Design and Synthesis of α,β-Epoxyketones as New Anticancer Agents

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As epoxy functional group has high anticancer activity, α,β-epoxyketones were designed and synthesized as new anticancer agents, and their structures were confirmed by UV, ^{1}H NMR, IR, MS technigeces and elemental analysis. Their *in vitro* anticancer activities were evaluated by MTT method and the results showed that the compound **4c** exhibited good activity with IC_{50} of 17.8, 22.0 and 24.1 μg/mL against A-549, Hela and HepG2 cells, respectively. The dose of LD50 of the mice by intragastric administration was 1864.4 mg/kg. Therefore, the α,β-epoxyketones could potentially provide as new anticancer agents.

Keywords α,β-epoxyketones, thiochromanones, anticancer activity, Darzens condensation reaction

Introduction

Cancer is a global issue and represents a major public health problem. At present there are many anticancer drugs, such as Bleomycin, Carboplatin, Carmustine, Cisplatin, Cyclophosphamide, Fluorouracil, Oxaliplatin, Paclitaxel, Vincristine and so on. But they all have some drawbacks such as undesired side effects and multidrug resistance. Therefore, new effective anticancer drugs are desired all the time.

Epoxides were reported to have strong biological activity, such as anticancer activity,[1] pesticides activity,[2] sterilization activity,[3] growth regulating activity[4] and so on. For example, the triptolide is a diterpenoid epoxides in tripterygium wilfordii, which contains epoxy function groups and was regarded as the most promising anticancer agents, especially for the treatment of ovarian cancer, breast cancer and melanoma.[5,6] And epoxy ethane derivatives were also reported to have strong bactericidal activity.[7-9]

Besides epoxides, thiochromanones had been reported to possess important biological activities such as antifungal activity,[10] anti-inflammatory activities,[11] anticancer activity.[12] For example, 3-bromo-4-thiochromanone derivatives,[13,14] 3-benzylienethiochromanone derivatives,[15,16] the Mannich base of thiochromanone derivatives,[17,18] 3-halomethylene-thiochromanone derivatives,[19,20] 2,3,3a,4-tetrahydrothiochromeno[4,3-*c*]-pyrazole derivatives[21] and 6*H*-thiochromeno-[4,3-*b*]-quinoline derivatives[22] *etc.* had been synthesized and reported to have high antifungal activities. Furthermore, (*Z*)-3-(chloromethylene)-6-methyl-thiochroman-4-one was reported to be highly active to inhibit the growing of many kinds of cancer cells.[23]

In view of the above reports, our interest in thiochromanones containing epoxide groups came to being, and the design, synthesis and biological activities evaluation of α,β-epoxyketones including the structure of thiomanone or chromanone were done.

Results and discussion

Chemistry

The syntheses of these α,β-epoxyketones were carried out by the reaction of a properly substituted 3-bromo-4-(thio)chromanones with an equimolar amount of a suitable aliphatic or aromatic aldehydes in the presence of an excess of freshly prepared sodium methoxide in anhydrous toluene (Scheme 1). The products obtained were purified by conventional workup in satisfactory yield. The starting reagents for the synthesis of α,β-epoxyketones were (thio)chromanones **1**. In our experiment, compounds **1** were synthezied through substitution reaction under microwave irradiation and cyclization reaction in the concentrated sulfuric acid.[20,21,24] The yields were obviously higher than that of traditional methods.[25] The syntheses of compounds **2** were carried out according to reported procedure,[13,14] and all of the

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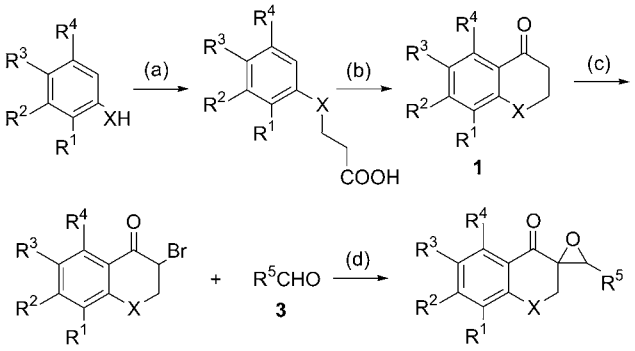
Received August 29, 2010; revised January 9, 2011; accepted February 23, 2011.
Project supported by the National Natural Science Foundation of China (Nos. 20375010, 20675084), the Hebei Province Programs for Science and Technology Development (Nos. 06276479B, 07276407D), Key Laboratory for Pharmaceutical Quality Control of Hebei Province Fund (No. 09265631D-14) and Hebei University Found (No. 2010Q33).

compounds **2** had good yields (85%—93%). The title compounds **4** were obtained by Darzens condensation reactions. Both analytical and ^{1}H NMR spectral data of all the synthesized compounds were in full agreement with the proposed structures.

Scheme 1 Synthetic route of target compounds

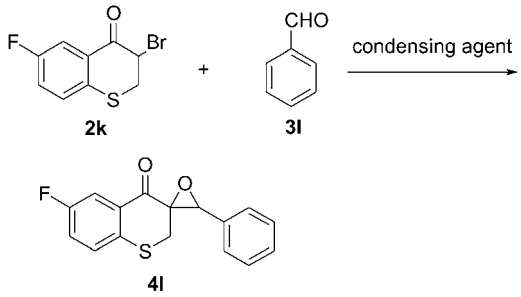


Reagents and conditions: (a) NaOH, $ClCH_2CH_2COOH$, MW; (b) H_2SO_4; (c) Br_2, CH_3COOH, 40 ℃; (d) CH_3ONa, toluene, 0 ℃

	R^1	R^2	R^3	R^4	R^5	X
a	H	H	CH_3	H	4-chlorophenyl	S
b	H	F	CH_3	H	4-chlorophenyl	S
c	H	H	Cl	H	4-chlorophenyl	S
d	Cl	H	H	H	4-chlorophenyl	S
e	Cl	H	H	Cl	4-chlorophenyl	S
f	Cl	H	Cl	H	4-chlorophenyl	S
g	H	Cl	Cl	H	4-chlorophenyl	S
h	H	H	OCH_3	H	4-chlorophenyl	S
i	H	H	$CH(CH_3)_2$	H	4-chlorophenyl	S
j	H	H	Cl	H	4-chlorophenyl	O
k	H	H	F	H	4-chlorophenyl	S
l	H	H	F	H	phenyl	S
m	H	H	F	H	benzo[*d*][1,3]dioxole-5-yl	S
n	H	H	F	H	4-methoxyphenyl	S
o	H	H	F	H	$CH_2CH_2CH_3$	S
p	H	H	F	H	$CH(CH_3)_2$	S

In order to obtain a good yield of the compound **4**, the effects of temperature and condensing agents on the yield of Darzens condensation reactions were investigated. Our study was initiated by the reaction of 3-bromo-6-fluorothiochroman-4-one (**2k**) and benzaldehyde (**3l**) (Eq. 1). The effect of various temperature conditions and different condensing agents on the condensation process was studied, and the results are summarized in Table 1.

