Anti-Leishmanial Activity of Flavanone Analogues Targeting Pteridine Reductase

Afroze Alam1,2, Vinay Pandit3, Shailendra Kumar4, Kamlesh Kumar Naik5, Mahfoozur Rahman6, Mohan Lal Verma7

1Narayan Institute of Pharmacy, Jamhar, Rohtas (Sasaram), Bihar, INDIA.
2Narayan Medical College and Hospital, Jamhar, Rohtas (Sasaram), Bihar, INDIA.
3Laureate Institute of Pharmacy, SH 22, VPO Kathog, Kangra, Himachal Pradesh, INDIA.
4Govt. Pharmacy Institute, Agam kuan, Patna, Bihar, INDIA.
5Nandha College of Pharmacy, Perundurai Main Road, Erode, Tamil Nadu, INDIA.
6Department of Pharmaceutical Sciences, Faculty of Health Sciences, Sam Higginbottom Institute of Agriculture, Technology and Sciences (SHIATS), Allahabad, Uttar Pradesh, INDIA.

ABSTRACT

Objectives: The aim of the study is to develop new synthetic anti-leishmanial agents as flavanone analogues, which should have low toxicity with noticeable yield.

Methodology: The starting materials for the synthesis of test compounds were 2'-hydroxypropiohenones, 2'-hydroxyacetophenone and substituted benzaldehyde. Test compounds were synthesized by three steps reaction starting from condensation, cyclization and reduction to yield 3-substituted flananone analogues. The synthesized compounds were screened by in vitro anti-leishmanial assay against promastigotes of *L. donovani*. Result: A series of flavanone analogues have been synthesized using cobalt (II) phthalocyanine and NaBH₄ in the equimolar ratio of the reactant with noticeable yield. The structures of the test compounds were elucidated and established by U.V. IR, ¹H-NMR, ¹³C-NMR and mass spectrometry. The synthesized compounds were screened by in vitro antileishmanial assay against promastigotes of *L. donovani*. Conclusion: Most of the compounds exhibited moderate leishmanicidal activity, while some compounds such as 4b, 10b, 5b, and 3a have shown promising antileishmanial activity against promastigotes of *L. donovani*.

Key words: Antileishmanial activity; Cobalt (II)phthalocyanine; Flavanone derivatives; *L. donovani*; Promastigotes.

INTRODUCTION

Leishmaniasis is a vector-borne disease and considered as one of the world’s most neglected diseases. The annual global incidence of leishmaniasis is approximately 12 million cases.¹ Leishmaniasis is endemic in 88 countries with more than 350 million peoples are at risk.² ³ The number of cases reported globally has increased over the past 10 years due to the increase in anti-leishmanial drug resistance and lack of adequate vector or reservoir control tools.⁴ Antimonial compounds are the first line drugs and drug of choice for the treatment of leishmaniasis but the emergence of drug resistance against these drugs is an emerging problem and spread of drug resistance strains of *Leishmania* is an alarming feature. Newer drugs like Miltefosine and Amphoterisin B is approved for the treatment of leishmaniasis are highly toxic and above all are very expensive. Thus, the control of leishmaniasis has become a costly affair due to the high cost of alternative drugs. For the control of leishmaniasis, there is a need to discover a novel compound which should not only be less toxic but also cost effective. During the last few years, researchers have focused their interest on the discovery of new anti-leishmaniasis compounds.
als by the isolation of bioactive molecules from plants, by semi synthetic and synthetic approach and some of synthetic molecules undergo clinical trials which have shown efficacy against Leishmania parasite. Chalcones and flavone analogues are the integral component of many pharmacological active compounds like antipROTOzoal, anti-inflammatory, immunomodulatory, nitric oxide inhibition, anticancer. The molecular basis of the anti-leishmanial action is still not well established. The Pteridine reductase (PTR1) is a member of oxido-reductase family, which has become an attractive target for the development of novel anti-leishmanial agents. Pteridine reductase is a flavoprotein, which is unique in leishmania and plays vital role in the growth of the leishmania via generation of intermediate which is required for the synthesis of DNA precursors. The enzyme PTR1 is predominantly involved in reduction of biopterin to dihydrobiopterin and tetrahydrobiopterin but it is also capable to reduce dihydrofolate to tetrahydrofolute. Hence, a combined strategy to target both PTR1 and Dihydrofolate reductase (DHFR) will be more effective to stop parasitic growth and survival. A number of other compounds have been screened against PTR1 in L. donovani. Several natural flavonoids were found to be inhibiting both DHFR and PTR1. So, the present strategy reflects the study of new flavanone analogues as promising anti-leishmanial agents targeting Pteridine reductase coupled with DHFR. This indicates that some novel flavanone analogues as an inhibitor are essentially required that targets both the enzymes simultaneously.

Researchers recently identified several types of flavonoids as antipROTOzoal principles of plant extracts, but a comprehensive study of their structure-activity relationships (SARs) has not been conducted so far. Therefore, it was our fine interest to assess the in vitro leishmanicidal activities of a large 3-substituted flavanone analogues library, draw SARs, and determine their anti-leishmanial activity without significant toxicity in mammalian cells.

**MATERIALS AND METHODS**

**Chemicals**

All the chemicals and solvents used were of AR-grade and LR-grade and were obtained from Sigma-Aldrich, Sisco Research Laboratories, Qualigens, Rankem, S.D. Fine, Hi-Media and Merck.

**Synthesis of 3-methyl chalcone**

To a solution of 0.01 mole of substituted 2'-hydroxyacetophenones in 10 ml of 40% KOH and 20 ml of ethyl alcohol, 0.01 mole of substituted benzaldehyde was added and mixture was stirred for 48-72 h. The coloured solution was poured into crushed ice and acidified with 1N HCl at 24-26°C. The precipitate so obtained was washed with cold water, filtered, dried and recrystallized with absolute alcohol.

**Synthesis of 3-methyl flavone**

To a solution of 0.01 mole of chalcone in 50 ml of dimethyl sulphoxide (DMSO) taken in 100 ml round bottom flask fitted with reflux condenser, 15-20 granules of iodine was added. The reaction mixture was refluxed for 3-4 h and kept overnight. The precipitate was neutralized with sodium thiosulphate to remove unreacted I₂ washed with water, fitted, dried and recrystallized with absolute alcohol.

**Synthesis of 3-methyl flavanone**

To 0.01 moles of 3-methyl flavone in 100ml round bottom, the equimolar amount of NaBH₄ and 10-15 ml of methanol in the presence of AlCl₃ was added and mixture was refluxed for 2-3 h. The resulting solution was cooled to room temperature followed by the addition of ice cold water. The solid was separated by filtration, washed with cold water and recrystallized from ethanol. The experiments suggest that the present reductive system initially reduces the conjugated double bond see Table 1.

