 7.45 (d, \(J = 8.39\) Hz, 1H, Ar-H), 5.31 (s, 2H, CH\textsubscript{2}).

Procedure for the synthesis of 2-[5-(6-chloropyridin-3-yl)-3-(trifluoromethyl)-1H-pyrazol-1-yl]benzoic acid (5).

To a solution of 1-(6-chloropyridin-3-yl)-4,4,4-trifluorobutane-1,3-dione (0.010 mol) in ethanol (25 ml) was added 2-Hydrazinobenzoic acid (0.010 mol) and catalytic amount of acetic acid (2-3 drops) contents were stirred for 8 h at 80 °C. The reaction was monitored by TLC, after completion of the reaction, solvent was removed by vacuum, the crude mass was purified by column chromatography using neutral alumina, by using pet ether/ethyl acetate (6:4) as eluent to obtain pale yellow solid and recrystallized by MDC/MeOH. Yield: 66%; LCMS: m/z: 368.8 (M +1).

HPLC: 97%.

Procedure for the synthesis of tert-butyl 4-(2-[5-(6-chloropyridin-3-yl)-3-(trifluoromethyl)-1H-pyrazol-1-yl]benzoic acid-1-carboxylate (7):

Intermediate 2-[5-(6-chloropyridin-3-yl)-3-(trifluoromethyl)-1H-pyrazol-1-yl]benzoic acid was taken in ethylene dichloride to this added triethylamine, stirred the reaction mixture for 10 min. To the reaction mixture, N-Boc piperazine taken in ethylene dichloride and propylphosphonic anhydride (\(T\textsubscript{3}P\textsuperscript{®}\)) was added. Stirred the reaction mixture for 10 h, completion of reaction was confirmed by TLC, organic layer was quenched by ice cold water. The separated organic layer was washed with brine and dried over anhydrous MgSO\textsubscript{4}. Obtained crude mass was purified by column chromatography using neutral alumina and eluents are pet ether/ethyl acetate (6:4). Purified off white solid was recrystallized by pet ether/ethyl acetate. Yield: 46 %. MS: (m/z) 536.99 (M +1). HPLC: Purity 94%. \text{\textsuperscript{1}H NMR (CDCl\textsubscript{3}) \delta ppm}: 8.37 (s, 1H, Ar-H), 7.68 (d, \(J = 7.99\) Hz, 1H, Ar-H), 7.44 (m, 6H, Ar-H & Pyrrole-H), 3.51 (t, \(J = 10.39\) Hz, 4H, -N (CH\textsubscript{2})\textsubscript{2}), 2.97 (t, \(J = 9.99\) Hz, -N (CH\textsubscript{2})\textsubscript{2}), 1.40 (s, 9H, -C(CH\textsubscript{3})\textsubscript{3}).
Procedure for the synthesis of (2-(5-(6-chloropyridin-3-yl)-3-(trifluoromethyl)-1H-pyrazol-1-yl)phenyl)(piperazin-1-yl)methanone (8):
The compound tert-butyl4-(2-(5-(6-chloropyridin-3-yl)-3-(trifluoromethyl)-1H-pyrazol-1-yl)benzoyl)piperazine-1-carboxylate was dissolved in ethylene dichloride to this added trifluoro acetic acid at 10 °C. Reaction mixture was stirred for 4 h at 60 °C, reaction was monitored by TLC, solvent was removed by vacuum, crude compound was purified by column chromatography using neutral alumina by using pet ether/ethyl acetate (6:4) as eluent to get pink color solid (8). Yield: 58 %. MS: (m/z) 436.85 (M +1). HPLC: Purity 97%. 1H-NMR (CDCl3) δ ppm: 8.94 (s, 1H, -NH), 8.37 (s, 1H, Ar-H), 7.68 (d, J = 7.99 Hz, 1H, Ar-H), 7.44-7.39 (m, 6H, Ar-H & Pyrrole-H), 3.18 (t, J = 4.79 Hz, 4H, -N (CH₂)₂), 3.08 (t, J = 5.19 Hz, -N (CH₂)₂).

General Procedure for the synthesis of pyrazoline derivatives (10a-e).
The key scaffold (8) (0.01 mol) was taken in 100 ml round bottomed flask containing ethylene dichloride, to this added trietylamine (0.03 mol) and the reaction mixture was stirred for 10 min. After 10 min substituted carboxylic acids (0.011 mol) were taken in ethylene dichloride and propylphosphonic anhydride (T₃P®) (0.03 mol) was added, reaction mixture was stirred for 15 h at 40 °C. Completion of reaction was confirmed by TLC, organic layer was quenched by ice cold water, separated organic layer, washed with brine and dried over anhydrous MgSO₄. Obtained crude mass was purified by column chromatography using pet ether/ethyl acetate as an eluent (7:3) to yield 10a-e. Physical data of all the final compounds are entered in Table 4.2.

Spectral interpretation for the final compounds (10a-e).