It turned out that better yields were obtained at 0 ℃ (Table 1, Entries 1—4). When the reaction temperature rose, on the one hand, self-condensation of the aldehyde



(1)

or ketone might increase; on the other hand, HBr might be removed from compound **2k**, and the **2k** was changed into 6-fluoro-4*H*-thiochromen-4-one which did not participate in Darzens condensation reaction. Therefore, it was better maintaining the condensation process at low temperature than high temperature. Comparing the results of Entries 1 and 2, it was found that the yield was not impoved greatly when the temperature was maintained at 0 ℃ during the whole process, so firstly we chose the condition of Entry 2. The same reaction was also run with several other condensing agents and the results are also included in Table 1 (Entries 5, 6). Comparing CH_3ONa with NaH and *t*-BuONa, the system of NaH and *t*-BuONa exhibited the similar yield to that of CH_3ONa. Because CH_3ONa was cheaper and more accessible than the others, we chose it as the condensing agent at last.

Table 1 The yields of **4l** under different reaction conditions

Entry	Condensing agent	Temperature/℃	Yield/%
1	CH_3ONa	0	69
2	CH_3ONa	0—r.t.	67
3	CH_3ONa	50—70	35
4	CH_3ONa	110	30
5	NaH	0—r.t.	69
6	*t*-BuONa	0—r.t.	71

The effects of R^1, R^2, R^3, R^4, R^5 and X on yields of compounds **4** are presented in Table 2. From the results we could conclude that the R^5 had a significant impact

Table 2 The yields of compounds **4**

Compound	Yield/%	Compound	Yield/%
4a	66	**4i**	62
4b	62	**4j**	67
4c	70	**4k**	71
4d	71	**4l**	67
4e	67	**4m**	64
4f	73	**4n**	60
4g	65	**4o**	51
4h	61	**4p**	45

on the yields of compounds **4**. When R^5 was aryl groups, the yields of compounds **4** were good and higher than 60%; when R^5 was aliphatic groups, the yields were lower than 51%. However, within the range of our experiments, it had not been found that R^1, R^2, R^3, R^4 and X had obvious effects on the yields of compounds **4**.

Biological activity

Evaluation of anticancer activity *in vitro* In order to investigate structure-activity relations of compounds **4** for anticancer activities against A-549 cells, HeLa cells and HepG2 cells, firstly, compounds **4k**—**4p** were designed and synthesized, and their anticancer activities were tested by MTT method with cisplatin as the positive control. From the results of their activities testing, it was shown that the designed compounds **4** possessed anticancer activities; moreover when R^5 was 4-chlorophenyl, the compound **4k** indicated the best anticancer activity with IC_{50} of 35.1, 38.9 and 43.0 μg/mL against A-549, Hela and HepG2 cells, respectively. Secondly, compounds **4a**—**4i** were designed and synthesized when R^5 was fixed as 4-chlorophenyl. Moreover their anticancer activities were investigated by the method as before. The biological activities results of these compounds revealed that 6'-chloro-3-(4-chlorophenyl)-spiro[oxirane-2,3'-thiochroman]-4'-one (**4c**) had the best activities against A-549, HeLa and HepG2 cells, and the IC_{50} values reached 17.8, 22.0 and 24.1 μg/mL, respectively. And the activities of compounds **4d** against HeLa and HepG2 were as high as that of **4c**. So it was indicated when R^1 was Cl or R^3 was Cl and the others among R^1—R^4 were H, compounds **4** possessed good anticancer activities. Especially, compounds **4** had broader range of anticancer avtivities when R^3 was Cl. Thirdly, in order to investigate the impact of X＝O on anticancer activity, 6-chloro-3'-(4-chlorophenyl)-spiro[chroman-3,2'-oxiran]-4-one (**4j**) was designed and synthesized. And from the biological activities of compound **4j**, it was shown that the growth of A-549, HeLa and HepG2 cells were also inhibited by compound **4j** to some extent, and the IC_{50} values were 27.7, 25.8 and 26.9 μg/mL, respectively. But its anticancer activity was lower than that of **4c**. The anticancer activities of all the target compounds are shown in Table 3.

From above, it was concluded that the structure of designed α,β-epoxyketones had anticancer activity, and when R^1, R^2 and R^4 were H, R^3 and R^5 were Cl, and X was S, compound **4** possesses the best anticancer activities. Although the mechanism of anticancer activities remains to be not clarified, the compounds **4** could potentially provide as new anticancer agents.

Evaluation of acute toxicity After screening *in vitro*, the acute toxicity of compound **4c** which had the best anticancer activity was investigated. The result of toxicity investigation indicated that the acute toxicity of compound **4c** on Kunming mice was low; and the mice were all dead at the dose of 3814.7 mg/kg. Moreover the LD50 of compound **4c** on Kunming mice was obtained by Statistical Program for Social Sciences (SPSS). The LD50 of compound **4c** was 1864.4 mg/kg. The observations of death in mice are shown in Table 4.

Table 3 Anticancer activities of title compounds

Compound	IC_{50}/(μg•mL^{-1})		
	A-549	Hela	HepG2
4a	40.0	39.6	37.9
4b	39.5	37.9	42.0
4c	17.8	22.0	24.1
4d	24.0	23.9	25.4
4e	29.5	28.1	28.9
4f	30.0	31.9	28.9
4g	31.9	33.0	32.1
4h	40.0	43.1	39.8
4i	35.9	39.7	38.0
4j	27.7	25.8	26.9
4k	35.1	38.9	43.0
4l	57.9	60.1	58.0
4m	60.7	54.0	49.0
4n	45.9	50.7	48.9
4o	52.0	50.1	47.0
4p	56.0	50.9	60.0
Cisplatin	30.0	9.0	10.0

Table 4 Observations of death in mice

Dose/(mg•kg^{-1})	Number of experimental animals	Number of death
1000.0	10	0
1250.0	10	2
1562.5	10	3
1953.1	10	5
2441.4	10	8
3051.7	10	9
3814.7	10	10

Conclusion

In summary, the designed α,β-epoxyketones were successfully synthesized and confirmed with spectra. And by optimizing the reaction condition, an economical preparing process of compounds **4** was constructed. Moreover, the biological activities of these compounds were evaluated, and all those were found to possess anticancer activities; especially compound **4c** had the best activities, and the IC_{50} values against A-549, HeLa and HepG2 cells reached 17.8, 22.0 and 24.1 μg/mL, respectively. Moreover the acute toxicity of compound **4c** was low, and its LD50 was 1864.4 mg/kg. Furthermore it was found that the anticancer activities of

α,*β*-epoxyketones were strongly dependent on its substituents. So the structure of *α*,*β*-epoxyketones could provide as a new group of anticancer agents, and the structures require optimizing deeply.

Experimental

Instruments

Melting points of all compounds synthesized were determined with an SGW X-4 melting point apparatus. The ^{1}H NMR spectra were recorded in $CDCl_3$ at 600 MHz with an AVANCE 600 spectrometer (internal standard TMS). The MS spectra were measured on LC-MSD Trap XCT spectrometer. Elemental analysis (C, H, S) was realized on Carlo Erba-1106 EA instrument.

General procedure for synthesis of compounds 1

Compounds **1** were synthesized according to published procedures.[20,21,23]

6-Methylthiochroman-4-one (**1a**): Light yellow solid, yield 61%, m.p. 70—72 ℃; UV-vis (MeOH) λ_{max}: 254 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 2.47 (s, 3H, CH_3), 2.83—2.85 (m, 2H, $COCH_2$), 3.26—3.28 (m, 2H, CH_2S), 6.89—7.01 (m, 1H, ArH), 7.12 (d, J=8.6 Hz, 1H, ArH), 7.55 (d, J=2.8 Hz, 1H, ArH); IR (KBr) ν: 2923, 2854, 1674, 1599, 1469, 1398 cm^{-1}; MS (APCI) m/z: 178.9 $(M+H)^+$. Anal. calcd for $C_{10}H_{10}OS$: C 67.38, H 5.65, S 17.99; found C 67.30, H 5.68, S 17.93.