**Synthesis of 2'-hydroxy chalcones**

To a solution of 0.01 mole of substituted 2'-hydroxyacetophenones in 10 ml of 40% KOH and 20 ml of ethyl alcohol, 0.01 mole of substituted benzaldehyde was added and mixture was stirred for 48-72 h. Completion of the reaction was monitored on TLC (20% Ethyl acetate in toluene). The coloured solution was poured into crushed ice and acidified with 1N HCl at 24-26°C. The precipitate so obtained was washed with cold water, filtered, dried and recrystallized from ethanol.

**Synthesis of 3-hydroxyflavones**

Figure 1(A): portrays the scheme-1 for synthesis of 3-methyl flavanone derivatives using various steps.
To a suspension of 0.01ml of chalcone, 50 ml of ethanol was added 10 ml of 20% aqueous sodium hydroxide with stirring, followed by the careful addition of 15 ml of 30% hydrogen peroxide over a period of 0.5-1 h. The reaction mixture was stirred for 3-5 h at 30°C and completion of the reaction was monitored on TLC (20% Ethyl acetate in toluene). After reaction completion, the mixture was poured into crushed ice containing 5 N HCl. The precipitate was filtered, washed, dried and recrystallized from ethyl acetate.

**Synthesis of 3-hydroxyflavanone analogues**

To a solution of 0.01 mole of 3-hydroxyflavone, 10-15 ml of methanol and 0.01 mole of NaBH₄ was added and the mixture was refluxed for 3-4 h in the presence of cobalt(II) phthalocyanine to yield 3-hydroxyflavanone analogues.

**Anti-leishmanial Activity**

**Chemicals**

*Parasite stock culture*: Axenic culture of *L. donovani* (LdMIPL-1) was maintained at 25°C in RPMI 1640 (Himedia, India) medium supplemented with 10% heat inactivated Fetal Bovine Serum (FBS) (Himedia, Mumbai, India), streptomycin (150 μg/ml), penicillin G (100 μg/ml) and gentamycin (150 μg/ml) at pH 7.2

**Anti-leishmanial assay**

For antileishmanial activity, pro-mastigotes of *L. donovani* were sub-cultured in Schneider’s Insect Medium (Himedia, Mumbai, India) supplemented with 10% heat inactivated FBS, streptomycin (150 μg/ml), penicillin G (100 μg/ml) and gentamycin (150 μg/ml). The antileishmanial screening was performed in 96-well flat bottom tissue culture plates (Corning Life Science, Corning USA) 100 microliters of cell suspension containing 2 × 10⁶ to 3 × 10⁶ cells/ml was poured in each well of the plate. Four different concentrations of the methanolic extracts i.e. 100, 250, 350 and 500 μg/ml were used as positive controls and cell suspension with 0.025% DMSO was used as a negative control. Inhibition of the promastigotes was assessed by measuring the cleavage of 10 mg/mL of MTT [3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide]. The absorbance was measured using an ELISA plate reader (BioTek, USA) at 595 nm. Percent growth inhibition was calculated by the following formula:

\[
\text{% of Inhibition} = \frac{\text{OD Control} - \text{OD Treated}}{\text{OD Control}} \times 100
\]

**Molecular docking into bioppterin/dihydrofolate binding site of Pteridine reductase (PTR1).**

Library of 20 substituted flavanone analogues were prepared by using chemdraw ultra 11.0. All ligands were prepared through Auto Dock Tools. The 3D crystal structure was obtained from Protein Data Bank (PDB code: 2XOX). The 3D structures of PTR1, (from 2XOX), was used for virtual screening. Docking parameters were set to default values on the basis of Lamarckian genetic algorithm principle. Autogrid program of AutoDock suit was used for generation of grid around binding pocket within target protein. Finally, docking simulation was carried out with AutoDock 4.2. Ligplot and UCSF Chimera version 1.8.1 were used for analysis of docking results (protein ligand interaction) and visualization of docked protein ligand complexes.

**RESULTS**

**Chemistry**

Synthesis of 3-hydroxy and 3-methylflavanone analogues involve 3 steps (See Figure 1A and 1B). Spectral Analysis of 1a-10a (3.1.1.1 to 3.1.1.10) and 1b-10b (3.1.1.11 to 3.1.1.20) are available in supplementary file.

2-(4-fluorophenyl)-2,3-dihydro-3-methylchromen-4-one (1a)

Pale yellow solid, physical data is summarized in Table 2. IR spectra (KBr cm⁻¹): 3041(\text{ArC-H}), 2762 (C–H), 1707 (C=O), 1514 (aromatic C=C), 1156 (C-F).

1H-NMR (400MHz, CDCl₃, TMS=0): 8.2(1H, d, J=7.72 Hz, 5-H), 7.66 (1H, dd, J=2.76 Hz, J=7.88 Hz, 7-H), 7.65 (2H, m, 2',6'-H), 7.46 (1H, d, J=7.84 Hz, 7-H)
2.3-dihydro-2-(3-hydroxy-4-methoxyphenyl)-3-methylchromen-4-one (4a)

Yellow solid, physical data is summarized in Table 2.

IR Spectra (KBr cm⁻¹): 3074 (Ar-C-H), 2851 (C-H), 1685 (C=O), 1514 (aromatic C=C), 1182 (C-O).

'H-NMR (400 MHz, CDCl₃, TMS=0): 7.94 (1H, d, J=9.56 Hz, 5-H), 7.51 (1H, dd, J=8.64 Hz, 2.8 Hz, 7-H), 7.47 (1H, dd, J=7.45 Hz, J= 2.8 Hz, 6-H), 7.07 (1H, d, J=8.2 Hz, 8-H ), 7.02 (1H, d, J=8.45 Hz, 6' -H ), 6.98( 1H,d, J=1.96Hz, 2'-H), 6.91(1H,d,J=7.88Hz, 5'H), 5.02(1H, s, 3'-OH) Exchangeable with D₂O, 4.30 (1H, d, 3-H J= 7.67 Hz), 5.56 (1H, d, J= 12.36 Hz), 3.92(3H, s, 4'-OCH₃), 2.19(3H, s, 3-CH₃).

13C NMR (400 MHz, δ, CDCl₃, TMS=0): Aromatic Ring-A (156.29, 135.24, 132.24, 129.03, 123.94, 116.06) 182.88 (C=O), 145.36 (2-C), 117.28 (3-C), Aromatic Ring B (155.47, 128.45, 128.03, 123.44, 115.13, 116.68), 60.5 –OCH₃, (10.09 -3 CH₃), TOF MS ES+ m/z 287.5

2-(benzo[d][1,3]dioxol-6-yl)-2,3-dihydro-3-methylchromene-4-one (5a)

Yellow solid, physical data is summarized in Table 2.

