Synthesis and biological evaluation of some innovative coumarin derivatives containing thiazolidin-4-one ring

RamaGanesh C K, Yadav D Bodke* & Venkatesh K B
Department of Studies and Research in Industrial Chemistry, Jnana Sahyadri, Kuvempu University, Shankaraghatta 577 451, India
Email: ydbodke@gmail.com

Received 25 May 2009; accepted (revised) 27 April 2010

The reaction of ethyl 2-oxo-2H-chromene-3-carboxylate with hydrazine followed by condensation of the resulting hydrazone with different aromatic aldehydes give the corresponding Schiff bases 5a-e. Reaction of these Schiff bases with mercaptoacetic acid furnishes the target thiazolidinone molecules 6a-e. The newly synthesized compounds have been screened for antibacterial and analgesic activities.

Keywords: Salicylaldehyde, biological agent, Schiff base, mercaptoacetic acid, thiazolidinone

The synthesis of coumarins and their derivatives has attracted considerable attention from organic and medicinal chemists for many years as large number of natural products contain this heterocyclic nucleus. They are widely used as additives in food, perfumes, cosmetics, pharmaceuticals, optical brighteners, dispersed fluorescent and laser dyes. A considerable number of natural and synthetic coumarin derivatives display pharmacological properties with a wide range of activity and others are useful for optical applications. Thus the synthesis of this heterocyclic nucleus is of much interest. 4-Thiazolidinones have been reported to show a broad spectrum of biological activities and a wide range of pharmacological activities such as hypnotic-sedative, analgesic activity, anticonvulsant, antifungal, antibacterial and antitubercular activity against M. tuberculosis H37Rv. β-Lactamase is generally considered to be responsible for microbial resistance against a broad spectrum of β-lactam antibiotics. In view of the pharmacological properties of 4-thiazolidinones, we were interested in synthesizing several new compounds bearing coumarin nucleus, attached to 4-thiazolidinone moieties.

Results and Discussion

As part of our aim to search for biologically active heterocycles containing sulfur and nitrogen, we report in this paper synthesis of a series of 2-oxo-N-(4-oxo-2-substituted phenyl-1,3-thiazolidin-3-yl)-2H-chromene-3-carboxamide 6a-e and on estimation of their biological properties. For this purpose, ethyl 2-oxo-2H-chromene-3-carboxylate 3 was a key intermediate which was originally prepared by the reaction of salicylaldehyde with diethyl malonate in the presence of catalytic amount of piperidine. Applying hydrazinolysis on ethyl 2-oxo-2H-chromene-3-carboxylate with hydrazine hydrate in methanol at room temperature, 2-oxo-2H-chromene-3-carboxyhydrazide 4 was obtained in good yield. The carbohydrazide 4 was then condensed with different aromatic aldehydes in methanol to furnish the corresponding Schiff’s bases, 5a-e. The structures of the products 5a-e were confirmed from their analytical and spectral data.

The reaction of Schiff bases 5a-e with mercaptoacetic acid in the presence of catalytic amount of anhydrous zinc chloride in DMF for about 8 hr furnished the compounds 6a-e (Scheme I) whose structures were assigned on the basis of spectral data and elemental analysis.

Biological studies

Antibacterial activity

The newly synthesized compounds were screened for their antibacterial activity by the cup plate method. The in vitro antibacterial activity was carried out against 24 hr old culture for four bacteria and four fungal organisms. The bacteria used were Staphylococcus aureus, Bacillus subtilis, Klebsiella pneumoniae and Escherichia coli. The compounds were tested at a concentration of 0.001 mol/mL in DMF against all the organisms. Ciprofloxacin (0.001 mol/mL) was used as standard for the comparison of antibacterial activity. The zone of inhibition was compared with the standard drug after 24 hr of incubation at 37°C.