(4-(2-(5-(6-chloropyridin-3-yl)-3-(trifluoromethyl)-1H-pyrazol-1-yl)benzoyl)piperazin-1-yl)(3-fluoro-4-methylphenyl)methanone(10a).
IR (KBr, cm⁻¹): 1632.45 (C=O), 1322.93 (CF₃ stretching) (Fig 4.18). 1H NMR ; CDCl₃ (ppm): 8.37 (s, 1H, Ar-H), 7.68 (d, J = 7.79 Hz, 1H, Ar-H ), 7.44-7.39 (m, 9H, Ar-H & Pyrazole-H), 3.81 (t, J = 12.78, 4H, CH₂NCH₂), 3.05 (t, J = 6.79, 4H, CH₂NCH₂), 2.37 (s, 3H, Ar-CH₃) (Fig 4.19). 13C NMR (100 MHz, CDCl₃) δ ppm: 166.72, 162.14 (C=O), 159.64 (C=O), 153.12, 148.08, 147.25, 137.88, 137.18, 134.28, 134.21, 132.43, 132.39, 131.58, 131.41, 129.34, 124.20, 123.18, 123.15,
114.64 (2C), 114.39 (2C), 47.21, 45.88, 42.12 (2C), 14.78 (Fig 4.20). CHN analysis complies with the calculated percentage (Fig 4.21).
Fig 4.18: IR spectrum of compound 10a
Fig 4.19: $^1$H NMR spectrum of compound 10a
Fig 4.20: $^{13}$C NMR spectrum of compound 10a
**Fig 4.21: CHN Analysis of compound 10a**

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(4-(2-(5-(6-chloropyridin-3-yl)-3-(trifluoromethyl)-1H-pyrazol-1-yl)benzoyl)piperazin-1-yl)(2,5-dimethoxyphenyl) methane (10b):

IR (KBr, cm\(^{-1}\)) : 1655 (C=O), 1365-1168 (CF\(_3\) stretching) (Fig 4.22); \(^1\)H NMR

CDCl\(_3\) (ppm): 8.41 (d, J = 1.7 Hz, 1H, Ar-H), 7.69 (d, J = 5.7 Hz, 1H, Ar-H), 7.41-7.39 (m, 7H, Ar-H & Pyrazole-H), 7.10 (d, J = 3.1 Hz, 1H, Ar-H), 6.98 (d, J = 9.0 Hz, 1H, Ar-H), 3.87 (s, 3H, -O-CH\(_3\)), 3.80 (s, 3H, -O-CH\(_3\)), 3.52 (t, J = 4.5 Hz, 4H, CH\(_2\)NCH\(_2\)), 3.31 (t, J = 5.1 Hz, 4H, CH\(_2\)NCH\(_2\)) (Fig 4.23); \(^13\)C NMR (100 MHz, CDCl\(_3\)) \(\delta\) ppm: 166.77, 153.17 (2 C=O), 152.99 (2C), 150.90 (2C), 148.05 (2C), 137.90 (2C), 129.66 (2C), 126.42 (2C), 124.43 (2C), 120.65 (2C), 116.00 (2C), 114.26 (2C), 58.80, 53.99, 47.84, 45.90, 42.35 (2C) (Fig 4.24); CHN analysis complies with the calculated percentage (Fig 4.25).

(4-(2-(5-(6-chloropyridin-3-yl)-3-(trifluoromethyl)-1H-pyrazol-1-yl)benzoyl)piperazin-1-yl)(3-fluorophenyl)methanone (10c):

IR (KBr, cm\(^{-1}\)) : 1653 (C=O), 1344 (CF\(_3\) stretching); \(^1\)H NMR

CDCl\(_3\) (ppm): 8.43 (s, 1H, Ar-H); 8.02 (d, J = 7.99 Hz, 1H, Ar-H); 7.73-7.70 (m, 2H, Ar-H), 7.42-7.37 (m, 8H, Ar-H & Pyrazole-H), 3.75-3.56 (m, 4H, -N(CH\(_2\))\(_2\)); 3.42-3.38 (m, 4H, -N(CH\(_2\))\(_2\)); HPLC: 99%.

(4-(2-(5-(6-chloropyridin-3-yl)-3-(trifluoromethyl)-1H-pyrazol-1-yl)benzoyl)piperazin-1-yl)(5-methylisoxazol-4-yl)methanone (10d):

IR (KBr, cm\(^{-1}\)) : 1628-1650 (C=O), 1311-1168 (CF\(_3\) stretching). \(^1\)H NMR

CDCl\(_3\) (ppm): 8.47 (s, 1H, Ar-H); 7.70 (d, J = 2.39 Hz, 1H, Ar-H), 7.45-7.40 (m, 7H, Ar-H, Pyrazole-H & Isoxazole-H), 3.73-3.68 (m, 8H, 2N(CH\(_2\))\(_2\)), 2.51 (s, 3H, Isoxazole-CH\(_3\)). MS: (m/z) (M+1): 545.91. HPLC: Purity 99%.

