3.1. Introduction

Heterocyclic ring systems have emerged as powerful scaffolds for many biological evaluations [1]. Heterocyclic compounds provide scaffolds on which pharmacophores can arrange to yield potent and selective drugs [2]. Benzofuran is an important class of heterocyclic compounds that are known to possess important biological properties. Benzofurans belong to one of the most studied structural units in both the synthetic and medicinal chemistry community. They are widely embedded in many biologically interesting natural products and synthetic analogues [3, 4]. Specifically, several benzofuran ring systems bearing various substituents at C-2 position. Benzofurans are omnipresent in nature, the stilbenoids structural compounds, e.g. an oligostilbene derivative viniferin are known to possess antimicrobial, antiviral, antioxidant, antifungal and antitumor activities [5, 6]. Among this large family, benzofuranols represent a fairly small family featured with the furan-2,3 double bond shifting from inside to outside of the furan fragment and with a C-2-hemiacetal function (Figure 3.1.1) [7]. This series of compounds were barely studied due to the limited source and very few available synthetic protocols.

![Figure 3.1.1: Mixture of 3, 3-aryl-benzofuran-2ones](image)

The, Irganox®HP-136 (a mixture of new 3-aryl-benzofuran-2ones) (Figure 3.1.2) has been used as an excellent stabilizer during polymer processing at high temperatures under oxygen deficient extrusion conditions. Systems based on benzofuranones provide equivalent melt processing performances at significantly reduced concentrations compared to those based on combinations of phenols and phosphites alone [8]. Their stabilizing effect is explained by the formation of stable benzofuranonyl radicals, which can react with alkyl radicals terminating chain reactions [9, 10]. Unfortunately, very little is known on the formal kinetics of oxidation of polyolefins in the presence of lactones at elevated temperatures.
In addition, phenyl (2-phenylbenzofuran-3-yl)methanone derivatives had been demonstrated to possess neuroprotective and antitumor activities [11]. On the other hand, pyrazole scaffold represents a common motif in many pharmaceutical active and remarkable compounds (Figure 3.1.3) demonstrating a wide range of pharmacological activities; the most important activities are the anti-inflammatory, antibacterial, antifungal, hypoglycemic and anti-hyperlipidemic [12, 13]. Heterocyclic rings and in particular, the pyrazole ring represent an advantageous choice for the synthesis of pharmaceutical compounds with different activities and good safety profiles [14]. These privileged pharmacophore containing molecules exhibit therapeutical properties over wide range of targets [15]. Owing to their prevalence in natural products as well as pharmaceuticals have stimulated significant interest in the synthesis of benzofuran containing heterocycles.

The literature survey reveals that the synthesis of amide linked pyrazole gathered with benzofuran moiety has not been established, which promotes us to synthesis of above moieties and to study their biological efficacy. In continuation of our research in the synthesis and biological evaluation of novel heterocyclic analogues [16, 17], the present study aimed at gathering the two bioactive entities like benzofuran with pyrazole amides in one compact structure for the purpose of increasing the antioxidant potentials.
3.2. Experimental

3.2.1. Instruments and materials

All reagents and solvents were purchased from Merck (Darmstadt, Germany) chemical AR grade and were used as provided. DPPH and BHA were purchased from Sigma-Aldrich chemical Co. (St. Louis, MO, USA). TLC analysis was performed on alumina sheets precoated with silica gel 60F-254 and SiO₂, 200-400 mesh (Merck) was used for column chromatography. ¹H NMR (400 MHz) and ¹³C NMR (100 MHz) spectra were obtained in AC Bruker spectrometer with the appropriate (DMSO-d₆) solvent. Melting points were recorded on a reichert thermopan melting point apparatus, equipped with a microscope and are uncorrected. Mass spectra were obtained by Water-Q-TOF ultima spectrometer. Micro analytical data were obtained by elemental-Vario EL-III.

3.2.2. Synthetic procedure

3.2.2.1. Synthesis of 2-acetyl benzofuran (2).

A mixture of salicylaldehyde (1) (2 mmol), chloroacetone (2 mmol) and 1,8-diaza bicyclo[5.4.0]undec-7-ene (DBU) (2 mmol) in 10 mL of dry dichloromethane (DCM) containing molecular sieves was stirred for 6 hrs at room temperature. Progress of the reaction was monitored by TLC using hexane: ethyl acetate (8:2) mixture as mobile phase. After the completion of the reaction, the reaction mixture was washed with 10% HCl solution followed by water. The organics were dried over anhydrous sodium sulfate. The yellow solid was obtained by desolventized in a rotary evaporator at room temperature affords 2-acetyl benzofuran (2).

3.2.2.2. 4-hydroxy-3-methoxyphenyl) prop-2-en-1-one (22).

Compound (2) (1 mmol) in DCM (5 mL) was treated with ZrCl₄ (46.6 mg, 20 mol%) followed by addition vanillin (1 mmol). The solution stirred at room temperature under an air atmosphere for 1 hr. After the completion of the reaction monitored by TLC, the crude mixture was worked up in ice cold brine solution and then extracted with ethyl acetate solution (3×10 mL). The combined
ethyl acetate extract was dried over anhydrous Na$_2$SO$_4$ and concentrated in vacuo, and the resulting product was purified by column chromatography using ethyl acetate/n-hexane as mobile phase (7:3) to afford the pure product (22) as a dark yellow solid. The product was recrystallized by methanol.

3.2.2.3. **Synthesis of 4-(3-(benzofuran-2-yl)-4,5-dihydro-1H-pyrazol-5-yl)-2-methoxyphenol (23).**

A mixture of benzofuran chalcone (22) (1 mmol) and hydrazine hydrate 98% (3 mmol) in absolute ethanol was refluxed for 3 hrs. Upon cooling, white solid was obtained, filtered off and recrystallized from methanol to obtain the compound (23).