Among the compounds tested for antibacterial activity (Table I), compound 6a showed high activity against S. aureus, B. subtilis and moderate activity against K. pneumoniae and E. coli. Compounds 6b and 6c have exhibited moderate activity against S. aureus, B. subtilis and weak activity against K. pneumoniae and E. coli.
**Analgesic activity**

The analgesic activity of the newly synthesized compounds was tested using adult Swiss albino mice (6 animals per group) by the abdominal constriction method\(^{21}\). The mice were housed individually in polypropylene cages with paddy husk as bedding. The animals were maintained at a temperature of 25-27°C and relative humidity of 30-70%. Male Swiss albino mice were procured from Virus Diagnostic Laboratory, Shivamoga. Five groups of six mice each (25-30 g) were selected and 0.6% acetic acid (dose 10 mL/kg) was injected intraperitoneally. The number of writhes was counted for 20 min, after 5 min of the injection of acetic acid to each mouse. This reading was taken as a control. Next day the same groups of mice were used for evaluating analgesic activity. Each group was administered orally with the suspension of the test compound in 0.1% Tween-80 solution (100 mg/kg body weight), 1 hr before injection of acetic acid. After 5 min, the mice were observed for the number of writhes for the duration of 20 min. The mean value for each group was calculated and compared with the control. Acetyl salicylic acid was used as standard for the comparison of analgesic activity. The percent protection was calculated using the formula \((1-V_c/V_t) \times 100\); \(V_t\) = mean number of writhing in test animals, \(V_c\) = mean number of writhing in control.

Among the compounds tested for analgesic activity (Table II), the compounds 6a and 6c exhibit good activity and the remaining compounds showed moderate to weak activity as compared with the standard drug.
Table I — Antibacterial activity of synthesized compounds

<table>
<thead>
<tr>
<th>Compd</th>
<th>Gram Positive</th>
<th>Gram Negative</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S. aureus</td>
<td>B. subtilis</td>
</tr>
<tr>
<td>6a</td>
<td>16</td>
<td>20</td>
</tr>
<tr>
<td>6b</td>
<td>14</td>
<td>12</td>
</tr>
<tr>
<td>6c</td>
<td>12</td>
<td>13</td>
</tr>
<tr>
<td>6d</td>
<td>07</td>
<td>05</td>
</tr>
<tr>
<td>6e</td>
<td>11</td>
<td>09</td>
</tr>
<tr>
<td>Standard</td>
<td>22.89</td>
<td>28.72</td>
</tr>
<tr>
<td>Control</td>
<td>0.89</td>
<td>0.94</td>
</tr>
</tbody>
</table>

Key for interpretation:
- <8mm: Inactive
- 8-12mm: Weakly active
- 13-15mm: Moderately active
- >16 mm: Highly active

Table II — Analgesic activity of synthesized compounds

<table>
<thead>
<tr>
<th>Compd</th>
<th>Dose mg/kg</th>
<th>Before administration of drug</th>
<th>After administration of drug</th>
<th>Protection %</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>100</td>
<td>22.83±2.13</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>Standard</td>
<td>100</td>
<td>24.00±1.41</td>
<td>11.00±1.26</td>
<td>54.20</td>
</tr>
<tr>
<td>(Aspirin)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>6a</td>
<td>100</td>
<td>21.33±0.55</td>
<td>13.16±0.30</td>
<td>37.80</td>
</tr>
<tr>
<td>6b</td>
<td>100</td>
<td>18.65±0.53</td>
<td>14.83±1.01</td>
<td>20.48</td>
</tr>
<tr>
<td>6c</td>
<td>100</td>
<td>22.66±0.49</td>
<td>17.00±0.77</td>
<td>24.97</td>
</tr>
<tr>
<td>6d</td>
<td>100</td>
<td>23.02±0.60</td>
<td>19.33±0.47</td>
<td>16.02</td>
</tr>
<tr>
<td>6e</td>
<td>100</td>
<td>25.50±0.51</td>
<td>22.00±0.85</td>
<td>13.72</td>
</tr>
</tbody>
</table>

Values are in mean ± SEM.