7-Fluoro-6-methylthiochroman-4-one (**1b**): Yellow solid, yield 65%, m.p. 48—49 ℃; UV-vis (MeOH) λ_{max}: 254 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 2.45 (s, 3H, CH_3), 2.86—2.89 (m, 2H, $COCH_2$), 3.25—3.27 (m, 2H, SCH_2), 6.89—7.01 (m, 1H, ArH), 7.59 (d, J=2.9 Hz, 1H, ArH); IR (KBr) ν: 2925, 2854, 1671, 1588, 1469, 1469, 1398 cm^{-1}; MS (APCI) m/z: 196.9 $(M+H)^+$. Anal. calcd for $C_{10}H_9FOS$: C 61.20, H 4.62, S 16.34; found C 61.16, H 4.68, S 16.30.

6-Chlorothiochroman-4-one (**1c**): Yellow solid, yield 65%, m.p. 69—70 ℃; UV-vis (MeOH) λ_{max}: 243 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 2.95—2.97 (m, 2H, $COCH_2$), 3.17—3.19 (m, 2H, SCH_2), 6.97—7.01 (m, 1H, ArH), 7.19 (d, J=8.4 Hz, 1H, ArH), 7.60 (d, J=3.2 Hz, 1H, ArH); IR (KBr) ν: 2910, 2846, 1678, 1598, 1479 cm^{-1}; MS (APCI) m/z: 198.9 $(M+H)^+$, 200.9 $(M+2+H)^+$. Anal. calcd for C_9H_7ClOS: C 54.41, H 3.55, S 16.14; found C 54.45, H 3.50, S 16.10.

8-Chlorothiochroman-4-one (**1d**): Light yellow solid, yield 67%, m.p. 93—95 ℃; UV-vis (MeOH) λ_{max}: 244 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 2.95—2.97 (m, 2H, $COCH_2$), 3.24—3.27 (m, 2H, SCH_2), 7.13 (t, J=7.85 Hz, 1H, ArH), 7.49 (dd, J=7.84, 1.35 Hz, 1H, ArH), 8.05 (dd, J=7.92, 1.36 Hz, 1H, ArH); IR (KBr) ν: 2923, 1664, 1574, 1427 cm^{-1}; MS (APCI) m/z: 198.9 $(M+H)^+$, 200.9 $(M+2+H)^+$. Anal. calcd for C_9H_7ClOS: C 54.41, H 3.55, S 16.14; found C 54.40, H 3.58, S 16.10.

5,8-Dichlorothiochroman-4-one (**1e**): Yellow solid, yield 70%, m.p. 74—75 ℃; UV-vis (MeOH) λ_{max}: 239 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 3.01—3.03 (m, 2H, $COCH_2$), 3.24—3.29 (m, 2H, SCH_2), 7.16 (d, J=8.41 Hz, 1H, ArH), 7.34 (d, J=8.42 Hz, 1H, ArH); IR (KBr) ν: 2929, 2853, 1697, 1558, 1507, 1469 cm^{-1}; MS (APCI) m/z: 232.9 $(M+H)^+$, 234.9 $(M+2+H)^+$. Anal. calcd for $C_9H_6Cl_2OS$: C 46.37, H 2.59, S 13.76; found C 46.40, H 2.53, S 13.70.

6,8-Dichlorothiochroman-4-one (**1f**): Yellow solid, yield 70%, m.p. 84—85 ℃; UV-vis (MeOH) λ_{max}: 248 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 2.96—2.98 (m, 2H, $COCH_2$), 3.24—3.27 (m, 2H, SCH_2), 7.50 (d, J=2.33 Hz, 1H, ArH), 8.03 (d, J=2.35 Hz, 1H, ArH); IR (KBr) ν: 2933, 2833, 1685, 1566, 1471 cm^{-1}; MS (APCI) m/z: 232.9 $(M+H)^+$, 234.9 $(M+2+H)^+$. Anal. calcd for $C_9H_6Cl_2OS$: C 46.37, H 2.59, S 13.76; found C 46.40, H 2.54, S 13.72.

6,7-Dichlorothiochroman-4-one (**1g**) Pink solid, yield 73%, m.p. 133—135 ℃; UV-vis (MeOH) λ_{max}: 248 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 2.93—2.99 (m, 2H, $COCH_2$), 3.23—3.28 (m, 2H, SCH_2), 7.40 (s, 1H, ArH), 8.16 (s, 1H, ArH); IR (KBr) ν: 2947, 2847, 1668, 1569, 1507, 1444 cm^{-1}; MS (APCI) m/z: 232.9 $(M+H)^+$, 234.9 $(M+2+H)^+$. Anal. calcd for $C_9H_6Cl_2OS$: C 46.37, H 2.59, S 13.76; found C 46.40, H 2.53, S 13.73.

6-Methoxythiochroman-4-one (**1h**): Yellow solid, yield 55%, m.p. 30—33 ℃; UV-vis (MeOH) λ_{max}: 240 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 2.95—2.97 (m, 2H, SCH_2), 3.19—3.23 (m, 2H, SCH_2), 3.82 (s, 3H, OCH_3), 7.01 (dd, J=8.68, 2.96 Hz, 1H, ArH), 7.19 (d, J=8.68 Hz, 1H, ArH), 7.62 (d, J=2.97 Hz, 1H, ArH); IR (KBr) ν: 2937, 2833, 1681, 1598, 1473 cm^{-1}; MS (APCI) m/z: 194.9 $(M+H)^+$. Anal. calcd for $C_{10}H_{10}O_2S$: C 61.83, H 5.19, S 16.51; found C 61.90, H 5.15, S 16.57.

6-Isopropylthiochroman-4-one (1i): Red oil, yield 67%, UV-vis (MeOH) λ_{max}: 238 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 1.24 [d, J=7.04 Hz, 6H, CH-(C**H**$_3$)$_2$], 2.86—2.93 [m, 1H, C**H**-$(CH_3)_2$], 2.96—2.98 (m, 2H, $COCH_2$), 3.20—3.24 (m, 2H, SCH_2), 7.21 (d, J=8.17 Hz, 1H, ArH), 7.26—7.28 (m, 1H, ArH), 7.99 (d, J=1.89 Hz, 1H, ArH); IR (KBr) ν: 2960, 2869, 1677, 1598, 1471 cm^{-1}; MS (APCI) m/z: 207 $(M+H)^+$. Anal. calcd for $C_{12}H_{14}OS$: C 69.86, H 6.84, S 15.54; found C 68.80, H 6.88, S 15.50.

6-Chlorochroman-4-one (**1j**): White solid, yield 65%, m.p. 102—103 ℃; UV-vis (MeOH) λ_{max}: 222 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 2.81 (t, J=6.60 Hz, 2H, OCH_2), 4.54 (t, J=6.60 Hz, 2H, OCH_2), 6.94 (d, J=8.85 Hz, 1H, ArH), 7.41 (dd, J=8.86, 2.64 Hz, 1H, ArH), 7.85 (d, J=2.60 Hz, 1H, ArH); IR (KBr) ν: 2933, 2840, 1677, 1573, 1469 cm^{-1}; MS (APCI) m/z: 182.9 $(M+H)^+$, 184.9 $(M+2+H)^+$. Anal. calcd for $C_9H_7ClO_2$: C 59.20, H 3.86; found C 59.27, H 3.90.