IR Spectra (KBr cm⁻¹): 3074 (Ar-C-H), 2857 (C-H), 1682 (C=O), 1490 (aromatic C=C), 1249 (C-O).

'H-NMR (400MHz, CDCl₃, δ, TMS=0): 8.32(1H, d, J=8.32 Hz, 5-H), 7.69(1H, d, J=7.08 Hz, 7-H), 7.55(1H, dd, J=8.4 Hz, J=8.53 Hz, 6-H ), 7.42(1H, d, J= 6.89 Hz, 8-H), 7.38 (1H,d,J = 7.00 Hz,2'-H), 6.96 (1H, d, J=8.24Hz, 5'-H), 6.71(1H,d,J = 7.56Hz,6'-H), 6.0(2H, s, 4'-CH₂). 4.40 (1H, d, 3-H J= 7.80 Hz), 5.60 (1H, d, 2-H J= 12.56Hz), 1.8 (3H, s, 3-CH₃).

13C NMR (400 MHz, δ, CDCl₃, TMS=0): Aromatic Ring-A (157.29, 133.24, 132.24, 128.03, 123.94, 115.06) 183.88 (C=O), 155.36 (2-C), 110.78 (3-C), Aromatic Ring B (149.47, 148.45, 126.03, 120.34, 116.13, 112.68, 107.23), (9.09 -3 CH₃). TOF MS ES+ m/z 283.5

2-(2-chlorophenyl)-2,3-dihydro-3-methylchromen-4-one (6a)

Light yellow solid, the physical data is summarized in Table 2.

IR Spectra (KBr cm⁻¹): 3050 (Ar-C-H), 2805 (C-H), 1645 (C=O), 1510 (aromatic C=C), 745 (C=O).

'H-NMR (400MHz, CDCl₃, δ, TMS=0): 8.2 (1H, d, J=8.64 Hz, 5-H), 7.8 (1H, d, J=8.24 Hz, 7-H), 7.44 (1H, d, J=7.6 Hz, 3'-H ) , 7.23 (1H, d, J=7.96 Hz, 6'-H), 7.01 (1H, dd, J=2.2 Hz, J=7.8 Hz, 4'-H ), 6.92 (1H, dd, J=2.0 Hz, J=7.8 Hz, 5'-H).

13C NMR (400 MHz, δ, CDCl₃, TMS=0): Aromatic Ring-A (156.19, 135.24, 131.24, 130.03, 122.94, 115.06) 183.88 (C=O), 125.36 (2-C), 115.78 (3-C), Aromatic Ring B (152.47, 126.45, 125.03, 123.44, 117.13, 116.68), 10-3CH₃, TOF MS ES+ m/z 255.5

Table 1: List of various substitutions on Ring A and Ring B

<table>
<thead>
<tr>
<th>COMPOUNDS</th>
<th>R₁</th>
<th>R₂</th>
</tr>
</thead>
<tbody>
<tr>
<td>1a</td>
<td>H</td>
<td>F</td>
</tr>
<tr>
<td>2a</td>
<td>H</td>
<td>Cl</td>
</tr>
<tr>
<td>3a</td>
<td>H</td>
<td>OH</td>
</tr>
<tr>
<td>4a</td>
<td>H</td>
<td>OCH₃</td>
</tr>
<tr>
<td>5a</td>
<td>Methylene dioxide</td>
<td>R₃ and R₅</td>
</tr>
<tr>
<td>6a</td>
<td>Cl</td>
<td>H</td>
</tr>
<tr>
<td>7Δ</td>
<td>H</td>
<td>CH₃</td>
</tr>
<tr>
<td>8Δ</td>
<td>H</td>
<td>Br</td>
</tr>
<tr>
<td>9Δ</td>
<td>OCH₃</td>
<td>H</td>
</tr>
<tr>
<td>10Δ</td>
<td>H</td>
<td>H</td>
</tr>
</tbody>
</table>

R₁, R₂, R₃, and R₅ are the different substitution of ring B.
Table 2: Physicochemical characterization of 3-methylflavonone analogues

<table>
<thead>
<tr>
<th>Code</th>
<th>Molecular Formula</th>
<th>Molecular Weight</th>
<th>(% Yield)</th>
<th>Melting Point (°C)</th>
<th>λ&lt;sub&gt;max&lt;/sub&gt; nm</th>
<th>R&lt;sub&gt;1&lt;/sub&gt; Value</th>
<th>Elemental Analysis Calculated (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1a</td>
<td>C&lt;sub&gt;9&lt;/sub&gt;H&lt;sub&gt;12&lt;/sub&gt;FO&lt;sub&gt;2&lt;/sub&gt;</td>
<td>256.02</td>
<td>73</td>
<td>95-100</td>
<td>341</td>
<td>0.73</td>
<td>C 77.20 H 5.27 O 13.06 N 7.90</td>
</tr>
<tr>
<td>2a</td>
<td>C&lt;sub&gt;9&lt;/sub&gt;H&lt;sub&gt;12&lt;/sub&gt;ClO&lt;sub&gt;2&lt;/sub&gt;</td>
<td>272.71</td>
<td>80</td>
<td>170-175</td>
<td>351</td>
<td>0.62</td>
<td>C 69.67 H 4.93 O 12.60 Cl/F 12.00</td>
</tr>
<tr>
<td>3a</td>
<td>C&lt;sub&gt;9&lt;/sub&gt;H&lt;sub&gt;12&lt;/sub&gt;ClO&lt;sub&gt;3&lt;/sub&gt;</td>
<td>254.08</td>
<td>82</td>
<td>125-130</td>
<td>348</td>
<td>0.71</td>
<td>C 77.42 H 5.23 O 19.56 Cl/F -</td>
</tr>
<tr>
<td>4a</td>
<td>C&lt;sub&gt;9&lt;/sub&gt;H&lt;sub&gt;12&lt;/sub&gt;O&lt;sub&gt;4&lt;/sub&gt;</td>
<td>268.29</td>
<td>90</td>
<td>90-95</td>
<td>343</td>
<td>0.57</td>
<td>C 77.44 H 5.00 O 19.56 Cl/F -</td>
</tr>
<tr>
<td>5a</td>
<td>C&lt;sub&gt;9&lt;/sub&gt;H&lt;sub&gt;12&lt;/sub&gt;ClO&lt;sub&gt;4&lt;/sub&gt;</td>
<td>262.27</td>
<td>70</td>
<td>110-115</td>
<td>308</td>
<td>0.65</td>
<td>C 73.48 H 5.66 O 21.14 Cl/F -</td>
</tr>
<tr>
<td>6a</td>
<td>C&lt;sub&gt;9&lt;/sub&gt;H&lt;sub&gt;12&lt;/sub&gt;ClO&lt;sub&gt;5&lt;/sub&gt;</td>
<td>272.04</td>
<td>80</td>
<td>165-170</td>
<td>328</td>
<td>0.56</td>
<td>C 71.12 H 5.00 O 12.98 Cl/F 13.90</td>
</tr>
<tr>
<td>7a</td>
<td>C&lt;sub&gt;9&lt;/sub&gt;H&lt;sub&gt;12&lt;/sub&gt;O&lt;sub&gt;6&lt;/sub&gt;</td>
<td>252.29</td>
<td>90</td>
<td>102-105</td>
<td>336</td>
<td>0.58</td>
<td>C 82.20 H 6.10 O 13.25 Cl/F -</td>
</tr>
<tr>
<td>8a</td>
<td>C&lt;sub&gt;9&lt;/sub&gt;H&lt;sub&gt;12&lt;/sub&gt;BrO&lt;sub&gt;2&lt;/sub&gt;</td>
<td>315.99</td>
<td>70</td>
<td>173-177</td>
<td>344</td>
<td>0.87</td>
<td>C 61.03 H 4.57 O 11.24 Br/F 26.19 (Br)</td>
</tr>
<tr>
<td>9a</td>
<td>C&lt;sub&gt;9&lt;/sub&gt;H&lt;sub&gt;12&lt;/sub&gt;O&lt;sub&gt;7&lt;/sub&gt;</td>
<td>268.29</td>
<td>90</td>
<td>110-114</td>
<td>345</td>
<td>0.55</td>
<td>C 77.44 H 5.00 O 19.56 Cl/F -</td>
</tr>
<tr>
<td>10a</td>
<td>C&lt;sub&gt;9&lt;/sub&gt;H&lt;sub&gt;12&lt;/sub&gt;O&lt;sub&gt;8&lt;/sub&gt;</td>
<td>238.27</td>
<td>90</td>
<td>98-112</td>
<td>310</td>
<td>0.66</td>
<td>C 80.12 H 5.94 O 14.25 Cl/F -</td>
</tr>
</tbody>
</table>