3.2.2.4. **Synthesis of 3-(benzofuran-2-yl)-5-(4-hydroxy-3-methoxyphenyl)-4,5-dihydro-1H-pyrazole-1-carbonyl chloride (24).**

To the well stirred solution of compound (23) (2.5 mmol) in dry tetrahydrofuran (THF), phosgene (3 mmol) in 5 mL of dry THF was added drop by drop for about 15 min. Then the reaction mixture was stirred at room temperature for about 4 hrs. Progress of the reaction was monitored by TLC using hexane: ethyl acetate mixture (6:4) as mobile phase. After the completion of reaction, the reaction mass was quenched in an ice cold water (about 25 mL) and extracted in diethyl ether. The ether layer was washed twice with 5% NaHCO$_3$ and twice with distilled water. Finally, the ether layer was dried over anhydrous Na$_2$SO$_4$. The brown colored solid product was obtained by desolventation through rotary evaporator at 30 °C. The obtained product was recrystallized by using dichloromethane (DCM).

3.2.2.5. **General procedure for the synthesis of benzofuran based 1,3,5-substituted pyrazole derivatives (25-36).**

Substituted anilines (1.2 mmol) in DCM (5 mL) were treated with anh. K$_2$CO$_3$ (600 mg) under N$_2$ atmosphere. Later the solution of 2-(3-(benzofuran-2-yl)-5-(4-hydroxy-3-methoxyphenyl)-4,5-dihydro-1H-pyrazol-1-yl)acetyl chloride (24) (1 mmol) in DCM (5 mL) was added drop by drop for 15 min. The reaction
mixture was refluxed for 3-5 hrs. The progress of the reaction mixture was monitored by TLC using hexane: ethyl acetate: mixture (6:4). The reaction mixture was then desolventized in rotary evaporator and the compound is extracted in ethyl acetate. The ethyl acetate layer was washed with water and dried over anhydrous Na₂SO₄. The final product was obtained by further desolventation in rotary evaporator at 50 °C. The respective products were purified through column chromatography using hexane: ethyl acetate mixture (60:40).

Scheme 3.2.1: Synthesis of 2-acetyl benzofuran (2)
Scheme 3.2.2: Reaction pathway for the synthesis of benzofuran based 1,3,5-substituted pyrazole derivatives (25-36)

(Published: Bioorganic & Medicinal Chemistry Letters. 22 (2012) 4773–4777)
Table 3.2.1: Chemical structure and yield of synthesized compounds (25-36)

<table>
<thead>
<tr>
<th>Compound No</th>
<th>Entry R</th>
<th>Yield %</th>
<th>Compound No</th>
<th>Entry R</th>
<th>Yield %</th>
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<td>83.4</td>
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<td>-------------------</td>
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<tr>
<td><strong>IUPAC Name</strong></td>
<td>(E)-1-(benzofuran-2-yl)-3-(4-hydroxy-3-methoxyphenyl)prop-2-en-1-one</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
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<tr>
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<td></td>
</tr>
<tr>
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<tr>
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<td>(DMSO-d_6 400 MHz) δ ppm: 6.70-7.89 (m, 8H, Ar-H), 7.68 (s, 1H, CH), 6.23 (s, 1H, CH), 5.21 (s, 1H, OH), 3.80 (s, 3H, OCH_3).</td>
<td></td>
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<td></td>
<td></td>
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<tr>
<td><strong>\text{\textsuperscript{13}}C NMR</strong></td>
<td>(DMSO-d_6 100 MHz) δ ppm: 177.8, 160.4, 155.2, 149.1, 147.8, 145.0, 127.8, 127.5, 124.7, 123.3, 122.9, 121.2, 120.9, 116.7, 111.9, 111.4, 56.1.</td>
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**ELEMENTAL ANALYSIS**

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<th>Calculated</th>
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<td>C, 73.46; H, 4.79%</td>
<td>C, 73.44; H, 4.78%.</td>
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**Compound (23)**

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<td>IUPAC Name</td>
<td>4-(3-(benzofuran-2-yl)-4,5-dihydro-1H-pyrazol-5-yl)-2-methoxyphenol</td>
</tr>
<tr>
<td>Chemical Formula</td>
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</tr>
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<tr>
<td>Physical State</td>
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<td>Melting Point</td>
<td>155-157 °C</td>
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<td>IR ( (\text{KBr})\nu_{\text{max}}(\text{cm}^{-1}) ):</td>
<td>3211 (N-H) and 1595 (C=N).</td>
</tr>
<tr>
<td>Mass ( (\text{ESI})\ m/z ):</td>
<td>308.12 (M⁺)</td>
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<td>(^{1}\text{H NMR} ) ( (\text{DMSO-d}_6\ 400 \text{ MHz})\ \delta \text{ ppm} ):</td>
<td>6.65-7.85 (m, 8H, Ar-H), 7.10 (s, 1H, N-H), 5.23 (s, 1H, OH), 3.90 (d, 2H, CH₂, pyrazoline), 3.84-3.88 (t, 1H, CH, pyrazoline), 3.78 (s, 3H, OCH₃).</td>
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<tr>
<td>(^{13}\text{C NMR} ) ( (\text{DMSO-d}_6\ 100 \text{ MHz})\ \delta \text{ ppm} ):</td>
<td>155.6, 155.0, 147.2, 146.7, 137.0, 134.5, 125.1, 124.7, 123.3, 120.9, 119.4, 115.4, 115.5, 110.2, 104.9, 56.1, 49.0, 40.9.</td>
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**ELEMENTAL ANALYSIS**

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<td>C</td>
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<tr>
<td>H</td>
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<td>H, 5.25%</td>
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<td>N</td>
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<td>N, 9.11%</td>
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### Compound (24)

