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Synthesis and biological activity of flavanone derivatives

Lei Shi [a], Xiu E Feng [a], Jing Rong Cui [b], Lian Hua Fang [c], Guan Hua Du [c], Qing Shan Li [a,b,*]

[a] School of Pharmaceutical Science, Shanxi Medical University, Taiyuan 030001, China
[b] State Key Laboratory of Natural and Biomimetic Drugs, Peking University, Beijing 100083, China
[c] Institute of Materia Medica Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing 100050, China

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ABSTRACT

A series of new flavanone derivatives of farrerol was synthesized by a convenient method. The in vitro anti-tumor activity of these compounds was evaluated against human Bel-7402, HL-60, BGC-823 and KB cell lines, the protein tyrosine kinase (PTK) inhibitor activity was also tested. Their cytoprotective activity was tested using hydrogen peroxide (H_2O_2)-induced injury in human umbilical vein endothelial cells. Their in vitro anti-atherosclerosis activity was tested on vascular smooth muscle cells by the MTT method using tetrandrine as a positive contrast drug. The structures of all compounds synthesized were confirmed by ^1H, ^{13}C NMR and ESI-MS. Most of the compounds exhibited good pharmacological activity and the preliminary structure–activity relationships were described.

Flavanone is an important natural compound with significant potential to cure, treat and prevent tumor, senescence and cardiovascular diseases. Flavanones have been a potential source in the search for lead compounds and biologically active components and have been the focus of much researches and development in the last 30 years.[1,2] Flavanone and 2-OH flavanone markedly inhibited the invasion, motility and cell-matrix adhesion of A549 cells.[3] Lavandulyl flavanones showed potent β-secretase inhibitory activity which was strongly implicated in the cause of Alzheimer's disease.[4] Naringin has been shown to play a role in preventing the development of cardiovascular disease.[5] However, biological utilization rates are low because of their low content in natural products, which limits flavanone applications.[6] Thus, investigation of synthetic routes and chemical modification is a new direction in flavanone research. Furthermore, structure–activity relationship (SAR) studies of flavonoids are hampered by their structural diversity and the different mechanisms of their effects. Thus, SAR of flavanones is also an important area of research.

Farrerol is a flavanone-type compound and thus may have potential anti-hypertension and anti-atherosclerosis uses.[7,8] In recent research, our team found that farrerol has good in vitro anti-tumor activity against SGC-7901 cells in a time and dose-dependant manner. The IC_{50} value is 40 ± 0.4 μM for 24 h.[9] These interesting pharmacological activities prompted us to design a novel series of flavanone derivatives in attempt to improve their activity. Here, we report on the synthesis and in vitro activity of a series of farrerol derivatives, as well as the primary SAR. To investigate the positional effects of different groups on the B ring (Fig. 1) on bioactivity, compounds were prepared using a straightforward chemical approach, as shown in Scheme 1.

In a typical synthetic procedure, *m*-xylene was subjected to a nitration reaction using a mixture of hydrochloric acid and nitric acid to produce **1**. Compound **1** was mixed with Sn powder and hydrochloric acid and stirred at 60 °C until the solid was completely dissolved. The mixture was then heated at 100 °C for 3 h. The pH of the solution was adjusted to 3–4 by adding 40% NaOH and the solution was then refluxed for 24 h to afford **3**. To a solution of compound **3** in acetonitrile, zinc chloride and HCl gas were added and the solution was reacted at 0 °C until a solid was produced. The solid was redissolved in water and the solution was refluxed for 30 min to yield **4**. Finally, **4** was reacted with benzaldehyde, which has a different substituent group, and boracic acid in ethylene glycol at 130 °C to yield the target compounds.[10–13]

Generally the flavanone compounds could be prepared by the method used in this article and by the Claisen–Schmidt

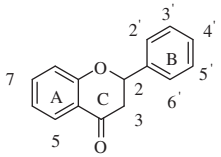

Figure 1.

* Corresponding author. Tel.: +86 351 4690322; fax: +86 351 4690114.
E-mail address: qingshanl@yahoo.com (Q.S. Li).