Physical Characterizations of 3-methylflavonone derivatives: (TLC Solvent Used: Toluene : Ethyl Acetate 8:2) % yields range between 70-90 and other physical parameters were determined by usual methods.

Table 2: Physicochemical characterization of 3-methylflavonone analogues

**J=7.32 Hz, 5'-H), 6.90((1H,d,J=7.89 Hz, 6-H), 6.88 (1H, d, J=7.50 Hz, 8-H), 4.50 (1H, d, 3-H J= 7.80 Hz), 5.65 (1H, d, 2-H J=12.56Hz), 1.9 (3H, s, 2-CH). <sup>1</sup>H NMR (400 MHz, δ, CDCl<sub>3</sub>, TMS=0): Aromatic Ring-A (156.29, 135.24, 132.24, 129.03, 123.94, 116.16) 182.88 (C=O), 158.26 (2-C), 112.43 (3-C), Aromatic Ring B (136.23, 132.56, 131.89, 129.34, 126.13, 124.68), (8.23 -3 CH). TOF MS ES+ m/z 273.5.

**2,3-dihydro-3-methyl-2-p-tolylchromen-4-one** (7a): White yellow solid, the physical data is summarized in Table 2. IR spectra (KBr cm<sup>-1</sup>): 3080 (A=H<sub>2</sub>-C-H), 2910 (C-H), 1690 (Ar=H-C-C), 1182 (C-O). <sup>1</sup>H NMR (400MHz, δ, CDCl<sub>3</sub>, TMS=0): 8.21(1H, d, J=7.72 Hz, 5-H), 7.98 (1H, dd, J=8.64Hz, 7.04Hz, 7-H), 7.82 (2H, m, 3',6'-H), 7.75 (2H, m, 3', 5', -H), 7.38 (1H,d,J=7.26Hz, 6-H), 7.05 (1H,d,J=7.89Hz, 8-H), 4.50 (1H, d, 3-H J= 7.80 Hz), 5.65 (1H, d, 2-H J=12.56Hz), 2.06 (3H, s, 4'CH) 1.9 (3H, s, 3-CH). <sup>13</sup>C NMR (400 MHz, δ, CDCl<sub>3</sub>, TMS=0): Aromatic Ring-A (157.29, 135.26, 133.24, 130.03, 125.94, 118.24), 183.88 (C=O), 159.26 (2-C), 111.43 (3-C), Aromatic Ring B (138.23, 130.56, 129.89, 127.34, 126.13, 125.68), (30.67-4'CH) (10.33 -3 CH). TOF MS ES+ m/z 253.1.

**2-(2-bromophenyl)-2,3-dihydro-3-methylchromen-4-one (6a)**

Light yellow solid, the physical data is summarized in Table 2.

8a: IR spectra (KBr cm<sup>-1</sup>): 3072 (A=H<sub>2</sub>-C-H), 2875 (C-H), 1663 (C=O), 1525 (Ar C=C), 1182 (C-O). <sup>1</sup>H NMR (400MHz, δ, CDCl<sub>3</sub>, TMS=0): 7.92(1H, d, J=7.70 Hz, 5-H), 7.82 (1H, dd, J=8.04Hz, 7-H), 7.82 (2H, m, 2',6'-H), 7.75 (2H, m, 3', -H), 7.38 (1H,d, J= 7.26 Hz, 6-H), 7.05(1H,d, J=7.89Hz, 8-H), 4.50 (1H, d, 3-H J= 7.80 Hz), 5.65 (1H, d, 2-H J= 12.56Hz), 2.00 (3H, s, 3-CH). <sup>13</sup>C NMR (400 MHz, δ, CDCl<sub>3</sub>, TMS=0): Aromatic Ring-A (156.29, 135.24, 132.24, 129.03, 123.94, 116.06) 182.88 (C=O), 145.36 (2-C), 112.78 (3-C), Aromatic Ring B (159.47, 131.45, 129.03, 118.34, 115.13, 110.68), 59.5 -OCH<sub>3</sub>, (9.09 -3 CH). TOF MS ES+ m/z 269.5.