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<td>IUPAC Name</td>
<td>3-(benzofuran-2-yl)-5-(4-hydroxy-3-methoxyphenyl)-4,5-dihydro-1H-pyrazole-1-carbonyl chloride</td>
</tr>
<tr>
<td>Chemical Formula</td>
<td>C_{19}H_{15}ClN_{2}O_{4}</td>
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<tr>
<td>Molecular Weight</td>
<td>370.786</td>
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<td>Physical State</td>
<td>Brown colored solid</td>
</tr>
<tr>
<td>Melting Point</td>
<td>162-165 °C</td>
</tr>
<tr>
<td>IR</td>
<td>(KBr)\nu_{\text{max}}(\text{cm}^{-1}) : 3211 (N-H) and 1595 (C=N).</td>
</tr>
<tr>
<td>Mass</td>
<td>(ESI) m/z: 370.79 (M+)</td>
</tr>
<tr>
<td>$^1$H NMR</td>
<td>(DMSO-$d_6$ 400 MHz) $\delta$ ppm: 6.65–7.85 (m, 8H, Ar-H), 7.10 (s, 1H, NH), 5.23 (s, 1H, OH), 3.91 (d, 2H, CH$_2$, pyrazoline), 3.83–3.88 (t, 1H, CH, pyrazoline), 3.75 (s, 3H, OCH$_3$).</td>
</tr>
<tr>
<td>$^{13}$C NMR</td>
<td>(DMSO-$d_6$ 100 MHz) $\delta$ ppm: 155.5, 155.1, 147.3, 146.6, 134.5, 125.0, 124.7, 123.3, 120.8, 119.3, 115.4, 111.5, 110.3, 104.9, 65.6, 56.0, 37.2.</td>
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<tr>
<td>ELEMENTAL ANALYSIS Calculated</td>
<td>C, 61.55; H, 4.08; N, 7.56%</td>
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<td>Found</td>
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# Compound (25)

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<td><strong>IUPAC Name</strong></td>
<td>3-(benzofuran-2-yl)-5-(4-hydroxy-3-methoxyphenyl)-N-phenyl-4,5-dihydro-1H-pyrazole-1-carboxamide</td>
</tr>
<tr>
<td><strong>Chemical Formula</strong></td>
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<td><strong>Molecular Weight</strong></td>
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<td><strong>Physical State</strong></td>
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</tr>
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<td><strong>Melting Point</strong></td>
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<td><strong>IR</strong></td>
<td>(KBr)ν_{max}(cm^{-1}): 3131-2973 (Ar-CH), 1680 (C=O), 1624 (C=N pyrazole), 1365 (C-N).</td>
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<tr>
<td><strong>Mass</strong></td>
<td>(ESI) m/z: 427.15 (M^{+})</td>
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<tr>
<td><strong>{^1}H NMR</strong></td>
<td>(DMSO-\textit{d}_6 400 MHz) δ ppm: 6.78-8.01 (m, 13H, Ar-H), 6.00 (s, 1H, NH), 5.35 (s, 1H, OH), 4.80-4.83 (t, 1H, CH of pyrazole), 3.95 (d, 2H, CH\textsubscript{2} of pyrazole), 3.75 (s, 3H, OCH\textsubscript{3}).</td>
</tr>
<tr>
<td><strong>{^{13}}C NMR</strong></td>
<td>(DMSO-\textit{d}_6 100 MHz) δ ppm: 155.7, 155.0, 153.0, 147.2, 146.8, 139.4, 134.8, 134.5, 128.8, 128.1, 125.2, 124.7, 123.4, 121.5, 120.9, 119.3, 115.2, 111.4, 110.3, 104.8, 66.2, 56.0, 37.4.</td>
</tr>
<tr>
<td><strong>ELEMENTAL ANALYSIS</strong></td>
<td>Calculated: C, 70.25; H, 4.95; N, 9.83%</td>
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### Compound (26)

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<td><strong>IUPAC Name</strong></td>
<td>3-(benzofuran-2-yl)-5-(4-hydroxy-3-methoxyphenyl)-N-(4-hydroxyphenyl)-4,5-dihydro-1H-pyrazole-1-carboxamide</td>
</tr>
<tr>
<td><strong>Chemical Formula</strong></td>
<td>C_{25}H_{21}N_{3}O_{5}</td>
</tr>
<tr>
<td><strong>Molecular Weight</strong></td>
<td>443.451</td>
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<tr>
<td><strong>Physical State</strong></td>
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<tr>
<td><strong>IR</strong></td>
<td>(KBr)ν_{max}(cm^{-1}): 3471 (N-H), 3132-2832 (Ar-CH), 1672 (C=O), 1566 (C=N pyrazole), 1307 (C-N).</td>
</tr>
<tr>
<td><strong>Mass</strong></td>
<td>(ESI) m/z: 443.15 (M+)</td>
</tr>
<tr>
<td><strong>{^1}H NMR</strong></td>
<td>(DMSO-d_{6} 400 MHz) δ ppm: 6.92-7.94 (m, 12H, Ar-H), 6.96 (s, 1H, NH), 5.31 (s, 2H, OH), 3.92-3.96 (t, 1H, CH of pyrazole), 3.80 (s, 3H, OCH_{3}), 3.62 (d, 2H, CH_{2} of pyrazole).</td>
</tr>
<tr>
<td><strong>{^{13}}C NMR</strong></td>
<td>(DMSO-d_{6} 100 MHz) δ ppm: 155.7, 155.0, 154.2, 153.0, 147.4, 146.8, 134.6, 134.5, 132.2, 125.3, 124.7, 123.3, 123.0, 120.9, 119.3, 116.1, 115.3, 111.4, 110.4, 104.8, 66.3, 56.0, 37.5.</td>
</tr>
<tr>
<td><strong>ELEMENTAL ANALYSIS</strong></td>
<td><strong>Calculated</strong> C, 67.71; H, 4.77; N, 9.48%</td>
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<td><strong>Found</strong>                      C: 67.70, H: 4.73, N: 9.52%.</td>
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## Physico-chemical and spectral characterization