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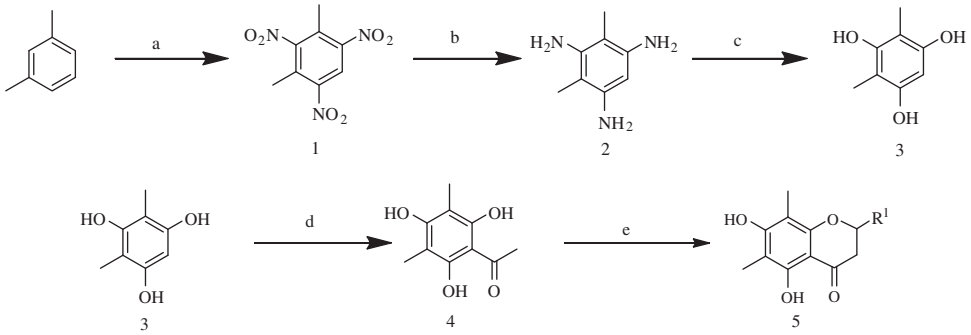


Scheme 1. Reagent and conditions: (a) HCl (37%), HNO_3 (69%), HNO_3 (98%), 110 °C, 87%; (b) Sn powder, HCl (37%), 90–100 °C; (c) NaOH, reflux, 24 h, 62%; (d) CH_3CN, $ZnCl_2$, HCl (gas), 0 °C, reflux, 58%; (e) EG, boracic acid, 130, 3 h, 20–30%.

condensation. We tried the above two methods to synthesize the target compounds. Compared with the Claisen–Schmidt condensation, the method used here has less steps with only one column chromatography step to obtain the target compounds, and the corresponding yield is similar to that obtained by the Claisen–Schmidt condensation method. So, we chosed this method to prepare flavanone derivatives.

To examine the changes of bioactivity resulting from different substituents and positions of the B ring, different benzaldehyde was used to yield the target compounds. To investigate the effect of changing the B ring to a heterocycle on bioactivity, different heterocyclic aldehydes were used to synthesize the target compounds according to the method described in Scheme 1. The structures of target compounds were described in Scheme 2.

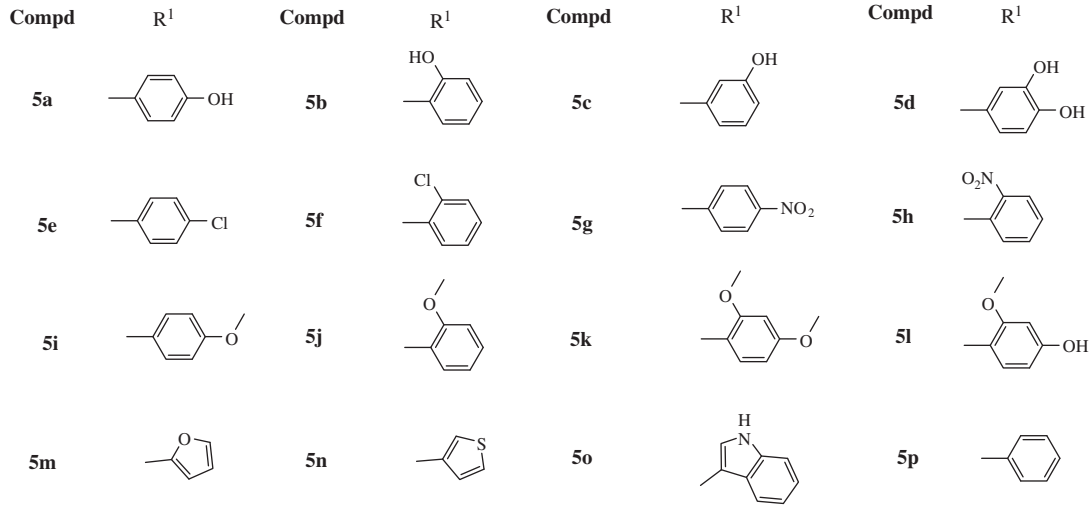
As presented in Table 1,[14] Compounds **5f**, **5h** exhibited significant cytotoxic activity against all of the human tumor cell lines except for the leukemia cell line HL-60, confirming the importance of *ortho* electron-withdrawing groups on the B ring for flavonoid cytotoxicity. Comparison of the activity of **5e**, **5g** and **5i** suggests that substitution of a *para* electron-withdrawing group or an electron-donating group decreases this activity. In addition, a hydroxy group plays an important role in the anti-tumor activity of these compounds. KB tumor cells seemed more sensitive to compounds with a single hydroxy group on the B ring in the *ortho*- or *meta*-position, as shown in compounds **5b–c**. A single *para*-substituted analog had lower anti-tumor potency compared with these of the other compounds. These results suggested that *ortho* and *meta* substituents may be crucial for anti-tumor activity.