**2,3-dihydro-3-methyl-2-phenylchromen-4-one** (10a)

Yellow solid, the physical data is summarized in Table 2. IR spectra (KBr, cm<sup>-1</sup>): 3085(A=H<sub>2</sub>-C-H), 2920 (C-H), 1690 (C=O), 1520 (Ar C=C), 1182 (C-O). <sup>1</sup>H NMR (400MHz, δ, CDCl<sub>3</sub>, TMS=0): 7.20(1H, d, J=7.70Hz,5-H), 7.09 (1H, dd, J=8.60Hz, 7.04Hz,7-H), 7.00 (2H, m, 2',6'-H), 6.98 (2H, m, 3', 5', -H), 6.90 (1H,d,J=7.80Hz ,4'-H),
Table 3: Physicochemical characterization of 3-hydroxyflavanone analogues

<table>
<thead>
<tr>
<th>Com. Code</th>
<th>Molecular Formula</th>
<th>Molecular Weight</th>
<th>(% Yield)</th>
<th>Melting Point (°C)</th>
<th>λ_{max} (nm)</th>
<th>Rf Value</th>
<th>Elemental Analysis Calculated (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1b</td>
<td>C_{11}H_{14}NO_{3}</td>
<td>238.32</td>
<td>88.24</td>
<td>134-136</td>
<td>341</td>
<td>0.63</td>
<td>C - 54.78, H - 7.42, O - 37.80, N - 0.90, Cl - -</td>
</tr>
<tr>
<td>2b</td>
<td>C_{10}H_{15}ClO_{3}</td>
<td>274.68</td>
<td>88.23</td>
<td>105-107</td>
<td>351</td>
<td>0.48</td>
<td>C - 55.93, H - 7.90, O - 36.17, N - -</td>
</tr>
<tr>
<td>3b</td>
<td>C_{10}H_{15}O_{3}</td>
<td>230.20</td>
<td>91.00</td>
<td>110-112</td>
<td>348</td>
<td>0.59</td>
<td>C - 62.42, H - 7.32, O - 30.26, N - -</td>
</tr>
<tr>
<td>4b</td>
<td>C_{10}H_{15}NO_{4}</td>
<td>313.33</td>
<td>86.00</td>
<td>168-170</td>
<td>343</td>
<td>0.56</td>
<td>C - 62.42, H - 7.32, O - 30.26, N - -</td>
</tr>
<tr>
<td>5b</td>
<td>C_{10}H_{15}ClO_{4}</td>
<td>304.70</td>
<td>83.00</td>
<td>107-109</td>
<td>308</td>
<td>0.44</td>
<td>C - 62.42, H - 7.32, O - 30.26, N - -</td>
</tr>
<tr>
<td>6b</td>
<td>C_{10}H_{15}O_{5}</td>
<td>260.22</td>
<td>90.32</td>
<td>120-122</td>
<td>328</td>
<td>0.50</td>
<td>C - 62.42, H - 7.32, O - 30.26, N - -</td>
</tr>
<tr>
<td>7b</td>
<td>C_{10}H_{15}NO_{5}</td>
<td>297.33</td>
<td>79.00</td>
<td>180-182</td>
<td>336</td>
<td>0.57</td>
<td>C - 62.42, H - 7.32, O - 30.26, N - -</td>
</tr>
<tr>
<td>8b</td>
<td>C_{10}H_{15}O_{5}</td>
<td>288.70</td>
<td>70.00</td>
<td>112-114</td>
<td>344</td>
<td>0.42</td>
<td>C - 62.42, H - 7.32, O - 30.26, N - -</td>
</tr>
<tr>
<td>9b</td>
<td>C_{10}H_{15}O_{5}</td>
<td>244.22</td>
<td>79.00</td>
<td>147-149</td>
<td>345</td>
<td>0.49</td>
<td>C - 62.42, H - 7.32, O - 30.26, N - -</td>
</tr>
<tr>
<td>10b</td>
<td>C_{10}H_{15}NO_{5}</td>
<td>285.05</td>
<td>85</td>
<td>135-139</td>
<td>310</td>
<td>0.67</td>
<td>C - 62.42, H - 7.32, O - 30.26, N - -</td>
</tr>
</tbody>
</table>

Physicochemical characterization of 3-hydroxyflavanone analogues (1b-10b)

- 6.88 (1H,d, J= 7.21 Hz, 6-H), 6.75 (1H,d, J= 7.82 Hz, 8-H), 4.50 (1H, d, 3-H J= 7.80 Hz), 5.65 (1H, d, 2-H J= 12.56Hz), 1.98 (3H, s, 3-CH3). 1^C NMR (400 MHz, δ, CDCl3, TMS=0): Aromatic Ring-A (159.29, 134.24, 132.27, 129.83, 122.94, 117.06) 183.88 (C=O), 157.36 (2-C), 110.78 (3-C), Aromatic Ring B (160.47, 130.45, 130.03, 128.34, 128.13, 127.68), (9.09 -3 CH3), TOF MS ES+ (m/z) 239.5

2-(4-dimethylamino)phenyl-2,3-dihydro-3-hydroxychromen-4-one (1b)

Yellow-brown solid, physical data is summarized in Table 3. IR (KBr, cm⁻¹): 3355 (Ar-OH), 1559 (Ar=C=C str), 1689 (C=O str), 1327 (C-O str), 2895 (C-H str), 3027 (Ar-H), 1318, (C=N). 1H NMR (400 MHz, DMSO, δ, TMS=0): δ = 12.69 (1H, s, 3-OH, Exchangeable with D₂O), 7.59 (1H, d, 5-H J= 8.42 Hz), 7.51 (1H, d, 7-H J= 8.24 Hz), 7.44 (2H, m, '2'-H), 7.39 (1H, d, 6-H J= 7.76 Hz), 6.97 (1H, d, 8-H, J= 8.23 Hz), 6.74 (2H, m, 3'-H), 5.60 (1H, d, 3-H, J= 12.56Hz), 5.53 (1H, d, 2-H, J= 8.65Hz), 2.97 (6H, s, 4'-Dimethyl amimo), 13C NMR (400 MHz, δ, CDCl3, TMS=0): Aromatic Ring-A (152.24, 132.12, 131.87, 124.43, 122.49, 117.10) 178.88 (C=O), 148.32 (2-C), 138.35 (3-C), Aromatic Ring B (153.44, 147.89, 127.35, 127.36, 118.68, 118.68), 40.16, 40.17, N-(CH₃)₂, TOF MS ES+ (m/z)= 282.