### Compound (27)

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<td><strong>IUPAC Name</strong></td>
<td>3-(benzofuran-2-yl)-5-(4-hydroxy-3-methoxyphenyl)-N-(4-hydroxyphenyl)-4,5-dihydro-1H-pyrazole-1-carboxamide</td>
</tr>
<tr>
<td><strong>Chemical Formula</strong></td>
<td>C_{25}H_{21}N_{3}O_{5}</td>
</tr>
<tr>
<td><strong>Molecular Weight</strong></td>
<td>443.451</td>
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<tr>
<td><strong>Physical State</strong></td>
<td>Light yellow solid</td>
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<tr>
<td><strong>Melting Point</strong></td>
<td>195-197 °C</td>
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</table>
| **IR**                          | (KBr)\(v_{\text{max}}\) (cm\(^{-1}\)): 3136-2969 (Ar-CH), 1682 (C=O), 1629 (C=N
|                                 | (C=N), 1358 (C-N).                                                    |
| **Mass**                        | (ESI) m/z: 443.15 (M\(^+\))                                        |
| **\(^1\)H NMR**                 | (DMSO-\(d_6\) 400 MHz) \(\delta\) ppm: 6.75-7.81 (m, 12H, Ar-H), 6.20 (s, 1H, NH), 5.33 (s, 2H, OH), 4.85-4.90 (t, 1H, CH of pyrazole), 3.90 (s, 3H, OCH\(_3\)), 3.79 (d, 2H, CH\(_2\) of pyrazole). |
| **\(^{13}\)C NMR**              | (DMSO-\(d_6\) 100 MHz) \(\delta\) ppm: 155.5, 155.0, 154.1, 153.1, 147.6, 146.7, 134.6, 134.5, 132.3, 125.2, 124.8, 123.3, 123.0, 120.9, 119.3, 116.1, 115.3, 111.4, 110.4, 104.8, 66.3, 56.0, 37.5. |
| **ELEMENTAL ANALYSIS**          | **Calculated**                                                       |
|                                 | C, 67.71; H, 4.77; N, 9.48%                                          |
|                                 | **Found**                                                            |
|                                 | C: 67.72, H: 4.75, N: 9.50%.                                         |
### Compound (28)

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<tr>
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<td><strong>Physical State</strong></td>
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<tr>
<td><strong>Mass</strong></td>
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<tr>
<td><strong>^{13}C NMR</strong></td>
<td>(DMSO-d$_6$ 100 MHz) δ ppm: 155.6, 154.9, 153.0, 147.4, 146.7, 144.2, 134.6, 134.5, 129.4, 125.2, 124.8, 123.3, 120.8, 119.2, 117.1, 115.3, 111.3, 110.4, 106.0, 104.8, 66.3, 56.0, 37.6.</td>
</tr>
<tr>
<td><strong>ELEMENTAL ANALYSIS</strong></td>
<td>Calculated</td>
</tr>
<tr>
<td><strong>Found</strong></td>
<td>C, 65.92; H, 4.88; N, 8.85%.</td>
</tr>
</tbody>
</table>

**Calculated**

C, 65.95; H, 4.90; N, 8.87%
## Compound (29)

<table>
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<th>Property</th>
<th>Value</th>
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<tbody>
<tr>
<td><strong>IUPAC Name</strong></td>
<td>3-(benzofuran-2-yl)-5-(4-hydroxy-3-methoxyphenyl)-N-(3-hydroxyphenyl)-4,5-dihydro-1H-pyrazole-1-carboxamide</td>
</tr>
<tr>
<td><strong>Chemical Formula</strong></td>
<td>C_{25}H_{21}N_{3}O_{5}</td>
</tr>
<tr>
<td><strong>Molecular Weight</strong></td>
<td>443.451</td>
</tr>
<tr>
<td><strong>Physical State</strong></td>
<td>Yellow semisolid</td>
</tr>
<tr>
<td><strong>Melting Point</strong></td>
<td>---</td>
</tr>
<tr>
<td><strong>IR</strong></td>
<td>(KBr)$\nu_{\text{max}}$(cm$^{-1}$): 3128-2970 (Ar-CH), 1669 (C=O), 1630 (C=N pyrazole), 1359 (C-N).</td>
</tr>
<tr>
<td><strong>Mass</strong></td>
<td>(ESI) m/z: 443.15 (M$^+$)</td>
</tr>
<tr>
<td><strong>$^1$H NMR</strong></td>
<td>(DMSO-$d_6$ 400 MHz) δ ppm: 6.79-7.65 (m, 12H, Ar-H), 6.00 (s, 1H, NH), 5.33 (s, 2H, OH), 4.86-4.90 (t, 1H, CH of pyrazole), 3.89 (s, 3H, OCH$_3$), 3.70 (d, 2H, CH$_2$ of pyrazole).</td>
</tr>
<tr>
<td><strong>$^{13}$C NMR</strong></td>
<td>(DMSO-$d_6$ 100 MHz) δ ppm: 155.6, 155.0, 154.2, 153.0, 147.4, 146.8, 134.6, 134.5, 132.2, 125.2, 124.6, 123.4, 123.0, 120.8, 119.2, 116.1, 115.2, 111.4, 110.4, 104.8, 66.4, 56.2, 37.6.</td>
</tr>
<tr>
<td><strong>ELEMENTAL ANALYSIS</strong></td>
<td>Calculated: C, 67.71; H, 4.77; N, 9.48%</td>
</tr>
<tr>
<td></td>
<td>Found: C: 67.70, H: 4.73, N: 9.52%.</td>
</tr>
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</table>
Table 3.1: Physico-chemical and spectral characterization of Compound (30)

<table>
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<tr>
<th>Property</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>IUPAC Name</td>
<td>3-(benzofuran-2-yl)-N-(3,4-dihydroxyphenyl)-5-(4-hydroxy-3-methoxyphenyl)-4,5-dihydro-1H-pyrazole-1-carboxamide</td>
</tr>
<tr>
<td>Chemical Formula</td>
<td>C$<em>{25}$H$</em>{21}$N$<em>{3}$O$</em>{6}$</td>
</tr>
<tr>
<td>Molecular Weight</td>
<td>459.450</td>
</tr>
<tr>
<td>Physical State</td>
<td>Light brown semisolid</td>
</tr>
<tr>
<td>Melting Point</td>
<td>221-223 ºC</td>
</tr>
<tr>
<td>IR</td>
<td>(KBr)ν$_{\text{max}}$ (cm$^{-1}$): 3316 (N-H), 3130-2762 (Ar-CH), 1682 (C=O), 1620 (C=N pyrazole), 1378 (C-N).</td>
</tr>
<tr>
<td>Mass</td>
<td>(ESI) m/z: 459.14 (M$^+$)</td>
</tr>
<tr>
<td>$^1$H NMR</td>
<td>(DMSO-$d_6$ 400 MHz) δ ppm: 6.60-7.90 (m, 11H, Ar-H), 7.08 (s, 1H, NH), 5.20 (s, 2H, OH), 4.96 (s, 1H, OH), 4.89 (s, 1H, CH of pyrazole), 3.75 (s, 3H, OCH$_3$), 3.68 (d, 2H, CH$_2$ of pyrazole).</td>
</tr>
<tr>
<td>$^{13}$C NMR</td>
<td>(DMSO-$d_6$ 100 MHz) δ ppm: 155.4, 155.0, 154.1, 153.1, 147.6, 146.7, 134.6, 134.5, 132.2, 125.1, 124.9, 123.3, 123.0, 120.8, 119.3, 116.1, 115.3, 111.4, 110.4, 104.8, 66.3, 56.1, 37.3.</td>
</tr>
<tr>
<td><strong>ELEMENTAL ANALYSIS</strong></td>
<td><strong>Calculated</strong></td>
</tr>
<tr>
<td></td>
<td>C, 65.35; H, 4.61; N, 9.15%</td>
</tr>
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## Compound (31)