6-Fluorothiochroman-4-one (**1k**): Light yellow solid, yield 64%, m.p. 92—94 ℃; UV-vis (MeOH) λ_{max}: 254 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 2.96—2.99 (m, 2H, $COCH_2$), 3.23—3.25 (m, 2H, SCH_2), 7.11—7.16 (m, 2H, ArH), 8.07 (dd, J=7.96, 1.39 Hz, 1H, ArH); IR (KBr) ν: 2923, 2854, 1660, 1588, 1469 cm^{-1}; MS

(APCI) *m/z*: 182.9 $(M+H)^+$. Anal. calcd for C_9H_7FOS: C 59.32, H 3.87, S 17.60; found C 59.27, H 3.90, S 17.64.

General procedure for the synthesis of compounds 2

15.0 mmol compound **1** was dissolved in 15 mL of glacial acetic acid at about 40 ℃, then a solution of 15.0 mmol bromine in 8 mL of glacial acetic acid was added dropwise, over a period of 40 min at 40 ℃. The reaction mixture was stirred for 3.5 h, then cooled and added 80 mL of saturated $NaHSO_3$ solution, the solid precipitated was filtered, abundantly washed with water, then air dried. The crude product was recrystallized from 95% (ϕ) EtOH to afford the compound **2**.

3-Bromo-6-methylthiochroman-4-one (**2a**): Brown solid, yield 88%, m.p. 57—58 ℃; UV-vis (MeOH) λ_{max}: 250 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 2.19—2.52 (m, 3H, CH_3), 3.50 (dd, J=14.02, 8.44 Hz, 1H, SCH_2), 3.66 (dd, J=14.04, 3.31 Hz, 1H, SCH_2), 4.98 (dd, J=8.46, 3.31 Hz, 1H, CHBr), 7.21 (d, J=8.09 Hz, 1H, ArH), 7.28—7.31 (m, 1H, ArH), 7.90—8.29 (m, 1H, ArH); IR (KBr) ν: 2977, 2844, 1672, 1606, 1589, 1469, 1398 cm^{-1}; MS (APCI) *m/z*: 176.9 $(M-HBr+H)^+$. Anal. calcd for $C_{10}H_9BrOS$: C 46.71, H 3.53, S 12.47; found C 46.67, H 3.58, S 12.43.

3-Bromo-7-fluoro-6-methylthiochroman-4-one (**2b**): Pale brown solid, yield 90%, m.p. 143—145 ℃; UV-vis (MeOH) λ_{max}: 245 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 2.25—2.39 (m, 3H, CH_3), 3.48 (dd, J=14.08, 8.23 Hz, 1H), 3.66 (dd, J=14.11, 3.27 Hz, 1H, SCH_2), 4.95 (dd, J=8.24, 3.26 Hz, 1H, CHBr), 7.09—7.23 (m, 1H, ArH), 7.73—7.90 (m, 1H, ArH); IR (KBr) ν: 2925, 2854, 1683, 1606, 1586, 1510, 1473, 1382 cm^{-1}; MS (APCI) *m/z*: 194.9 $(M-HBr+H)^+$. Anal. calcd for $C_{10}H_8BrFOS$: C 43.65, H 3.53, S 11.65; found C 43.61, H 2.88, S 11.60.

3-Bromo-6-chlorothiochroman-4-one (**2c**): Light yellow solid, yield 90%, m.p. 108—110 ℃; UV-vis (MeOH) λ_{max}: 245 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 3.50 (dd, J=14.17, 8.12 Hz, 1H, SCH_2), 3.70 (dd, J=14.18, 3.25 Hz, 1H, SCH_2), 4.96 (dd, J=8.11, 3.24 Hz, 1H, CHBr), 7.26 (d, J=8.52 Hz, 1H, ArH), 7.36—7.51 (m, 1H, ArH), 8.14 (d, J=2.40 Hz, 1H, ArH); IR (KBr) ν: 2920, 2850, 1687, 1577, 1458 cm^{-1}; MS (APCI) *m/z*: 196.9 $(M-HBr+H)^+$, 198.9 $(M-HBr+2+H)^+$. Anal. calcd for $C_9H_6BrClOS$: C 38.94, H 2.18, S 11.55; found C 38.89, H 2.15, S 11.59.

3-Bromo-8-chlorothiochroman-4-one (**2d**): Yellow oil, yield 90%, UV-vis (MeOH) λ_{max}: 245 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 3.50 (dd, J=14.26, 8.31 Hz, 1H, SCH_2), 3.68 (dd, J=14.23, 3.29 Hz, 1H, SCH_2), 4.95 (dd, J=8.35, 3.18 Hz, 1H, CHBr), 7.19 (t, J=7.90 Hz, 1H, ArH), 7.54 (dd, J=7.79, 1.26 Hz, 1H, ArH), 8.10 (dd, J=8.03, 1.30 Hz, 1H, ArH); IR (KBr) ν: 2920, 1685 1577, 1550, 1402 cm^{-1}; MS (APCI) *m/z*: 196.9 $(M-HBr+H)^+$, 198.9 $(M-HBr+2+H)^+$. Anal. calcd for $C_9H_6BrClOS$: C 38.94, H 2.18, S 11.55; found C 38.90, H 2.15, S 11.51.

3-Bromo-5,8-dichlorothiochroman-4-one (**2e**): Yellow solid, yield 91%, m.p. 70—73 ℃; UV-vis (MeOH) λ_{max}: 250 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 3.51 (dd, J=14.14, 8.44 Hz, 1H, SCH_2), 3.72 (dd, J=14.17, 3.77 Hz, 1H, SCH_2), 4.93 (dd, J=8.46, 3.77 Hz, 1H, CHBr), 7.22 (d, J=8.53 Hz, 1H, ArH), 7.39 (d, J=8.48 Hz, 1H, ArH); IR (KBr) ν: 2948, 1699, 1558, 1542, 1419 cm^{-1}; MS (APCI) *m/z*: 230.9 $(M-HBr+H)^+$, 232.9 $(M-HBr+2+H)^+$. Anal. calcd for $C_9H_5BrCl_2OS$: C 34.65, H 1.62, S 10.28; found C 34.69, H 1.65, S 10.20.

3-Bromo-6,8-dichlorothiochroman-4-one (**2f**): Light yellow solid, yield 90%, m.p. 113—115 ℃; UV-vis (MeOH) λ_{max}: 250 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 3.48 (dd, J=14.34, 7.91 Hz, 1H), 3.68 (dd, J=14.33, 3.13 Hz, 1H), 4.92 (dd, J=7.91, 3.18 Hz, 1H), 7.55 (d, J=2.31 Hz, 1H), 8.08 (d, J=2.30 Hz, 1H); IR (KBr) ν: 2916, 1681, 1569, 1400 cm^{-1}; MS (APCI) *m/z* : 230.9 $(M-HBr+H)^+$, 232.9 $(M-HBr+2+H)^+$. Anal. calcd for $C_9H_5BrCl_2OS$: C 34.65, H 1.62, S 10.28; found C 34.62, H 1.68, S 10.23.