Index for analgesic activity study

Method: Acetic acid induced writhing

(Acetic acid-0.6% concentration)

Animal: Albino mice

No. of animals per group: 6(25-30g)

Route of administration: IP (Intraperitoneally)

Standard drug used: Acetyl salicylic acid (Aspirin)

Experimental Section

The melting points were determined in an open capillary tube and are uncorrected. The purity of the compounds was checked by TLC on silica gel and was purified by column chromatography. $^1$H NMR spectra (300 MHz) were recorded on a Bruker Supercon FT NMR instrument using TMS as an internal standard and chemical shifts are expressed in δ units. IR spectra were recorded on a Perkin-Elmer 157 infrared spectrophotometer and mass spectra were recorded on a Jeol JMS-D 300 mass spectrometer.

Synthesis of ethyl 2-oxo-2H-chromene-3-carboxylate 3

To the solution of salicylaldehyde 1 (0.05 mole, 6 mL) and diethyl malonate (0.05 mole, 7 mL) in absolute methanol (15 mL) was added 0.1 mL piperidine and it was refluxed on water-bath for 5-6 hr. Completion of reaction was judged by TLC. After cooling, the yellow solid separated was collected by filtration and recrystallized from ethanol. m.p. 80-82°C; yield 90%. IR (KBr): 1650 and 1759 (C=O) cm$^{-1}$; $^1$H NMR (300 MHz, DMSO-$_d_6$): δ 1.32 (s, 3H); 4.29 (q, 2H); 7.95-7.40 (m, 5H); MS: m/z 218 (M$^+$).

Synthesis of 2-oxo-2H-chromene-3-carbohydrazide 4

An intimate mixture of ethyl 2-oxo-2H-chromene-3-carboxylate 3 (0.01 mole, 1.9 g) and hydrazine hydrate (0.02 mole, 2 mL) in absolute methanol was refluxed on water-bath for 5-6 hr. Completion of reaction was judged by TLC. After cooling, the reaction-mixture was poured into ice cold water. The solid, thus separated, was collected by filtration, dried and recrystallized from ethanol. m.p. 206-209ºC, yield 80%. IR (KBr): 1652, 1710 and 3043-3271 cm$^{-1}$; $^1$H NMR(300 MHz, DMSO-$_d_6$): δ 5.71 (s, 2H); 8.99 (s, 1H); 7.68-6.90 (m, 5H).

Synthesis of 2-oxo-N’-[substituted phenylmethylidene]-2H-chromene-3-carbohydrazides 5a-e

General procedure

An equimolar mixture of the carbohydrazide 4 and an aromatic aldehyde was refluxed on water-bath for 5-6 hr. The completion of reaction was judged by TLC. After keeping the mixture overnight, it was poured into ice cold water. The solid, thus separated, was collected by filtration, dried and recrystallized from ethanol. The products obtained were identified as 2-oxo-N’-[substituted phenylmethylidene]-2H-chromene-3-carbohydrazides 5a-e.

5a: Pale yellow solid, crystallized from ethanol, yield 82 %, m.p. 205-207ºC, IR (KBr): 1620, 1676, 1690 and 3400 cm$^{-1}$; $^1$H NMR (300 MHz, DMSO-$_d_6$): δ 6.55 (s, 1H); 8.01-6.90 (m, 10H) and 8.99 (s, 1H).