<table>
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<th>Value</th>
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<tbody>
<tr>
<td>IUPAC Name</td>
<td>3-((benzofuran-2-yl)-N-(4-bromophenyl)-5-(4-hydroxy-3-methoxyphenyl)-4,5-dihydro-1H-pyrazole-1-carboxamide</td>
</tr>
<tr>
<td>Chemical Formula</td>
<td>C_{25}H_{20}BrN_{3}O_{4}</td>
</tr>
<tr>
<td>Molecular Weight</td>
<td>506.348</td>
</tr>
<tr>
<td>Physical State</td>
<td>Light brown solid</td>
</tr>
<tr>
<td>Melting Point</td>
<td>268-271 °C</td>
</tr>
<tr>
<td>IR</td>
<td>(KBr)ν_{\text{max}}(\text{cm}^{-1}): 3124-2981 (Ar-CH), 1675 (C=O), 1624 (C=N pyrazole), 1356 (C-N).</td>
</tr>
<tr>
<td>Mass</td>
<td>(ESI) m/z: 505.06 (M^+)</td>
</tr>
<tr>
<td>^1H NMR</td>
<td>(DMSO-d_{6} 400 MHz) δ ppm: 6.70-7.85 (m, 12H, Ar-H), 6.15 (s, 1H, NH), 5.30 (s, 1H, OH), 4.90 (s, 1H, CH of pyrazole), 3.95 (d, 2H, CH$_2$ of pyrazole), 3.81 (s, 3H, OCH$_3$).</td>
</tr>
<tr>
<td>^13C NMR</td>
<td>(DMSO-d$_6$ 100 MHz) δ ppm: 155.5, 155.0, 153.1, 147.4, 146.7, 138.4, 134.6, 134.5, 131.8, 125.2, 124.8, 123.3, 122.3, 121.9, 120.9, 119.3, 115.3, 111.4, 110.4, 104.8, 66.3, 56.0, 37.5.</td>
</tr>
<tr>
<td>ELEMENTAL ANALYSIS</td>
<td>Calculated C, 59.30; H, 3.98; N, 8.30%</td>
</tr>
<tr>
<td></td>
<td>Found C, 59.28; H, 3.97; N, 8.32%.</td>
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</table>
## Compound (32)

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<th>Description</th>
</tr>
</thead>
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<td>IUPAC Name</td>
<td>3-(benzofuran-2-yl)-N-(4-chlorophenyl)-5-(4-hydroxy-3-methoxyphenyl)-4,5-dihydro-1H-pyrazole-1-carboxamide</td>
</tr>
<tr>
<td>Chemical Formula</td>
<td>C_{25}H_{20}ClN_{3}O_{4}</td>
</tr>
<tr>
<td>Molecular Weight</td>
<td>461.897</td>
</tr>
<tr>
<td>Physical State</td>
<td>Light brown semi solid</td>
</tr>
<tr>
<td>Melting Point</td>
<td>---</td>
</tr>
<tr>
<td>IR</td>
<td>(KBr)ν_{max}(cm(^{-1})) = 3127-2973 (Ar-CH), 1685 (C=O), 1628 (C=N pyrazole), 1362 (C-N).</td>
</tr>
<tr>
<td>Mass</td>
<td>(ESI) m/z: 461.11 (M(^+))</td>
</tr>
<tr>
<td>(^1)H NMR</td>
<td>(DMSO-d(_6) 400 MHz) δ ppm: 6.70-7.80 (m, 12H, Ar-H), 5.90 (s, 1H, NH), 5.40 (s, 1H, OH), 4.70-4.73 (t, 1H, CH of pyrazole), 3.86 (s, 3H, OCH(_3)), 3.65 (d, 2H, CH(_2) of pyrazole).</td>
</tr>
<tr>
<td>(^13)C NMR</td>
<td>(DMSO-d(_6) 100 MHz) δ ppm: 155.7, 155.1, 153.0, 147.2, 146.8, 137.4, 134.6, 133.4, 129.1, 125.2, 124.8, 123.3, 120.9, 120.8, 119.4, 115.4, 111.3, 110.3, 104.8, 66.0, 56.2, 37.6.</td>
</tr>
<tr>
<td>ELEMENTAL ANALYSIS</td>
<td>Calculated Found</td>
</tr>
<tr>
<td></td>
<td>C, 65.03; H, 4.34; N, 9.11%</td>
</tr>
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</table>
### Compound (33)