3-Bromo-6,7-dichlorothiochroman-4-one (**2g**): Yellow solid, yield 89%, m.p. 123—125 ℃; UV-vis (MeOH) λ_{max}: 254 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 3.69 (dd, J=14.25, 3.15 Hz, 1H, SCH_2), 3.47 (dd, J=14.23, 7.83 Hz, 1H, SCH_2), 4.92 (dd, J=7.84, 3.21 Hz, 1H, CHBr), 7.42 (s, 1H, ArH), 8.21 (s, 1H, ArH); IR (KBr) ν: 2918, 1683, 1573, 1442 cm^{-1}; MS (APCI) *m/z*: 230.9 $(M-HBr+H)^+$, 232.9 $(M-HBr+2+H)^+$. Anal. calcd for $C_9H_5BrCl_2OS$: C 34.65, H 1.62, S 10.28; found C 34.61, H 1.65, S 10.32.

3-Bromo-6-methoxythiochroman-4-one (**2h**): Yellow solid, yield 92%, m.p. 62—64 ℃; UV-vis (MeOH) λ_{max}: 245 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 3.46 (dd, J=14.00, 8.33 Hz, 1H, SCH_2), 3.63 (dd, J=13.99, 3.34 Hz, 1H, SCH_2), 3.83 (s, 3H, OCH_3), 4.96 (dd, J=8.32, 3.33 Hz, 1H, CHBr), 7.06 (dd, J=8.77, 2.90 Hz, 1H, ArH), 7.20 (d, J=8.69 Hz, 1H, ArH), 7.64 (d, J=2.93 Hz, 1H, ArH); IR (KBr) ν: 2960, 2831, 1662, 1596, 1475, 1226, 1024 cm^{-1}; MS (APCI) *m/z*: 193 $(M-HBr+H)^+$. Anal. calcd for $C_{10}H_9BrO_2S$: C 43.97, H 3.32, S 11.74; found C 43.93, H 3.36, S 11.69.

3-Bromo-6-isopropylthiochroman-4-one (**2i**): Red oil; yield 89%, UV-vis (MeOH) λ_{max}: 245 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 1.24 [d, J = 6.98 Hz, 6H, CH-(C**H**$_3$)$_2$], 2.87—2.94 [m, 1H, C**H**-(CH$_3$)$_2$], 3.45—3.49 (m, 1H, SCH_2), 3.64 (dd, J=14.03, 3.32 Hz, 1H, SCH_2), 4.96 (dd, J=8.43, 3.31 Hz, 1H, CHBr), 7.22 (d, J=8.18 Hz, 1H, ArH), 7.32 (dd, J=8.23, 2.16 Hz, 1H, ArH), 8.02 (d, J=2.15 Hz, 1H, ArH); IR (KBr) ν: 2960, 2869, 1681, 1598, 1475 cm^{-1}; MS (APCI) *m/z*: 205 $(M-HBr+H)^+$. Anal. calcd for $C_{12}H_{13}BrOS$: C 50.54, H 4.59, S 11.24; found C 50.50, H 4.56, S 11.20.

3-Bromo-6-chlorochroman-4-one (**2j**): White solid, yield 93%, m.p. 97—99 ℃; UV-vis (MeOH) λ_{max}: 245 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 4.58—4.67 (m, 3H, OCH_2 and CHBr), 7.01 (d, J=8.86 Hz, 1H, ArH), 7.49 (dd, J=8.92, 2.66 Hz, 1H, ArH), 7.90 (d, J=2.66 Hz,

1H, ArH); IR (KBr) *v*: 2929, 1697, 1598, 1473 cm^{-1}; MS (APCI) *m/z*: 180.9 $(M-HBr+H)^+$, 182.9 $(M-HBr+2+H)^+$. Anal. calcd for $C_9H_6BrClO_2$: C 41.34, H 2.31; found C 41.37, H 2.30.

3-Bromo-6-fluorothiochroman-4-one (**2k**): Pale brown solid, yield 91%, m.p. 103—105 ℃; UV-vis (MeOH) λ_{max}: 250 nm; ^1H NMR ($CDCl_3$, 600 MHz) δ: 3.50 (dd, J=14.13, 8.26 Hz, 1H), 3.68 (dd, J=14.12, 3.26 Hz, 1H), 4.98 (dd, J=8.25, 3.26 Hz, 1H), 7.10—7.24 (m, 1H), 7.30 (dd, J=8.73, 4.88 Hz, 1H), 7.87 (dd, J=9.16, 2.88 Hz, 1H); IR (KBr) *v*: 2920, 2831, 1687, 1600, 1573, 1469 cm^{-1}; MS (APCI) *m/z*: 180.9 $(M-HBr+H)^+$. Anal. calcd for C_9H_6BrFOS: C 41.40, H 2.32, S 12.28; found C 41.37, H 2.36, S 12.23.

General procedure for the synthesis of compounds 4

10.0 mmol compound **2** and 10.0 mmol substituted aldehyde **3** were dissolved in 25 mL of dry toluene and cooled to 0 ℃, then added 20.0 mmol freshly prepared sodium methoxide. After all the base was added, the reaction mixture was permitted to warm slowly to room temperature and stirred for 8—10 h. Then the reaction mixture was poured into 30 mL of water, the organic layer was obtained and dried over anhydrous $MgSO_4$, then filtered and evaporated. The crude product was purified by silica-gel column chromatography [*V*(ethyl acetate)∶*V*(petroleum)=1∶60—1∶10] to afford the compound **4**.

3-(4-Chlorophenyl)-6'-methylspiro[oxirane-2,3'-thiochroman]-4'-one (**4a**): White solid, yield 66%, m.p. 154—156 ℃; UV-vis (MeOH) λ_{max}: 247 nm; ^1H NMR ($CDCl_3$, 600 MHz) δ: 2.36 (s, 3H, CH_3), 2.51 (d, J=13.45 Hz, 1H, SCH_2), 3.75 (d, J=13.50 Hz, 1H, SCH_2), 4.37 (s, 1H, OCH), 7.16 (d, J=8.02 Hz, 1H, ArH), 7.24—7.27 (m, 2H, ArH), 7.38 (s, 4H, ArH), 7.96 (d, J=0.89 Hz, 1H, ArH); IR (KBr) *v*: 2923, 2854, 1674, 1596, 1469, 1398, 1286 cm^{-1}; MS (APCI) *m/z*: 317 $(M+H)^+$, 319 $(M+2+H)^+$. Anal. calcd for $C_{17}H_{13}ClO_2S$: C 64.45, H 4.14, S 10.12; found C 64.39, H 4.19, S 10.08.

3-(4-Chlorophenyl)-7'-fluoro-6'-methylspiro[oxirane-2,3'-thiochroman]-4'-one (**4b**): Yellow-green solid, yield 62%, m.p. 139—141 ℃; UV-vis (MeOH) λ_{max}: 247 nm; ^1H NMR ($CDCl_3$, 600 MHz) δ: 2.29 (s, 3H, CH_3), 2.52 (d, J=13.50 Hz, 1H, SCH_2), 3.74 (d, J=13.50 Hz, 1H, SCH_2), 4.37 (s, 1H, OCH), 7.09 (d, J=6.60 Hz, 1H, ArH), 7.36—7.40 (m, 4H, ArH), 7.76 (d, J=9.89 Hz, 1H, ArH); IR (KBr) *v*: 2923, 2854, 1681, 1608, 1494, 1471, 1394, 1272 cm^{-1}; MS (APCI) *m/z*: 335 $(M+H)^+$, 337 $(M+2+H)^+$. Anal. calcd for $C_{17}H_{12}ClFO_2S$: C 60.99, H 3.61, S 9.58; found C 60.94, H 3.65, S 9.51.