5b: Yellow solid, crystallized from ethanol, yield 65%, m.p. 194-96°C, IR (KBr): 1600, 1650, 1700 and 3370 cm$^{-1}$; $^1$H NMR (300 MHz, DMSO-$_d_6$): δ 3.60 (s,
3H), 6.58 (s, 1H), 8.07-6.92 (m, 9H) and 8.90 (s, 1H).  
5c: Yellow solid, crystallized from ethanol, yield 81%, m.p. 216-18°C, IR (KBr): 1628, 1668, 1680 and 3386 cm\(^{-1}\); \(^1\)H NMR (300 MHz, DMF-d\(_6\)): δ 6.44 (s, 1H), 8.12-7.01 (m, 9H) and 8.84 (s, 1H).  
5d: Yellow solid, crystallized from ethanol, yield 72%, m.p. 218-20°C, IR (KBr): 1616, 1668, 1680 and 3394 cm\(^{-1}\); \(^1\)H NMR (300 MHz, DMF-d\(_6\)): δ 6.56 (s, 1H), 8.08-7.00 (m, 9H), 8.91 (s, 1H) and 11.12 (s, 1H).  
5e: Yellow solid, crystallized from ethanol, yield 61%, m.p. 200-203°C IR (KBr): 1630, 1676, 1690 and 3395 cm\(^{-1}\); \(^1\)H NMR (300 MHz, DMF-d\(_6\)): δ 1.36 (s, 3H), 8.79 (s, 1H), 8.11-6.91 (m, 10H) and 6.52 (s, 1H).

**Synthesis of 2-oxo-N-(4-oxo-2-substituted phenyl-1,3-thiazolidin-3-yl)-2H-chromene-3-carboxamides, 6a-e**

**General procedure**

Schiff’s bases 5a-e (0.001 mole) were refluxed with mercaptoacetic acid (0.001 mole, 0.3 mL) in the presence of a catalytic amount of anhydrous ZnCl\(_2\) in DMF (15 mL) for 8 hr. The mixture was then cooled and poured into crushed ice. The product separated was filtered, dried and recrystallized from ethanol in hot condition to give 6a-e.  
6a: Light yellow solid, crystallized from ethanol, m.p. 248-52°C, yield 75%, IR (KBr): 1590, 1680, 1710 and 3375 cm\(^{-1}\); \(^1\)H NMR (300 MHz, DMF-d\(_6\)): δ 3.70 (s, 2H), 3.78 (s, 1H), 8.99 (s, 1H) and 7.90-6.9 (m, 10H); MS: m/z 366 (M\(^+\)).  
6b: Pale yellow solid, crystallized from ethanol, yield 70%, m.p. 185-87°C; IR (KBr): 1600, 1680, 1722 and 3324 cm\(^{-1}\); \(^1\)H NMR (300 MHz, DMF-d\(_6\)): δ 3.65 (s, 3H), 3.75 (s, 2H), 3.80 (s, 1H), 11.12 (s, 1H) and 8.60-7.60 (m, 9H); MS: m/z 396 (M\(^+\)).  
6c: Pale yellow solid, crystallized from ethanol, yield 70%, m.p. 215-18°C; IR (KBr): 1590, 1650, 1725 and 3424 cm\(^{-1}\); \(^1\)H NMR (300 MHz, DMF-d\(_6\)): δ 3.65 (s, 2H), 3.72 (s, 1H), 9.51 (s, 1H) and 8.50-7.65 (m, 9H); MS: m/z 400 (M\(^+\)); m/z 402 (M+2)\(^+\).  
6d: Pale yellow solid, crystallized from ethanol, yield 68%, m.p. 236-39°C; IR (KBr): 1610, 1680, 1700, 3390 and 3450 cm\(^{-1}\); \(^1\)H NMR (300 MHz, DMF-d\(_6\)): δ 3.50 (s, 2H), 3.72 (s, 1H), 8.00 (1H), 10.11 (s, 1H) and 8.90-7.9 (m, 9H); MS: m/z 382 (M\(^+\)).  
6e: Pale yellow solid, crystallized from ethanol, yield 68%, m.p. 191-93°C; IR (KBr): 1600, 1670, 1720 and 3390 cm\(^{-1}\); \(^1\)H NMR (300 MHz, DMF-d\(_6\)): δ 1.05 (s, 3H), 3.60 (s, 2H), 3.70 (s, 1H), 8.0 (s, 1H) and 8.90-7.9 (m, 9H); MS: m/z 380 (M\(^+\)).

**Acknowledgement**

The authors thank Indian Institute of Science, Bangalore and Karnataka University Dharwad for providing spectral data. One of the authors (C.K.R) thanks to Kuvempu University for awarding a Research Fellowship.

**References**