2-(4-chlorophenyl)-2,3-dihydro-3-hydroxychromen-4-one (2b)

Yellow-white solid, physical data is summarized in Table 3. IR (KBr, cm⁻¹): 3388 (Ar-OH), 1527 (Ar=C=C str), 1685 (C=O str), 1352 (C-O str), 3071 (Ar-H). 1H NMR (400 MHz, CMSO, δ, TMS=0): δ = 11.90 (1H, s, 3-OH, Exchangeable with D₂O), 7.69 (1H, d, 5-H J= 8.02 Hz), 7.38 (1H, dd, 7-H, J=10.60 Hz), 7.20 (1H, dd, 6-H, J= 4.04 Hz), 7.09 (1H, d, 3'-H, J=8.5Hz), 6.99 (1H, d, 8-H, J= 8.04 Hz), 6.77 (1H, m, 4'-H), 5.58 (1H, d, 3-H, J= 12.56Hz), 5.52 (1H, d, 2-H, J= 8.63Hz), 13C NMR (400 MHz, δ, CDCl3, TMS=0): Aromatic Ring-A (152.85, 133.03, 130.31, 129.54, 119.58, 116.31), 187.38 (C=O), 155.67 (3-C), 140.59 (2-C), Aromatic Ring B (154.16, 142.68, 123.86, 112.73), TOF MS ES+ (m/z)= 229.5

2-(4-dimethylamino)phenyl-2,3-dihydro-3-hydroxy-7-methoxychromen-4-one (4b)

Yellow-brown, physical data is summarized in Table 3. IR (KBr, cm⁻¹): 3356 (Ar-OH), 1570 (Ar=C=C str), 1688 (C=O str), 1340 (C-O str), 2930 (H str), 3025 (Ar-H), 1247 (O-C), 1313, (C=N). 1H NMR (400 MHz, DMSO, δ, TMS=0): δ = 12.00 (1H, s, 3-OH, Exchangeable with D₂O), 7.62 (1H, d, 5-H J= 8.00 Hz), 7.35 (1H, dd, 7-H, J=7.68Hz), 7.25 (2H, m, 2'-H), 7.19 (2H, m, 3',5'-H), 7.03 (1H, d, 6-H, J= 3.05 Hz), 6.94 (1H, d, 8-H, J= 8.04 Hz), 5.60 (1H, d, 3-H, J= 12.56Hz), 5.53 (1H, d, 2-H, J= 8.65Hz), 13C NMR (400 MHz, δ, CDCl3, TMS=0): Aromatic Ring-A (153.25, 132.56, 131.81, 125.17, 121.49, 115.37), 187.49 (C=O), 153.46 (2-C), 145.59 (3-C), Aromatic Ring B (146.09, 138.79, 135.20, 135.91, 126.67, 126.37), TOF MS ES+ (m/z)= 275.5.
with water), 7.57 (1H, d, 5-H, J= 8.00 Hz), 7.24 (2H, m, 2',6'-H), 7.05 (2H, m, 3'-5'-H), 6.98 (1H, d, 6-H,J=7.76 Hz), 6.95 (1H, s, 8-H), 5.58(1H, d, 3-H, J=12.56Hz), 5.52 (1H,d, 2-H,J= 8.63Hz), 3.89(3Hs, 7-OCH3), 2.06 (6H, s,4'-Dimethyl aminno), 1^3C NMR (400 MHz, δ, CDCl3, TMS=0): Aromatic Ring-A (157.77, 152.34, 134.75, 134.74, 127.23, 127.23, 115.68, 115.60) 40.01, 39.98, N-(CH3)_2, TOF MS ES+ (m/z)= 314.

2-(4-chlorophenyl)-2,3-dihydro-3-hydroxy-7-methoxychromen-4-one (5b)

Yellow-brown, physical data is summarized in Table 3. IR (KBr, cm^{-1}): 3356 (Ar-OH), 1659 (C=O str), 1509 (C=N), 1309 (C-O str), 2939, (C-H str), 3090 (Ar-H), 7625 (C-Cl).

1H NMR (400 MHz, δ, CDCl3, TMS=0): δ= 12.11 (1H,s, 3-OH, Exchangeable with D2O), 7.68 (1H,d, 5-H,J= 8.00 Hz), 7.35 (2H, m, 2',6'-H), 7.26 (2H, m, 3'-5'-H). 7.03(1H,d, 6'-H,J= 8.04Hz), 6.98 (1H, s, 8-H), 5.58(1H, d, 3-H, J=12.56Hz), 5.52 (1H,d, 2-H,J= 8.63Hz), 3.88(3Hs,7-7-OCH3), 2.06 (6H, s,4'-Dimethyl aminno), 1^3C NMR (400 MHz, δ, CDCl3, TMS=0): Aromatic Ring-A (157.77, 152.34, 134.75, 134.74, 127.23, 127.23, 115.68, 115.60) 39.98, N-(CH3)_2, TOF MS ES+ (m/z)= 298.

2-(4-chlorophenyl)-2,3-dihydro-3-hydroxy-7-methylchromen-4-one (8b)

Yellow solid, physical data is summarized in Table 3. IR (KBr, cm^{-1}): 3395 (Ar-OH), 1565 (Ar=C=C str), 1688 (C=O str), 1324 (C-O str), 2960, (C-H str), 3070 (Ar-H), 776 (C=C). 1^H NMR (400 MHz, δ, CDCl3, TMS=0): δ= 11.98 (1H,s, 3-OH, Exchangeable with D2O), 7.71(1H,d, 5-H,J=8.00Hz), 7.31(2H, m, 2',6'-H), 188.87 (C=O), 141.36 (3-C), 155.99 (2-C), Aromatic Ring B (160.43, 145.67, 131.66, 131.56, 124.37, 124.29). TOF MS ES+ (m/z)= 305.

2-(furan-2-yl)-2,3-dihydro-3-hydroxy-6-methylchromen-4-one (9b)

Yellow-brown, physical data is summarized in Table 3. IR (KBr, cm^{-1}): 3356 (Ar-OH), 1509 (C=O str), 1309 (C-O str), 2939, (C-H str), 3090 (Ar-H), 7625 (C-Cl).

1H NMR (400 MHz, δ, CDCl3, TMS=0): δ= 12.01 (1H,s,3-OH, Exchangeable with D2O), 7.65(1H,d, 5-H,J=8.04Hz), 7.31(2H, m, 2',6'-H), 188.38 (C=O), 141.36 (3-C), 155.99 (2-C), Aromatic Ring B (160.43, 145.67, 131.66, 131.56, 124.37, 124.29). TOF MS ES+ (m/z)= 287.