6'-Chloro-3-(4-chlorophenyl)spiro[oxirane-2,3'-thiochroman]-4'-one (**4c**): White solid, yield 70%, m.p. 148—149 ℃; UV-vis (MeOH) λ_{max}: 248 nm; ^1H NMR ($CDCl_3$, 600 MHz) δ: 2.55 (d, J=13.54 Hz, 1H, SCH_2), 3.76 (d, J=13.49 Hz, 1H, SCH_2), 4.38 (s, 1H, OCH), 7.22 (d, J=8.45 Hz, 1H, ArH), 7.38—7.40 (m, 5H, ArH), 8.11 (d, J=2.37 Hz, 1H, ArH); IR (KBr) *v*: 1679, 1577, 1490, 1454, 1240 cm^{-1}; MS (APCI) *m/z*: 337 $(M+H)^+$, 339 $(M+2+H)^+$. Anal. calcd for $C_{16}H_{10}Cl_2O_2S$: C 56.99, H 2.99, S 9.51; found C 56.92, H 2.90, S 9.55.

8'-Chloro-3-(4-chlorophenyl)spiro[oxirane-2,3'-thiochroman]-4'-one (**4d**): Light yellow solid, yield 71%, m.p. 136—139 ℃; UV-vis (MeOH) λ_{max}: 248 nm; ^1H NMR ($CDCl_3$, 600 MHz) δ: 2.60 (d, J=13.57 Hz, 1H, SCH_2), 3.75 (d, J=13.58 Hz, 1H, SCH_2), 4.37 (s, 1H, OCH), 7.22 (t, J=7.89 Hz, 1H, ArH), 7.36—7.43 (m, 4H, ArH), 7.55 (d, J=7.80 Hz, 1H, ArH), 8.10 (d, J=7.96 Hz, 1H, ArH); IR (KBr) *v*: 1685, 1573, 1488, 1278 cm^{-1}; MS (APCI) *m/z*: 337 $(M+H)^+$, 339 $(M+2+H)^+$. Anal. calcd for $C_{16}H_{10}Cl_2O_2S$: C 56.99, H 2.99, S 9.51; found C 56.93, H 2.91, S 9.54.

5',8'-Dichloro-3-(4-chlorophenyl)spiro[oxirane-2,3'-thiochroman]-4'-one (**4e**): Yellow solid, yield 67%, m.p. 131—133 ℃; UV-vis (MeOH) λ_{max}: 248 nm; ^1H NMR ($CDCl_3$, 600 MHz) δ: 2.71 (d, J=13.77 Hz, 1H, SCH_2), 3.48 (d, J=13.77 Hz, 1H, SCH_2), 4.51 (s, 1H, OCH), 7.26 (d, J=3.50 Hz, 1H, ArH), 7.34—7.42 (m, 5H, ArH); IR (KBr) *v*: 1697, 1598, 1490, 1274 cm^{-1}; MS (APCI) *m/z*: 371 $(M+H)^+$, 373 $(M+2+H)^+$, 375 $(M+4+H)^+$. Anal. calcd for $C_{16}H_9Cl_3O_2S$: C 51.71, H 2.44, S 8.63; found C 51.76, H 2.40, S 8.68.

6',8'-Dichloro-3-(4-chlorophenyl)spiro[oxirane-2,3'-thiochroman]-4'-one (**4f**): Yellow solid, yield 73%, m.p. 153—154 ℃; UV-vis (MeOH) λ_{max}: 254 nm; ^1H NMR ($CDCl_3$, 600 MHz) δ: 2.61 (d, J=13.68 Hz, 1H, SCH_2), 3.74 (d, J=13.57 Hz, 1H, SCH_2), 4.37 (s, 1H, OCH), 7.41 (d, J=8.57 Hz, 2H, ArH), 7.38 (d, J=8.55 Hz, 2H, ArH), 7.55 (d, J=2.28 Hz, 1H, ArH), 8.07 (d, J=2.27 Hz, 1H, ArH); IR (KBr) *v*: 1691, 1567, 1494, 1261 cm^{-1}; MS (APCI) *m/z*: 371 $(M+H)^+$, 373 $(M+2+H)^+$, 375 $(M+4+H)^+$. Anal. calcd for $C_{16}H_9Cl_3O_2S$: C 51.71, H 2.44, S 8.63; found C 51.75, H 2.48, S 8.60.

6',7'-Dichloro-3-(4-chlorophenyl)spiro[oxirane-2,3'-thiochroman]-4'-one (**4g**): Yellow solid, yield 65%, m.p. 166—167 ℃; UV-vis (MeOH) λ_{max}: 254 nm; ^1H NMR ($CDCl_3$, 600 MHz) δ: 2.56 (d, J=13.59 Hz, 1H, SCH_2), 3.76 (d, J=13.57 Hz, 1H, SCH_2), 4.38 (s, 1H, OCH), 7.41—7.36 (m, 5H, ArH), 8.19 (s, 1H, ArH),; IR (KBr) *v*: 1697, 1568, 1488, 1257 cm^{-1}; MS (APCI) *m/z*: 371 $(M+H)^+$, 373 $(M+2+H)^+$, 375 $(M+4+H)^+$. Anal. calcd for $C_{16}H_9Cl_3O_2S$: C 56.99, H 2.99, S 9.51; found C 56.93, H 2.93, S 9.55.

3-(4-Chlorophenyl)-6'-methoxyspiro[oxirane-2,3'-thiochroman]-4'-one (**4h**): Yellow solid, yield 61%, m.p. 130—134 ℃; UV-vis (MeOH) λ_{max}: 248 nm; ^1H NMR ($CDCl_3$, 600 MHz) δ: 2.51 (d, J=13.41 Hz, 1H, SCH_2), 3.75 (d, J=13.33 Hz, 1H, SCH_2), 3.85 (s, 3H, OCH_3), 4.38 (s, 1H, OCH), 7.05 (dd, J=8.66, 2.96 Hz, 1H, ArH), 7.17 (d, J=8.65 Hz, 1H, ArH), 7.39 (s, 4H, ArH), 7.62 (d, J=2.92 Hz, 1H, ArH); IR (KBr) *v*: 2923, 2825, 1679, 1595, 1558, 1473, 1398, 1290 cm^{-1}; MS (APCI) *m/z*: 333 $(M+H)^+$, 335 $(M+2+H)^+$. Anal. calcd for $C_{17}H_{13}ClO_3S$: C 61.35, H 3.94, S 9.63; found C 61.30, H 3.90, S 9.70.

3-(4-Chlorophenyl)-6'-isopropylspiro[oxirane-2,3'-thiochroman]-4'-one (**4i**): Yellow solid, yield 62%, m.p.

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161—163 ℃; UV-vis (MeOH) λ_{max}: 247 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 1.26 [d, J = 6.95 Hz, 6H, CH-(C**H**$_3$)$_2$], 2.51 (d, J=13.45 Hz, 1H, SCH_2), 2.89—2.96 [m, 1H, C**H**-(CH_3)$_2$], 3.76 (d, J=13.50 Hz, 1H, SCH_2), 4.37 (s, 1H, OCH), 7.20 (d, J=8.11 Hz, 1H, ArH), 7.32 (dd, J=8.12, 2.09 Hz, 1H, ArH), 7.36—7.40 (m, 4H, ArH), 8.02 (d, J=2.03 Hz, 1H, ArH); IR (KBr) v: 2923, 2864, 1681, 1596, 1490, 1471, 1370, 1267 cm^{-1}; MS (APCI) m/z: 345 $(M+H)^+$, 347 $(M+2+H)^+$. Anal. calcd for $C_{19}H_{17}ClO_2S$: C 66.17, H 4.97, S 9.30; found C 66.12, H 4.93, S 9.35.

