Introduction

The small molecules that target microtubules is an attractive and an active area in cancer drug discovery. These molecules bind to the tubulin, an α, β- heterodimer and disrupt the dynamics of microtubule. Microtubule targeting agents can be classified into two categories. Microtubule stabilizing agents such as paclitaxel, docetaxel, epothilones, and discodermolide binds to the tubulin polymer and stabilize the microtubules. Microtubule destabilizing agents such as vinca alkaloids, colchicine and combretastatins binds to tubulin dimers and cause destabilization. The equilibrium between tubulin and microtubule is altered, which results in disruption of mitotic spindle. This effects a critical transition in the cell cycle, leading to cell death. Out of these different pockets on tubulin, colchicine is a significant source of inspiration for the design of new drugs as the colchicine binding site inhibitors binds with high affinity at the interface of α and β-tubulin. These are effective against multidrug mechanisms but however, the potential clinical applications of colchicine site tubulin inhibitors have been nullified by the significant toxicities against the normal cells, low solubility, and low bioavailability.

In an endeavor to find the potential chemotherapeutic agents, a substantial effort has been devoted on the design and development of heterocycles embedded small organic molecules. The thiazolidinedione ring has been used as scaffold to develop novel class of anticancer agents with a wide range of cytotoxicity against many human cancer cells. From the literature, it was observed that numerous compounds containing thiazolidine 2,4-dione and rhodanine have been reported as potential anticancer agents. Also several chromene derivatives were reported as effective antitumor agents; particularly 2-Amino-3-cyano-4-aryl-4H-chromene derivatives are promising apoptosis inducing agents. The presence of the 4-aryl moiety, the 3-cyano group, and the 2-amino functionality were essential features for the anticancer activity of these chromene analogues.

Patil and co-workers have reported that naphthalene tethered 4-substituted-4H-chromene (Figure 2) was most active against all the cell lines and showed most potent tubulin polymerization inhibitory activity. Inspired by these results, we hereby report the design and synthesis of azolidinedione/thiazolidinediones tethered benzo[f]chromene analogues and their in silico evaluation as tubulin inhibitors.
Results and discussions

We are actively engaged in accessing hybrid heterocyclic scaffolds through multicomponent reactions by integrating the concepts of diversity oriented synthesis and bioisosterism particularly build/couple/pair strategy. Recently we have reported the synthesis of double ring replacement analogues of 4-heterocycle substituted 4H-chromenes. The synthesis of azolidinedione/thiazolidinediones tethered benzo[f]chromene analogues were carried out as outlined in Figure 3 & Figure 4. We have initiated our investigation by a model three component reaction between 1-formyl-2-hydroxynaphthalene (1), hydantoin (2) and malononitrile (3) in water, in the presence of piperidine, as a base. The reaction yielded benzo[f]chromene derivatives in 30 min with 94% yield. Later the reaction was optimized using different solvents and bases. It was observed that the best results were obtained when the reaction was conducted in water and piperidine as a base for 30min at room temperature. The product was confirmed by 1H, 13C NMR and HRMS spectra. A set of compounds (4a-d) were synthesized with different azolidinedione and thiazolidinedione derivatives (2a-d). The results were summarized in Figure 3.

Docking studies

After establishing a three component reaction to afford 4-heterocycle-4H benzo[f]chromenes, We have performed a molecular docking to investigate the possible interactions and binding conformation for this set of compounds at the colchicine binding site of tubulin, which may give a suggestion about their proposed mechanism of action. All the stereoisomers of the synthesized benzo[f]chromene analogues were docked at the colchicine binding site of tubulin. UCSF Chimera visualisation software was used to visualize the results. The overlap of RS isomer of 4a with the colchicine at the binding site of tubulin is displayed (Figure 5).

Table 1 Predicted binding affinities of synthesized compounds at the colchicine binding site of tubulin

<table>
<thead>
<tr>
<th>S No.</th>
<th>Compound</th>
<th>Binding affinity (Kcal/mol)</th>
<th>S No.</th>
<th>Compound</th>
<th>Binding affinity (Kcal/mol)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>4a-R,S</td>
<td>-9.9</td>
<td>9</td>
<td>4c-R,S</td>
<td>-9</td>
</tr>
<tr>
<td>2</td>
<td>4a-S,R</td>
<td>-9.7</td>
<td>10</td>
<td>4c-S,R</td>
<td>-9</td>
</tr>
<tr>
<td>3</td>
<td>4a-R,R</td>
<td>-9.7</td>
<td>11</td>
<td>4c-R,R</td>
<td>-9.4</td>
</tr>
<tr>
<td>4</td>
<td>4a-S,S</td>
<td>-9.5</td>
<td>12</td>
<td>4c-S,S</td>
<td>-8.9</td>
</tr>
<tr>
<td>5</td>
<td>4b-R,S</td>
<td>-9.5</td>
<td>13</td>
<td>4d-R,S</td>
<td>-9.5</td>
</tr>
<tr>
<td>6</td>
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<td>-9.5</td>
<td>14</td>
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<tr>
<td>7</td>
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<td>15</td>
<td>4d-R,R</td>
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<tr>
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<td>4b-S,S</td>
<td>-9.5</td>
<td>16</td>
<td>4d-S,S</td>
<td>-9.5</td>
</tr>
</tbody>
</table>

Note: R=R configuration; S= S configuration.