2-(furan-2-yl)-2,3-dihydro-3-hydroxy-6-methylchromen-4-one (10b)

Yellow-brown, physical data is summarized in Table 3. IR (KBr, cm^{-1}): 3356 (Ar-OH), 1659 (C=O str), 1509 (C=O str), 2960, (C-H str), 3070 (Ar-H), 776 (C=C). 1^H NMR (400 MHz, δ, CDCl3, TMS=0): δ= 11.98 (1H,s, 3-OH, Exchangeable with D2O), 7.71(1H,d, 5-H,J=8.00Hz), 7.31(2H, m, 2',6'-H), 188.87 (C=O), 141.36 (3-C), 155.99 (2-C), Aromatic Ring B (160.43, 145.67, 131.66, 131.56, 124.37, 124.29). TOF MS ES+ (m/z)= 245.
6.97 (1H, d, 6-H, J= 8.23 Hz ), 6.74 (1H, d, 8-H, J=7.50 Hz), 5.68(1H, d, 2-H, J=12.56Hz), 5.58 (1H,d, 3-H, J= 8.63Hz),\(^1^H\) NMR (400 MHz, δ, CDCl₃, TMS=0):

Aromatic Ring-A (152.24, 132.12, 131. 87, 124.43, 122.49, 117.10)  178.88 (C=O), 148.32 (2-C), 138.35 (3-C), Aromatic Ring B (153.44, 147.89, 127.35, 127.36, 118.68, 118.68, ) 40.16, 40.17, (C-NO\(_2\)). TOF MS ES+ (m/z)= 282.

Physicochemical characterization of 3-methylflavanone analogues (1a-10a)

Physicochemical characterizations of 3-methylflavanone analogues are listed in Table 2.

Physicochemical characterization of 3-hydroxyflavanone analogues (1b-10b)

Physicochemical characterization of 3-hydroxyflavanone analogues are listed in Table 3.

Anti-leishmanial Activity

The anti-leishmanial activities of synthesized compounds were screened against promastigotes of \(L.\) donovani. Amphotericin B and Sodium Stibugluconate were used as standard drugs and exhibited 100 % inhibition. The results are presented in Table 4.

Docking Study

All the twenty compounds were docked into the active site of the PTR1 and the estimated free binding energy were presented in Table 5 and the interaction of most promising test compounds with receptors were illustrated in Figure 2 A and 2 B respectively.

DISCUSSION

All the selected 20 compounds were investigated for their activities against axenic \(L.\) donovani promastigotes, grown in simple media have been used as test parasite to screen potential antileishmanial agents and the simplicity of this system accounts for its wide popularity, and the results were expressed as % inhibition illustrated in Table 4 with few exceptions, all 3- methyl and 3-hdroxy flavanone showed significant antileishmanial activity. The test compounds such as 4b (95%), 10b (93%), 5b (92%), 6b (91%), and 3a (90%) being the most potent. Their % inhibitions were almost comparable to that of Amphotericin B and Sodium Stibugluconate the antileishmanial drug used in the clinic. Starting with the flavanone, the insertion of a single OH group at the benzo-γ- chromone (at-3-position) portion of the flavone structure have a notable influence, but insertion of one more OH / OCH\(_3\) on ring A and B functions significantly enhanced the leishmanicidal potential. Particularly important positions were C-5, C-7 on ring A. Hydroxylation on ring B had some impact on the activity, but a clear SAR could not be observed. Further, compounds such as 9b (87%), 4a (84%), 5a (82%), 9a (81%), 3b (81%), 7a (80%) and 1b (80%) have shown appreciable leishmanicidal activity as compared with that of the standard drugs, as most of the test compounds were in conjugation with hydroxyl or electron donating groups except nitro group. However, compounds such as 8b (72%), 8a (71%), 10a (68%) and 2b (62%) exhibited moderate activity against promastigotes of \(L.\) donovani, as these compounds were having either mild...
electron withdrawing group or without electronic effect on main nucleus. The presence of strong electron withdrawing group on the entire structure significantly reduced the leishmanicidal activity as observed in compounds, such as 1a (12%), 2a (15%), 6a (18%) see Table 1. Based on the information obtained, it is difficult to decipher empirical SARs among the test compounds investigated in the current study. It is quite noteworthy in the finding that majority of leishmanicidal compounds have a typical 3-hydroxy flavanone structure with one or more substitution by hydroxyl or methoxy groups or electron donating group(s). Furthermore, drug receptor interaction was carried out to validate our findings. The ligands were ranked according to docking score /estimated free energy of binding. The free energy of binding of ligands was in the range between -3.15 to -7.84 Kcal/mole. Top ranked compound (4b) and (10b) with -7.84 and -7.77 Kcal/mole free energy of binding, respectively, were in correlation with wet lab experiments. The protein ligand analysis also has shown strong interactions with target protein and had five hydrogen bond interaction in (4b) and four hydrogen bond interaction in (10b). The residues involved in hydrogen bond interaction were Arg 17, Lys 199, Asn109, Asp 181 and Phe113 in (4b) (Figure 2A) and Lys 16, Ser 40, Asn 140, and Arg 17 in (10b) (Figure 2B) with the active site of PTR1. Virtual screening of 100 3-substituted flavanone analogues library resulted in the identification of 20 compounds. Out of 100 compounds, 20 compounds with the lowest estimated free energy of binding were selected for synthesis. Among synthesized compounds top ranked compounds (4b) and (10b) according to estimated free energy of binding (-7.84 and -7.77 Kcal/mole respectively), also had promising anti-leishmanial activity in wet lab experiments. The excellent interactions of PTR1 with all five top ranked compounds (4b), (10b), (5b), (6b) and (3a) indicated a high degree of coherent relationship between *in silico* approach and *in vitro* studies. An *in-vitro* promising anti-leishmanial activity of the compounds demands further *in-vitro* and clinical studies and these compounds might find an important place in the new array of molecules targeting PTR1 dependent biological functions as anti-leishmanial agents. However, there are some limitations of using promastigotes for *in vitro* anti-leishmanial study; the metabolism and ecology of promastigote differ so widely from those of amastigote that screening data obtained from *in vitro* test with promastigote have very little value in animals. Any condition which reduces leishmanicidal action *in vitro* is lower temperature (24°C) at which the culture normally grows, as opposed to the *in vivo* temperature of 37°C. The promastigote in culture at 37°C will survive but not multiply. Further, the promastigote culture represents an artificial situation and is not always showing a significant value for drug screening.