6-Chloro-3'-(4-chlorophenyl)spiro[chroman-3,2'-oxiran]-4-one (**4j**): White solid, yield 67%, m.p. 176—177 ℃; UV-vis (MeOH) λ_{max}: 224 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 4.09 (d, J=12.63 Hz, 1H, OCH_2), 4.52—4.55 (m, 2H, OCH and OCH_2), 6.94 (d, J=8.83 Hz, 1H, ArH), 7.30 (d, J=8.43 Hz, 2H, ArH), 7.41 (d, J=8.47 Hz, 2H, ArH), 7.47 (dd, J=8.89, 2.67 Hz, 1H, ArH), 7.93 (d, J=2.66 Hz, 1H, ArH); IR (KBr) v: 1683, 1604, 1571, 1490, 1261 cm^{-1}; MS (APCI) m/z: 321 $(M+H)^+$, 323 $(M+2+H)^+$. Anal. calcd for $C_{16}H_{10}Cl_2O_3$: C 59.84, H 3.14; found C 59.80, H 3.10.

3-(4-Chlorophenyl)-6'-fluorospiro[oxirane-2,3'-thiochroman]-4'-one (**4k**): Light yellow solid, yield 71%, m.p. 164—165 ℃; UV-vis (MeOH) λ_{max}: 240 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 2.54 (d, J=13.54 Hz, 1H, SCH_2), 3.76 (d, J=13.51 Hz, 1H, SCH_2), 4.39 (s, 1H, OCH), 7.16—7.20 (m, 1H, ArH), 7.24—7.26 (m, 1H, ArH), 7.37—7.41 (m, 4H, ArH), 7.82 (dd, J=9.07, 2.91 Hz, 1H, ArH); IR (KBr) v: 1681, 1600, 1488, 1244 cm^{-1}; MS (APCI) m/z: 321 $(M+H)^+$, 323 $(M+2+H)^+$. Anal. calcd for $C_{16}H_{10}ClFO_2S$: C 59.91, H 3.14, S 10.00; found C 59.89, H 3.10, S 10.03.

6'-Fluoro-3-phenylspiro[oxirane-2,3'-thiochroman]-4'-one (**4l**): Light yellow solid, yield 67%, m.p. 129—131 ℃; UV-vis (MeOH) λ_{max}: 244 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 2.61 (d, J=13.51 Hz, 1H, SCH_2), 3.76 (d, J=13.54 Hz, 1H, SCH_2), 4.44 (s, 1H, OCH), 7.15—7.19 (m, 1H, ArH), 7.25 (dd, J=8.68, 4.86 Hz, 1H, ArH), 7.38—7.45 (m, 5H, ArH), 7.83 (dd, J=9.11, 2.90 Hz, 1H, ArH); IR (KBr) v: 1685, 1598, 1471, 1276 cm^{-1}; MS (APCI) m/z: 287 $(M+H)^+$. Anal. calcd for $C_{16}H_{11}FO_2S$: C 67.12, H 3.87, S 11.20; found C 67.09, H 3.77, S 11.17.

3-(Benzo[d][1,3]dioxol-5-yl)-6'-fluorospiro[oxirane-2,3'-thiochroman]-4'-one (**4m**): Yellow solid, yield 64%, m.p. 200—202 ℃; UV-vis (MeOH) λ_{max}: 245 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 2.65 (d, J=13.43 Hz, 1H, SCH_2), 3.77 (d, J=13.38 Hz, 1H, SCH_2), 4.34 (s, 1H, OCH), 6.01 (s, 2H, OCH_2O), 6.84 (d, J=7.94 Hz, 1H, ArH), 6.89—6.93 (m, 2H, ArH), 7.17 (dt, J=8.55, 8.17, 2.85 Hz, 1H, ArH), 7.23—7.26 (m, 1H, ArH), 7.81 (dd, J=9.10, 2.82 Hz, 1H, ArH); IR (KBr) v: 1685, 1598, 1488, 1450, 1259 cm^{-1}; MS (APCI) m/z: 331 $(M+H)^+$. Anal. calcd for $C_{17}H_{11}FO_4S$: C 61.81, H 3.36, S 9.71; found C 61.89, H 3.29, S 9.68.

6'-Fluoro-3-(4-methoxyphenyl)spiro[oxirane-2,3'-thiochroman]-4'-one (**4n**): Yellow-green solid, yield 60%, m.p. 150—152 ℃; UV-vis (MeOH) λ_{max}: 236 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 2.51 (d, J=13.44 Hz, 1H, SCH_2), 3.74 (d, J=13.39 Hz, 1H, SCH_2), 3.85 (s, 3H, OCH_3), 4.38 (s, 1H, OCH), 7.05 (dd, J=8.65, 2.90 Hz, 1H, ArH), 7.25—7.41 (m, 5H, ArH), 7.62 (d, J=2.91 Hz, 1H, ArH); IR (KBr) v: 2923, 2852, 1681, 1595, 1558, 1471, 1228 cm^{-1}; MS (APCI) m/z: 317 $(M+H)^+$. Anal. calcd for $C_{17}H_{13}FO_3S$: C 64.54, H 4.14, S 10.14; found C 61.49, H 4.19, S 10.09.

6'-Fluoro-3-propylspiro[oxirane-2,3'-thiochroman]-4'-one (**4o**): White solid, yield 51%, m.p. 76—79 ℃; UV-vis (MeOH) λ_{max}: 242 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 1.03 (t, J=7.26 Hz, 3H, CH_3), 1.60—1.86 (m, 4H, CH_2CH_2), 2.87 (d, J=13.69 Hz, 1H, SCH_2), 3.23—3.26 (m, 1H, OCH), 3.82 (d, J=13.63 Hz, 1H, SCH_2), 7.15—7.19 (m, 1H, ArH), 7.30 (dd, J=8.71, 4.84 Hz, 1H, ArH), 7.80 (dd, J=9.14, 2.92 Hz, 1H, ArH); IR (KBr) v: 2960, 2869, 1456, 1398, 1683, 1598, 1558, 1471, 1263 cm^{-1}; MS (APCI) m/z: 253 $(M+H)^+$. Anal. calcd for $C_{13}H_{13}FO_2S$: C 61.89, H 5.19, S 12.71; found C 61.84, H 5.15, S 12.68.

6'-Fluoro-3-isopropylspiro[oxirane-2,3'-thiochroman]-4'-one (**4p**): White solid, yield 45%, m.p. 124—126 ℃; UV-vis (MeOH) λ_{max}: 242 nm; ^{1}H NMR ($CDCl_3$, 600 MHz) δ: 1.16 [dd, J=24.01, 6.69 Hz, 6H, CH-(C**H**$_3$)$_2$], 1.67 [ddd, J=13.38, 6.70, 2.75 Hz, 1H, C**H**-(CH_3)$_2$], 2.88 (d, J=13.73 Hz, 1H, SCH_2), 2.97 (d, J=9.47 Hz, 1H, OCH), 3.82 (d, J=13.71 Hz, 1H, SCH_2), 7.15—7.20 (m, 1H, ArH), 7.30 (dd, J=8.70, 4.84 Hz, 1H, ArH), 7.81 (dd, J=9.16, 2.91 Hz, 1H, ArH); IR (KBr) v: 2923, 2852, 1676, 1600, 1559, 1488, 1467, 1382, 1269 cm^{-1}; MS (APCI) m/z: 253 $(M+H)^+$. Anal. calcd for $C_{13}H_{13}FO_2S$: C 61.89, H 5.19, S 12.71; found C 61.84, H 5.13, S 12.76.