<table>
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<th>Value</th>
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<td>IUPAC Name</td>
<td>3-((benzofuran-2-yl)-5-(4-hydroxy-3-methoxyphenyl)-N-(4-methoxyphenyl)-4,5-dihydro-1H-pyrazole-1-carboxamide</td>
</tr>
<tr>
<td>Chemical Formula</td>
<td>C_{26}H_{23}N_{3}O_{5}</td>
</tr>
<tr>
<td>Molecular Weight</td>
<td>457.477</td>
</tr>
<tr>
<td>Physical State</td>
<td>Dark yellow solid</td>
</tr>
<tr>
<td>Melting Point</td>
<td>249-252 °C</td>
</tr>
<tr>
<td>IR</td>
<td>(KBr)ν_{max}(cm^{-1}): 3137-2971 (Ar-CH), 1678 (C=O), 1632 (C=N pyrazole), 1367 (C-N).</td>
</tr>
<tr>
<td>Mass</td>
<td>(ESI) m/z: 457.16 (M^{+})</td>
</tr>
<tr>
<td>^1H NMR</td>
<td>(DMSO-d_{6} 400 MHz) δ ppm: 6.64-7.90 (m, 12H, Ar-H), 6.10 (s, 1H, N-H), 5.29 (s, 1H, OH), 4.87-4.90 (t, 1H, CH of pyrazole), 4.00 (d, 2H, CH_2 of pyrazole), 3.83 (s, 6H, OCH_3).</td>
</tr>
<tr>
<td>^13C NMR</td>
<td>(DMSO-d_{6} 100 MHz) δ ppm: 158.8, 155.5, 155.2, 152.9, 147.2, 146.6, 134.7, 134.4, 131.6, 125.0, 124.8, 123.3, 120.9, 119.8, 119.3, 115.4, 114.5, 111.6, 110.2, 104.8, 66.1, 56.0, 55.8, 37.7.</td>
</tr>
<tr>
<td>ELEMENTAL ANALYSIS</td>
<td>Calculated: C, 68.26; H, 5.07; N, 9.19; O, 17.49%</td>
</tr>
<tr>
<td></td>
<td>Found: C, 68.25; H, 5.04; N, 9.15; O, 17.52%.</td>
</tr>
<tr>
<td><strong>Compound (34)</strong></td>
<td></td>
</tr>
<tr>
<td>--------------------</td>
<td></td>
</tr>
<tr>
<td><strong>IUPAC Name</strong></td>
<td>3-(benzofuran-2-yl)-5-(4-hydroxy-3-methoxyphenyl)-N-p-tolyl-4,5-dihydro-1H-pyrazole-1-carboxamide</td>
</tr>
<tr>
<td><strong>Chemical Formula</strong></td>
<td>C_{26}H_{33}N_{3}O_{4}</td>
</tr>
<tr>
<td><strong>Molecular Weight</strong></td>
<td>441.478</td>
</tr>
<tr>
<td><strong>Physical State</strong></td>
<td>Yellow semisolid</td>
</tr>
<tr>
<td><strong>Melting Point</strong></td>
<td>---</td>
</tr>
<tr>
<td><strong>IR</strong></td>
<td>(KBr) ν_{max}(cm^{-1}): 3132-2974 (Ar-CH), 1670 (C=O), 1630 (C=N pyrazole), 1371 (C-N).</td>
</tr>
<tr>
<td><strong>Mass</strong></td>
<td>(ESI) m/z: 441.17 (M^+)</td>
</tr>
<tr>
<td><strong>{H NMR</strong></td>
<td>(DMSO-d_{6} 400 MHz) δ ppm: 6.62-7.85 (m, 12H, Ar-H), 5.90 (s, 1H, NH), 5.31 (s, 1H, OH), 4.80-4.83 (s, 1H, CH of pyrazole), 4.10 (d, 2H, CH_{2} of pyrazole), 3.90 (s, 3H, OCH_{3}), 2.33 (s, 3H, CH_{3}).</td>
</tr>
<tr>
<td><strong>{C NMR</strong></td>
<td>(DMSO-d_{6} 100 MHz) δ ppm: 155.8, 155.0, 153.2, 147.3, 146.7, 136.7, 136.4, 134.6, 134.4, 129.1, 125.0, 124.7, 123.2, 121.4, 121.0, 119.8, 119.3, 115.4, 114.5, 111.6, 110.2, 104.8, 66.1, 56.0, 55.8, 37.7.</td>
</tr>
<tr>
<td><strong>ELEMENTAL ANALYSIS</strong></td>
<td><strong>Calculated</strong></td>
</tr>
<tr>
<td></td>
<td>C, 70.73; H, 5.25; N, 9.52; O, 14.50%</td>
</tr>
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</table>
### Compound (35)

<table>
<thead>
<tr>
<th><strong>IUPAC Name</strong></th>
<th>3-(benzofuran-2-yl)-5-(4-hydroxy-3-methoxyphenyl)-N-(4-nitrophenyl)-4,5-dihydro-1H-pyrazole-1-carboxamide</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Chemical Formula</strong></td>
<td>C_{26}H_{20}N_{4}O_{6}</td>
</tr>
<tr>
<td><strong>Molecular Weight</strong></td>
<td>472.449</td>
</tr>
<tr>
<td><strong>Physical State</strong></td>
<td>Reddish brown semisolid</td>
</tr>
<tr>
<td><strong>Melting Point</strong></td>
<td>---</td>
</tr>
</tbody>
</table>

**IR**

(KBr)\(\nu_{\text{max}}\) (cm\(^{-1}\)): 3132-2974 (Ar-CH), 1678 (C=O), 1630 (C=N Pyrazole), 1369 (C-N).

**Mass**

(ESI) m/z: 472.14 (M\(^+\)).

**\(^1\)H NMR**

(DMSO-\(d_6\) 400 MHz) \(\delta\) ppm: 6.89-7.76 (m, 12H, Ar-H), 6.00 (s, 1H, NH), 5.30 (s, 1H, OH), 4.86-4.90 (t, 1H, CH of pyrazole), 4.05 (d, 2H, CH\(_2\) of pyrazole), 3.88 (s, 3H, OCH\(_3\)).

**\(^{13}\)C NMR**

(DMSO-\(d_6\) 100 MHz) \(\delta\) ppm: 155.7, 155.0, 153.1, 147.2, 146.7, 145.4, 143.5, 134.6, 134.3, 125.2, 124.7, 124.1, 123.2, 120.9, 119.9, 119.4, 115.4, 111.5, 110.3, 104.9, 66.2, 56.0, 37.5.