**CONCLUSION**

This study reports on the *in vitro* anti-leishmanial activity of a number of flavanone analogues synthesized from

<table>
<thead>
<tr>
<th>Compounds</th>
<th>Estimated free energy of binding (Kcal/mol)</th>
</tr>
</thead>
<tbody>
<tr>
<td>4b</td>
<td>-7.84</td>
</tr>
<tr>
<td>10b</td>
<td>-7.77</td>
</tr>
<tr>
<td>5b</td>
<td>-7.76</td>
</tr>
<tr>
<td>6b</td>
<td>-7.62</td>
</tr>
<tr>
<td>3a</td>
<td>-7.56</td>
</tr>
<tr>
<td>9b</td>
<td>-6.95</td>
</tr>
<tr>
<td>4a</td>
<td>-6.51</td>
</tr>
<tr>
<td>5a</td>
<td>-6.40</td>
</tr>
<tr>
<td>9a</td>
<td>-6.16</td>
</tr>
<tr>
<td>3b</td>
<td>-6.01</td>
</tr>
<tr>
<td>7b</td>
<td>-5.73</td>
</tr>
<tr>
<td>1b</td>
<td>-5.61</td>
</tr>
<tr>
<td>8b</td>
<td>-5.54</td>
</tr>
<tr>
<td>8a</td>
<td>-4.78</td>
</tr>
<tr>
<td>2b</td>
<td>-4.06</td>
</tr>
<tr>
<td>1a</td>
<td>-3.66</td>
</tr>
<tr>
<td>2a</td>
<td>-3.52</td>
</tr>
<tr>
<td>6a</td>
<td>-3.39</td>
</tr>
<tr>
<td>7a</td>
<td>-3.21</td>
</tr>
<tr>
<td>10a</td>
<td>-3.15</td>
</tr>
</tbody>
</table>

3D structures of PTR1, (from 2XOX), was used for virtual screening.
2'-hydroxypropiohenones, 2'-hydroxyacetophenone, and substituted benzaldehyde. The study was prompted by the need of novel, efficacious and cost-effective medicines against this parasitic disease. After elucidating the chemical structure of the newly synthesized flavanone analogues, they were evaluated for their potential leishmanicidal activity against cultured *L. (*L.)* donovani* promastigotes. These experiments revealed promising anti-leishmanial activity of particularly compound 10b, which also showed the best docking into the active site of pteridine reductase.

**ACKNOWLEDGEMENT**

The authors thank to Narayan Institute of Pharmacy to facilitate part of the research work. We must thank to SAIF, Auvtar Singh and Manish Kumar, Panjab University, Chandigarh, (India) to get spectral data of 1HNMR, C13NMR, and Mass Spectrometry.

**CONFLICTS OF INTEREST**

The authors declare no conflict of interest.

**ABBREVIATIONS USED**

PTR1: Pteridine Reductase; DHFR: Dihydrofolate reductase; SARs: Structure Activity Relationships; DMSO: Dimethyl Sulphoxide; TLC: Thin Layer Chromatography; FBS: Fetal Bovine Serum; ELISA: Enzyme Linked Immunosorbent Assay; MTT: [3-(4,5-dimethylthiazol-2-yl)-2,5 diphenyl tetrazolium; TMS: Tetra Methyl Silane.

**REFERENCES**

SUMMARY

Leishmaniasis is a vector-borne disease and considered as one of the world’s most neglected diseases, is endemic in 88 countries with more than 350 million people at risk. The World Health Organization emphasized that plants used in traditional medicine and their synthetic analogues should primarily be investigated against leishmaniasis. The proposed work of in silico study exhibits the high degree of statistical significance and good predictive ability. The information obtained from this study could provide vital information for future development of potent anti-leishmanial agents as pteridine reductase inhibitors. A series of flavanones with various structural features has been synthesized and evaluated for in vitro anti-leishmanial activity; the potent compounds should be further evaluated in vivo anti-leishmanial efficacy in L. donovani / hamster model as future target. Herein, we have been able to document a comprehensive assessment of anti-leishmanial activity and structure-activity relationship analyses of promising flavanone analogues.

About Authors

Mohan Lal Verma is Principal and Director cum Technical Advisor in Narayan Medical College & Hospital, Jamuhar 821305, Sasaram Bihar. Earlier he served as Principal in Patna Medical College & Hospital as well as in Nalanda Medical College & Hospital. He has done MBBS, MD in Pharmacology. He has been awarded so many titles in the field of Medical Science. His work interest is multi-disciplinary research focus on development of new drugs related to leishmaniasis. He has a huge experience to handle academic and administrative profile of the colleges and University. Moreover, Dr. M.L. Verma published 6 books and more than 100 research and review papers in peer-reviewed journals. He is chairman of the various committees or advisory board of many government Universities.

Afroze Alam is specialized in Pharmaceutical Chemistry at M. Pharma level and did his Ph.D. in Pharmaceutical/Medicinal and Phytochemistry. He is working as Associate professor at Narayan Institute of Pharmacy, Jamuhar, Sasaram-821305, Rohtas Bihar, India. His main area of work is drug design and the natural product chemistry of anticancer, antibacterial, antileishmanial, antimalarial and anti-diabetic molecules. Moreover, his research interest includes nanobiomedicines. He published more than 30 papers in internationally recognized high impact biomedical and chemistry journals.

Mr. Kamlesh Naik did his post-graduation (2007) in Pharmaceutical Sciences from Nandha College of Pharmacy Erode, Chennai. Presently, he is working as a Ph.D. Research fellow at Pharmaceutical research lab, Department of Pharmaceutical Chemistry, Nandha College of Pharmacy Erode, Tamil Nadu, India. Her research work has focus predominantly on the flavonoids nucleus and its associated pharmacological activities such as anti-oxidant, hepatoprotective, anti-inflammatory and cytotoxic activity. He is going to face defense or Viva –Voce of his PhD work in April, 2018. He published 12 research and review article in reputed journals.
Mr. Mahfoozur-Rahman is currently working as a Pharmaceutics faculty at Department of Pharmaceutical Sciences, Faculty of Health Sciences, Sam Higginbottom Institute of Agriculture, Technology and Sciences (SHIATS), Allahabad, INDIA, Uttar Pradesh, India. He is a professional Pharmacy graduate; did his M.Pharm and pursuing Ph.D. in Pharmaceutical Science (specialization_ Pharmaceutics). He was awarded with senior fellowship from Council of Scientific and Industrial Research (CSIR), India. His research interests involve drug delivery and targeting, nanoparticulates and vesicular systems, nanotoxicology and pharmaceutical analysis. Mr. Mahfoozur-Rahman has published more than 50 papers including book chapters in peer-reviewed journals of high impact.

Shailendra Kumar did his M.Pharmacy from Manipal College of Pharmaceutical Sciences, Udupi Karnataka, India, He has been awarded PhD from Magadh University, Bodh Gaya, Presently he is working as Principal, Govt. Pharmacy Institute, Agam Kuan Patna Bihar-India. He is the members of many government regulatory bodies. He is specialized in Pharmaceutical Chemistry. His research area is profoundly related to the isolation of bioactive molecules from plant sources. He greatly contributes to the development of Pharmacy institute in Bihar. He published more than 15 papers in reputed journal with high impact.

Cite this article: Alam A, Pandit V, Kumar S, Naik KK, Rahman M, Verma ML. Anti-Leishmanial Activity of Flavanone Analogues Targeting Pteridine Reductase. Indian J of Pharmaceutical Education and Research. 2018;52(3)480-91.