Biological activity evaluation

***In vitro* anticancer activity** The anticancer activities of target compounds were evaluated *in vitro* on A-549 (human lung cancer cell lines), HeLa (human cervical cancer cell lines) and HepG2 (Human liver cancer cell lines) by measuring cell viability according to the MTT method described in the literature[26,27] with cisplatin as the positive control. The cells were seeded in RPMI 1640 medium (100 μL) in a 96-well plate at a concentration of 6000—7000 cells per well. After culturing for 12 h at 37 ℃ and 5% CO_2, cells were incubated with various concentrations of the samples for 24 h. Twenty microliter of MTT (5 mg/mL) was added and incubated with the cells for 4 h. The formazan product was dissolved by adding dimethyl sulfoxide (DMSO, 100 μL) to each well, and the plates were read at 570 nm. All measurements were performed in triplicate and each experiment was repeated at least three times. IC_{50} values were determined as the drug and sample concentration at 50% inhibition of the cell growth.

Acute toxicity test The experimental procedure was operated according to the reported procedure and guidelines.[28] Kunming mice, which weighed 20—25 g

(supplied from the Experimental Animal Center, Hebei Medical University), were randomly divided into 7 groups (10 in each group, male and female in half). Before the test, mice were fasted for 12 h. According to the results of previous experiments, LD100 was 3814.7 mg/kg, LD0 was 1000.0 mg/kg. Solutions of different concentrations were prepared, then the mice were fed with different doses of liquid by intragastric administration according to the weight, and the dose volume was 10 mL/kg. After administration, they were observed for 2 weeks continuously, toxic reactions and the death were recorded.

References

1 Chung, I. D.; Jung, E. Y.; Lee, Y. W.; Kim, S. H.; Ha, C. S.; Cho, W. J. *J. Polym. Sci. A: Polym. Chem.* **2000**, *38*, 4272.
2 Zhang, Y. B. *World Pestcides* **2005**, *27*, 6 (in Chinese).
3 Zocher, F.; Enzelberger, M. M.; Bornscheuer, U. T.; Hauer, B.; Wohlleben, W.; Schmid, R. D. *J. Biochem.* **2000**, *77*, 287.
4 Ozawa, Y.; Mori, K.; Matsui, M. *Agric. Biol. Chem.* **1974**, *38*, 1383.
5 Gao, X. P.; Li, B. G.; Zhou, J.; Li, W. B.; Zhong, S. D.; Tan, W. D. *Nat. Product. Res. Dev.* **1999**, *12*, 18 (in Chinese).
6 Yang, S.; Chen, J.; Guo, Z. *Mol. Cancer. Ther.* **2003**, *2*, 65.
7 Yang, B. K.; Jiang, C. H.; Hu, B. F. *Chin. J. Appl. Chem.* **1989**, *6*, 44 (in Chinese).
8 Wu, X. R.; Sun, B. Y. *Chin. J. Nosocomiol.* **2003**, *13*, 254 (in Chinese).
9 Huang, J. X. *Chin. J. Nosocomiol.* **2004**, *14*, 1435 (in Chinese).
10 Jerina, D. M.; Daly, J. W. *Science* **1974**, *185*, 573.
11 Akhtar, M. N.; Boyd, D. R. *J. Chem. Soc., Chem. Commun.* **1975**, 916.
12 Igarashi, M.; Midorikawa, H. *Bull. Chem. Soc. Jpn.* **1967**, *40*, 2624.
13 Nakazumi, H.; Ueyama, T.; Kitao, T. *J. Heterocycl. Chem.* **1984**, *21*, 193.
14 Khazi, I. A. M.; Gadad, A. K.; Mahajanshetti, C. S. *Indian J. Heterocycl. Chem.* **1995**, *4*, 243.
15 Hollshouser, M. H.; Loeffler, L. J. *J. Pharm. Sci.* **1982**, *71*, 715.
16 Qi, P.; Jin, Y. H.; Guo, C.; Fang, L. *Chin. J. Med. Chem.* **2003**, *13*, 205 (in Chinese).
17 Fang, L.; Guo, C.; Zhang, W. *J. Shenyang Pharm Univ.* **1998**, *15*, 8 (in Chinese).
18 Qi, P.; Jin, Y. H.; Guo, C.; Fang, L. *Chin. J. New Drugs* **2004**, *13*, 141 (in Chinese).
19 Nakib, T. A.; Bezjak, V.; Megan, M. J.; Chandy, R. *Eur. J. Med. Chem.* **1990**, *25*, 455.
20 Qi, P.; Jin, Y. H.; Guo, C.; Fang, L. *Chin. J. Med. Chem.* **2003**, *13*, 134 (in Chinese).
21 Zhu, Q. H.; Fang, L.; Zhang, G. L. *Chin. J. Med. Chem.* **2000**, *10*, 1 (in Chinese).
22 Fang, B. L.; Ma, Z. Y.; Yang, G. L.; Wang, G.; Tian, W.; Li, L. B. *Int. J. Chem.* **2010**, *2*, 143.
23 Tian, W.; Ma, Z. Y.; Yang, G. L.; Wang, G.; Fang, B. L.; Li, L. B. *Org. Chem. An Indian J.* **2010**, *6*, 8.
24 Ma, Z. Y.; Yang, G. L.; Yan, G. Y.; Zhu, S. G.; Guan, L. *Chin. J. Med. Chem.* **2008**, *18*, 170 (in Chinese).
25 Wang, G.; Yang, G. L.; Ma, Z. Y.; Tian, W.; Fang, B. L.; Li, L. B. *Int. J. Chem.* **2010**, *2*, 19.
26 Li, C. N.; Yang, C. L.; Liu, Y. X.; Yang, G. L.; Liu, T. F.; Lu, Q. *Chin. Sci. Bull.* **2010**, *55*, 3027 (in Chinese).
27 Ma, Z. Y.; Yan, G. Y.; Zhu, S. G.; Yang, G. L.; Luo, D. Q. *Chin. J. Chem.* **2009**, *27*, 987.
28 Fang, L.; Zhang, G. L.; Zhu, Q. H. *J. Shenyang Pharm. Univ.* **1998**, *15*, 157 (in Chinese).
29 Skehan, P.; Storeng, R.; Scudiero, D.; Monks, A.; McMahon, J.; Vistica, D.; Warren, J. T.; Bokesch, H.; Kenney, S.; Boyd, M. R. *J. Nat. Inst.* **1990**, *82*, 1107.
30 Ye, J. T.; Zheng, Y. L.; Liu, D. Y. *Chin. J. Chin. Mater. Med.* **2009**, *34*, 761 (in Chinese).
31 Pang, X. J.; Dong, Z.; Deng, J.; Fu, H. *Chin. J. Med. Guide* **2008**, *10*, 1067 (in Chinese).

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