**ELEMENTAL ANALYSIS**

**Calculated**

\(\text{C, 63.56\%; H, 4.27\%; N, 11.86\%}\)

**Found**

\(\text{C, 70.71\%; H, 5.23\%; N, 9.54\%}\).
**Compound (36)**

<table>
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<th>Value</th>
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<td><strong>IUPAC Name</strong></td>
<td>3-(benzofuran-2-yl)-N-(4-fluorophenyl)-5-(4-hydroxy-3-methoxyphenyl)-4,5-dihydro-1H-pyrazole-1-carboxamide</td>
</tr>
<tr>
<td><strong>Chemical Formula</strong></td>
<td>C$<em>{26}$H$</em>{20}$FN$_3$O$_4$</td>
</tr>
<tr>
<td><strong>Molecular Weight</strong></td>
<td>445.442</td>
</tr>
<tr>
<td><strong>Physical State</strong></td>
<td>Brown semisolid</td>
</tr>
<tr>
<td><strong>Melting Point</strong></td>
<td>---</td>
</tr>
<tr>
<td><strong>IR</strong></td>
<td>(KBr)$\nu_{\text{max}}$(cm$^{-1}$): 3135-2979 (Ar-CH), 1675 (C=O), 1625 (C=N pyrazole), 1374 (C-N).</td>
</tr>
<tr>
<td><strong>Mass</strong></td>
<td>(ESI) m/z: 445.14 (M$^+$)</td>
</tr>
<tr>
<td><strong>$^1$H NMR</strong></td>
<td>(DMSO-$_d^6$ 400 MHz) δ ppm: 6.60-7.90 (m, 12H, Ar-H), 6.00 (s, 1H, NH), 5.26 (s, 1H, OH), 4.80-4.83 (t, 1H, CH of pyrazole), 3.85 (s, 3H, OCH$_3$), 3.72 (d, 2H, CH$_2$ of pyrazole).</td>
</tr>
<tr>
<td><strong>$^{13}$C NMR</strong></td>
<td>(DMSO-$_d^6$ 100 MHz) δ ppm: 162.9, 155.6, 155.2, 153.0, 147.3, 146.8, 135.0, 134.5, 134.2, 125.1, 124.7, 123.3, 120.9, 119.3, 115.7, 115.4, 111.5, 110.2, 104.9, 66.1, 56.1, 37.6.</td>
</tr>
</tbody>
</table>

**ELEMENTAL ANALYSIS**

<table>
<thead>
<tr>
<th></th>
<th>Calculated</th>
<th>Found</th>
</tr>
</thead>
<tbody>
<tr>
<td>C, H, N</td>
<td>67.41; 4.53; 9.43%</td>
<td>67.40; 4.55; 9.41%</td>
</tr>
</tbody>
</table>
Figure 3.2.1: IR Spectrum of 3-(benzofuran-2-yl)-5-(4-hydroxy-3-methoxyphenyl)-N-(4-hydroxyphenyl)-4,5-dihydro-1H-pyrazole-1-carboxamide (26)
Figure 3.2.2: $^1$H NMR Spectrum of 3-(benzofuran-2-yl)-5-(4-hydroxy-3-methoxyphenyl)-N-(4-hydroxyphenyl)-4,5-dihydro-1H-pyrazole-1-carboxamide (26)
Figure 3.2.3: Mass Spectrum of 3-(benzofuran-2-yl)-5-(4-hydroxy-3-methoxyphenyl)-N-(4-hydroxyphenyl)-4,5-dihydro-1H-pyrazole-1-carboxamide (26)
Figure 3.2.4: IR Spectrum of 3-(benzofuran-2-yl)-N-(3,4-dihydroxyphenyl)-5-(4-hydroxy-3-methoxyphenyl)-4,5-dihydro-1H-pyrazole-1-carboxamide (30)
Figure 3.2.5: $^1$H NMR Spectrum of 3-(benzofuran-2-yl)-N-(3,4-dihydroxyphenyl)-5-(4-hydroxy-3-methoxyphenyl)-4,5-dihydro-1H-pyrazole-1-carboxamide (30)
Figure 3.2.6: Mass Spectrum of 3-(benzofuran-2-yl)-N-(3,4-dihydroxyphenyl)-5-(4-hydroxy-3-methoxyphenyl)-4,5-dihydro-1H-pyrazole-1-carboxamide (30)
Figure 3.2.7: $^{13}$C NMR Spectrum of 3-(benzofuran-2-yl)-5-(4-hydroxy-3-methoxyphenyl)-N-(4-hydroxyphenyl)-4,5-dihydro-1H-pyrazole-1-carboxamide (26)
3.3. Result and discussion

3.3.1. Chemistry

In this chapter, five step synthetic strategies were employed for the preparation of benzofuran based 1,3,5-substituted pyrazole derivatives (25-36). In the first step, the desired starting material 2-acetyl benzofuran (2) was obtained by cyclocondensation reaction. At present, several general methods for the preparation of 2-acetyl benzofuran are known [18-22]. In this communication, we described a mild variant 1,8-diaza bicyclo[5.4.0]undec-7-ene (DBU) assisted one pot synthesis of 2-acetyl benzofuran in presence of molecular sieves Scheme 3.2.1. Initially, o-alkylation of salicylaldehyde (1) with chloroacetone in presence of DBU as organic base furnished o-alkylated salicylaldehyde derivative (a) which subsequently generates enolate anion undergo intramolecular cyclocondensation reaction afforded 2-acetyl benzofuran in excellent yield. In the second step, zirconium catalyzed aldol condensation reaction of appropriately 2-acetyl benzofuran (2) with vanillin in dichloromethane (DCM) afforded benzofuran chalcone (E)-1-(benzofuran-2-yl)-3-(4-hydroxy-3-methoxyphenyl)prop-2-en-1-one (22)[23]. Generally, chalcones are considered to be useful intermediate in several cyclisation reactions to produce types of heterocyclic compounds of diverse biological importance, according to the reactants used and the reaction conditions [24]. In the next step, Claisen-Schmidt cyclocondensation of compound (22) with hydrazine hydrate in absolute ethanol obtained the corresponding pyrazole derivative 4-(3-(benzofuran-2-yl)-4, 5-dihydro-1H-pyrazol-5-yl)-2-methoxyphenol (23) [25]. Compound (23) further treated with phosgene in presence of triethylamine (TEA) to furnished 3-(benzofuran-2-yl)-5-(4-hydroxy-3-methoxyphenyl)-4,5-dihydro-1H-pyrazole-1-carbonyl chloride (24). In the final step, compound (24) was coupled with different substituted aromatic amines in presence of potassium carbonate accomplished the desired carboxamide products (25-36) Scheme 3.2.2. The structures of the compounds were elucidated by IR, $^1$H NMR, $^{13}$C NMR, mass and elemental analysis.
3.3.2. Antioxidant activity

Evaluation of antioxidant activity for the newly synthesized analogues was done by using two in vitro assays such as 2,2-diphenyl-1-picryl-hydrazyl (DPPH) radical scavenging activity and inhibition microsomal lipid peroxidation (LPO). The antioxidant properties were expressed as 50% inhibitory concentration (IC$_{50}$) values (Table 3.3.2.1).