(4-(2-(5-(6-chloropyridin-3-yl)-3-(trifluoromethyl)-1H-pyrazol-1-yl)benzoyl)piperazin-1-yl)(cyclopropyl)methanone (10e):

IR (KBr, cm\(^{-1}\)) : 1620-1642 (C=O), 1291-1260 (CF\(_3\) stretching). \(^1\)H NMR

CDCl\(_3\) (ppm): 8.46 (s, 1H, Ar-H); 7.70 (d, J = 2.39 Hz, 1H, Ar-H), 7.44-7.38 (m, 6H, Ar-H & Pyrazole-H), 3.73-3.68 (m, 8H, 2N(CH\(_2\))\(_2\)), 2.31-2.28 (m, 1H, Junction-H), 1.22-1.20 (m, 2H, CH\(_2\)), 1.18-0.99 (m, 2H, CH\(_2\)). MS: m/z: (M+1): 504.94. HPLC: Purity 96%.
Fig 4.22: IR spectrum of compound 10b
Fig 4.23: $^1$H NMR spectrum of compound 10b
Fig 4.24: $^{13}$C NMR spectrum of compound 10b
Fig 4.25: CHN Analysis of compound 10b

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Table 4.2: Physical data of final molecules (10a-e)

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<td>C_{27}H_{20}ClF_{4}N_{5}O_{2}</td>
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<td>178</td>
<td>58.12</td>
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<td>(58.11)</td>
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<td>C_{29}H_{25}ClF_{3}N_{5}O_{4}</td>
<td>57</td>
<td>143</td>
<td>58.05</td>
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<td>(58.01)</td>
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<td>167</td>
<td>55.10</td>
</tr>
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<td>(55.04)</td>
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<tr>
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<td>C_{24}H_{21}ClF_{3}N_{5}O_{2}</td>
<td>52</td>
<td>182</td>
<td>57.20</td>
</tr>
<tr>
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<td>(57.15)</td>
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</table>

4.4. Results and Discussion

In view of literature survey on pyrazole properties and preparation of amides of pyrazoline derivatives, compound (6) was prepared by the condensation and cyclisation reaction by reacting (4) with (5) in the presence of catalytic amount of AcOH. Then intermediate (6) was made to react with compound (7) in presence of TEA, T_{3}P® to give compound (8) which was then treated to TFA for the deprotection of boc group to get key scaffold (9) which was made to react with carboxylic acids (10a-j) in presence of TEA, T_{3}P® in ethylenedichloride to get the new compounds of amides containing pyridine pyrazoline nucleus (11a-j).

In IR spectrum of compound 11a, CH stretching, C=O stretching, CN Stretching and CF_{3} bending frequencies appears in the range 2859.92, 1639.24, 1475.38 and 1166.72 cm\(^{-1}\) respectively.
In $^1$H NMR spectrum of compound 11a, protons of the difluorobenzene, 2-fluorobenzene and benzene appears in the range 7.78-6.91 ppm. Pyrazole one proton appears at 6.46 as singlet. Piperazine protons appears in the range 3.65-3.44 (t, 8H) and methylene two protons appears at 2.64 (d, 3H), junction proton appears at 2.10-2.01 (m, 1H). Two methyl six protons appear at 1.02 (d, 6H).

In $^{13}$C NMR spectrum of compound 11a, carbons of difluorobenzene, 2-fluorobenzene and benzene appears in the range 136.21-113.12 ppm and ipso carbons C$_4$, C$_7$, C$_2$ and C$_{33}$ appears at 167.91, 161.44, 153.22 and 149.99 ppm respectively. The carbonyl (C=O) carbons appears at 165.71 and 165.14 ppm and aliphatic carbon of piperazine ring C$_{26}$, C$_{27}$, C$_{29}$ and C$_{30}$ appears at 46.82-45.21 ppm. Methylene carbon to pyrazole ring appears at 53.49 ppm. Junction carbon appears at 26.01 and two methyl carbons appear at 26.60. All these evidences complies the assigned structure for the compound. Similarly all the newly synthesized compounds from Scheme 4.1 and Scheme 4.2 were also characterized.

4.5. Conclusion

The research work was focused on the efficient synthesis of amides containing 3,5-disubstituted pyrazoline derivatives containing piperazine nucleus. The reactions performed are ecofriendly as they are carried out at moderate temperature 50-60 °C by using solvents ethanol and ethylene dichloride to get final compounds with good yield, short time interval and high purity. Newly synthesized compounds were well characterized by IR, $^1$H & $^{13}$C NMR, LCMS, HPLC and CHN analysis. The obtained compounds were evaluated for their biological activities like antibacterial, antifungal, anthelmintic, anti-inflammatory and anticancer activities. The details of the biological screening results have been presented in Chapter 6 of the thesis.
4.6. References