Figure 5 A view of the binding conformation of 4a-R,S (yellow) overlapping with colchicine (pink) at the colchicine binding site of tubulin. Tubulin is displayed as a flat ribbon with α-tubulin coloured red and β-tubulin coloured green.
Design and synthesis of Azolidinedione/Thiazolidinediones tethered benzo[f]chromene derivatives and their in silico evaluation as tubulin inhibitors

Experimental section

Chemistry and materials

Commercial reagents were used without further purification. Melting points were recorded on a Veego capillary melting point apparatus and were uncorrected. For compounds 1H NMR (400MHz, DMSO-d6) and 13C NMR (100MHz, DMSO-d6) spectra were recorded in DMSO-d6 on a Bruker 400MHz spectrometer using tetramethylsilane (TMS, δ=0) as an internal standard at room temperature. Mass spectra were recorded on Agilent 1200 LC/MS-6110 mass spectrometer.

General procedure for synthesis of 3-amino-1-(2,5-dioxoimidazolidin-4-yl)-1H-benzo[f]chromene-2-carbonitrile 4a

A mixture of 1-formyl-2-hydroxynapthalene (1mmol), hydantoin (1mmol) and malononitrile (1mmol) were taken in water (5ml) in a round bottom flask and the reaction mixture was allowed to stir at room temperature until the starting materials disappear on tlc. The reaction conditions. Docking results showed that all the four synthesized compounds bind at colchicine binding site of tubulin. In conclusion, we have designed and synthesized azolidinedione/thiazolidinediones tethered benzochromene analogues under greener reaction conditions. Docking results showed that all the four synthesized compounds bind at colchicine binding site of tubulin and exhibited good predicted binding affinities.

Yield (94%); mp 252-255°C; 1H NMR (400MHz, DMSO-d6) δ 11.69 (s, NH), 9.80 (s, NH), 7.91-7.99 (m, 4H), 7.49-7.54 (m, 2H), 7.12 (s, NH), 4.66 (d, J=4Hz, 1H), 4.38 (d, J=4Hz, 1H) ppm; δ 13C NMR (100MHz, DMSO-d6) δ 183.7, 174.29, 163.39, 148.20, 130.75, 129.49, 128.81, 127.55, 125.97, 125.07, 122.08, 121.83, 119.98, 112.54, 65.20, 47.22, 35.13 ppm. HRMS (ESI) Calcd for C17H12N2O3 [M+Na] 320.09amu, found 320.10amu.

3-amino-1-(5-oxo-2-thioxoimidazolidin-4-yl)-1H-benzo[f]chromene-2-carbonitrile 4b

Yield (86%); mp 260-265 °C; 1H NMR (400MHz, DMSO-d6) δ 7.8-8.3 (m, 3H), 7.89 (d, J=4Hz, 1H), 7.6-7.70 (m, 1H), 7.5-7.54 (m, 1H), 7.21 (d, J=8.8 Hz, 1H), 7.05 (s, NH2), 4.94 (d, J=2.4 Hz, 1H), 4.48 (d, J=2Hz, 1H)ppm; δ 13C NMR (100MHz, DMSO-d6) δ 173.16, 164.15, 158.16, 145.97, 145.53, 144.03, 131.22, 127.84, 119.87, 62.77, 58.12, 46.70, 34.83, 18.65ppm. HRMS (ESI) Calcd for C15H11N2O3S [M+Na] 353.03amu, found 353.03amu.

3-amino-1-(4-oxo-2-thiothiazolidin-5-yl)-1H-benzo[f]chromene-2-carbonitrile 4c

Yield (91%); mp 256-260°C; 1H NMR (400MHz, DMSO-d6) δ 11.85 (s, NH), 7.91-7.98 (m, 4H), 7.5-7.58 (m, 2H), 7.12 (s, NH2), 4.66 (d, J=4Hz, 1H), 4.38 (d, J=4Hz, 1H) ppm; δ 13C NMR (100MHz, DMSO-d6) δ 181.70, 170.29, 163.43, 147.20, 130.75, 129.37, 129.0, 128.95, 127.83, 125.22, 121.83, 120.19, 116.78, 115.82, 66.19, 49.34, 35.40ppm. HRMS (ESI) Calcd for C17H12N2O3S [M+Na] 337.05 amu, found 337.05amu.

3-amino-1-(2,4-dioxothiazolidin-5-yl)-1H-benzo[f]chromene-2-carbonitrile 4d

Yield (86%); mp 260-265 °C; 1H NMR (400MHz, DMSO-d6) δ 11.85 (s, NH), 7.9-7.98 (m, 4H), 7.5-7.58 (m, 2H), 7.12 (s, NH2), 4.66 (d, J=4Hz, 1H), 4.38 (d, J=4Hz, 1H) ppm; δ 13C NMR (100MHz, DMSO-d6) δ 181.70, 170.29, 163.43, 147.20, 130.75, 129.37, 129.0, 128.95, 127.83, 125.22, 121.85, 120.19, 116.78, 115.82, 66.19, 49.34, 35.40ppm. HRMS (ESI) Calcd for C17H12N2O3S [M+Na] 337.05 amu, found 337.06 amu.

Conclusion

In conclusion, we have designed and synthesized azolidinedione/thiazolidinediones tethered benzochromene analogues under greener reaction conditions. Docking results showed that all the four synthesized compounds bind at colchicine binding site of tubulin and exhibited good predicted binding affinities. In vitro studies of the synthesized compounds are in progress and will be reported in future publications.

Acknowledgements

Dr. G. Vasuki thank DST-SERB (SB/S1/OC-37/2014), Govt. of India, for financial support, and TSK thank CSIR for JRF and SRF (File No. 09/559(102)/2012-EMR-I). Authors also thank Central Instrumentation Facility (CIF), Pondicherry University, for high-resolution NMR and the Department of Chemistry for ESI-HRMS and single-crystal XRD.

Conflict of interest

Author declares that there is no conflict of interest.
Design and synthesis of Azolidinedione/Thiazolidinediones tethered benzo[f]chromene derivatives and their in silico evaluation as tubulin inhibitors

References