3.3.2.1. DPPH radical scavenging activity

The DPPH radical scavenging evaluation is a standard assay in antioxidant activity studies and offers a rapid technique for screening the radical scavenging activity (RSA) of specific compounds [26]. The reaction of synthesized compounds with stable DPPH free radical indicates their free radical scavenging ability. Initially, benzofuran scaffolds (23) and (24) exhibited considerable activities, this could be due to the presence of hydroxyl and electron donating methoxy group at 5-substituted phenyl ring and also presence of the N-H functional group in the scaffold (23) [27]. Further, coupling of substituted anilines gave the significant enhancement of activity. Majority of the tested compounds in these series (25-36) showed moderate to high activity. Dominant RSA was observed for compounds (28) and (30) this may be presence of hydroxyl and methoxy group on the 5-substituted phenyl ring of the pyrazole skeleton as well as in 1-substituted carboxamide phenyl ring. The coupling of substituted anilines (3-methoxy-4-hydroxy aniline and 3,4-dihydroxy aniline) to compound (24) have revealed ten to thirteen folds more activity compared to the benzofuran scaffolds (23) and (24). Antioxidant activity of these compounds is related with their electron- or hydrogen-donating ability to DPPH radical, so that it become stable diamagnetic molecules. This might be the reason for the higher antioxidant activity of the compounds (28) and (30) among the synthesize analogues as well as BHA. The introduction of electron withdrawing (Br, Cl, NO$_2$ and F) substituted aromatic amines to scaffold (24) was inadequate for the enhanced activity in compounds (31), (32), (35) and (36). Whereas, the compounds (26), (27), (29), (33) and (34)contain electron releasing single hydroxyl, methoxy and methyl groups at positions 3,4,5 exhibited five to six
folds more RSA activity than that of (23) and (24). Since compound (25) does not have any substituent on the ring it showed least activity compared to other compounds.

3.3.2.2. Inhibition of lipid peroxidation assay

Inhibition of lipid peroxidation property of newly synthesized compounds was performed by the formation of thiobarbituric acid reactive species (TBARS) using liver excised from adult male Wister rats. Lipid free radicals produced by ferric chloride and vitamin C rapidly react with oxygen molecule to give peroxyl radicals with the subsequent formation of the final product, malondialdehyde [28]. The results showed that all newly synthesized compounds (25-36) inhibited the ferric chloride induced lipid peroxidation and varying degree compared with standard antioxidant BHA Table 3.3.2.1. Compound (28) which contains p-OH, m-OCH₃ and compound (30) having 3,4-dihydroxy group at 1-substituted carboxamide phenyl ring showed maximum inhibition and even higher than that of the reference compound BHA. In general, the presence of electron-donating groups on the phenyl ring favors the activity. This might be the reason for the four-seven times enhancement of activity of the other compounds, which showed moderate to good activities in the order (26)>(27)>(29)>(33)>(34) which has free hydroxyl, methoxy and methyl as the electron donating substituent’s. The presence of electron withdrawing groups on the phenyl ring was not favor for the activity, thus might be the reason for the decreased activity for the compounds (31), (32), (33) and (36).
**Table 3.3.2.1:** 50% Inhibition of DPPH radical and microsomal LPO inhibition by compounds (25-36). Each value represents mean ± SD (n=3).

<table>
<thead>
<tr>
<th>Compound No</th>
<th>DPPH activity IC\textsubscript{50} (µM/mL)\textsuperscript{a}</th>
<th>LPO inhibition IC\textsubscript{50} (µM/mL)\textsuperscript{b}</th>
</tr>
</thead>
<tbody>
<tr>
<td>23</td>
<td>100±0.61</td>
<td>112±0.88</td>
</tr>
<tr>
<td>24</td>
<td>135±0.30</td>
<td>147±0.97</td>
</tr>
<tr>
<td>25</td>
<td>121±0.91</td>
<td>116±0.13</td>
</tr>
<tr>
<td>26</td>
<td>17±0.32</td>
<td>15±0.21</td>
</tr>
<tr>
<td>27</td>
<td>19±0.11</td>
<td>22±0.46</td>
</tr>
<tr>
<td>28</td>
<td>10±0.56</td>
<td>12±0.45</td>
</tr>
<tr>
<td>29</td>
<td>21±0.34</td>
<td>24±0.12</td>
</tr>
<tr>
<td>30</td>
<td>9.5±0.41</td>
<td>11±0.64</td>
</tr>
<tr>
<td>31</td>
<td>105±0.52</td>
<td>115±0.53</td>
</tr>
<tr>
<td>32</td>
<td>107±0.27</td>
<td>121±0.71</td>
</tr>
<tr>
<td>33</td>
<td>25±0.79</td>
<td>29±0.96</td>
</tr>
<tr>
<td>34</td>
<td>37±0.60</td>
<td>44±0.45</td>
</tr>
<tr>
<td>35</td>
<td>103±0.14</td>
<td>113±0.23</td>
</tr>
<tr>
<td>36</td>
<td>110±0.45</td>
<td>125±0.31</td>
</tr>
</tbody>
</table>

\textsuperscript{a}IC\textsubscript{50} = the concentration (µM/mL) exhibiting 50% inhibition of DPPH radical

\textsuperscript{b}IC\textsubscript{50} = the concentration (µM/mL) exhibiting 50% inhibition of LPO oxidation.
Figure 3.3.2.1: Concentration required for 50% inhibition of DPPH and LPO radical
3.4. Conclusion

In conclusion, we have described a mild variant DBU assisted one pot synthesis of 2-acetyl benzofuran. Aldol condensation of 2-acetyl benzofuran with vanillin afforded respective chalcone, which undergo further Claisen-Schmidt condensation with hydrazine hydrate followed by coupling of different substituted anilines furnished benzofuran based 1,3,5-substituted pyrazole derivatives (25-36) in reasonably good yields. The newly synthesized analogues were evaluated for their in vitro antioxidant activity. Among the analogues compounds (28) and (30) demonstrated potent antioxidant activity. The data obtained operate as a positive reinforce of the tendency to use antioxidant properties as a guideline of the rational design of this type of compounds.
3.5. References