Table 1
In vitro anti-tumor activities of compounds **5a–5p** (0.1 μmol L^{-1})

Compound	IR (%)			
	HL-60	BGC-823	Bel-7402	KB
Cisplatin	52.33	75.83	35.76	28.32
5a	−3.37	−23.52	−6.53	−7.21
5b	−6.47	−6.69	−1.80	44.54
5c	−4.04	−7.60	−5.48	28.66
5d	−4.85	−19.81	−7.68	45.30
5e	7.65	−22.45	−7.10	15.89
5f	2.94	36.98	18.70	28.63
5g	−0.46	−7.41	2.05	−9.11
5h	−6.16	52.00	27.18	29.04
5i	8.52	−28.89	15.15	−11.99
5l	−1.29	4.85	2.54	−18.83
5m	12.96	−9.68	3.27	−26.34
5n	10.82	50.71	11.59	16.07
5o	11.39	1.67	6.45	10.47
5p	ND[a]	13.51	2.63	3.64

[a] ND = Not detected.

The protein tyrosine kinase (PTK) plays critical roles in many of the signal transduction processes that control cell growth, differentiation, mitosis and apoptosis. So the inhibitory activity of PTK was believed to have the potential activity of anti-tumor.[15] In Table 2,[16] most of the compounds show certain activity, but Compounds **5e** and **5g** were inactive. Meanwhile, the heterocycle substituted analog, compounds **5m**, **5n** and **5o** have no activity on this assay. But compared to the results in Table 1, **5n** exhibited significant



Scheme 2. Target compounds.

Table 2
Inhibitory activity of PTK

Compound	PTK inhibitor activity IC_{50} (μM)	Compound	PTK inhibitor activity IC_{50} (μM)
5a	>200	**5i**	>100
5b	55	**5j**	>100
5c	51	**5k**	—[a]
5d	>50	**5l**	>200
5e	—	**5m**	—
5f	>50	**5n**	—
5g	—	**5o**	—
5h	>50	Genistein	13.6

[a] — = inactive.

Table 3
In vitro VSMC anti-vegetation activity and cytoprotective activity against H_2O_2-induced HUVEC injury

Compound	VSMC anti-vegetation activity IC_{50} (μM)	Cytoprotective activity EC_{50} (μM)
5a	—[a]	>20
5b	—	>20
5c	—	>20
5d	—	>20
5e	—	>50
5f	>20	>20
5g	—	>20
5h	9.9	>5
5i	>100	—
5j	—	>20
5k	—	—
5l	>100	>20
5m	6.7	>20
5n	—	>50
5o	—	>10
5p	>100	—
Tetrandrine	1.48	ND[b]

[a] — = inactive.
[b] ND = Not detected.

cytotoxic activity against BGC-823 cell line. So the mechanism of this compound may be not due to the inhibition of PTK.

The compounds were evaluated for their cytoprotective effects on HUVEC injury induced by H_2O_2.[17] The results in Table 3 show that most of the compounds exhibited moderate to good activity. Compound **5h** exhibited high cytoprotective activity. Subsequently, all the compounds were tested for their in vitro VSMC anti-vegetation activity.[18] The data indicate that compounds with good activity have an *ortho* electron-withdrawing substituent on the B ring. As a typical compound, **5h**, with an *ortho* nitro group on the B ring, respectively, had the strongest activity (IC_{50} = 9.9 μM). By contrast, compounds **5e** and **5g**, with a *para* electron-withdrawing group, displayed weak or no activity. Compounds **5b–d** also exhibited no VSMC anti-vegetation activity. The results indicate that an *ortho* electron-withdrawing substituent is crucial for the activity, whereas, a hydroxy group is unfavorable. Meanwhile, when the heterocycle is as the B ring instead of phenyl group, compound **5m** showed significant activity of VSMC anti-vegetation activity, but compounds **5n** and 5o were inactive.

HUVEC injury provokes VSMC vegetation. However, abnormal and excessive proliferation of VSMC plays a critical role in the genesis and development of hypertension and atherosclerosis.[19,20] It is interesting that we found that **5h** and **5m** exhibited high cytoprotective activity against HUVEC injury and good VSMC anti-vegetation activity. Thus, these compounds may be potent and specific therapeutic agents for cardiovascular disease.

In summary, a number of new flavanone derivatives were synthesized and evaluated for their in vitro anti-tumor activity and five compounds displayed good activity. Cytoprotective activity of the compounds on H_2O_2-induced HUVEC injury and in vitro VSMC anti-vegetation activity was also assayed, and the results showed that some compounds exhibited promising activities. Further in vivo tests of the compounds are under way.

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Supplementary data

Supplementary data (experimental procedures and characterization data for all final compounds) associated with this article can be found, in the online version, at doi:10.1016/j.bmcl.2010.07.090.

References and notes

1. Halsteen, B. *Biochem. Pharmacol.* **1983**, *32*, 1141.
2. Silberberg, M.; Gil-Izquierdo, A.; Combaret, L.; Remesy, C.; Scalbert, A.; Morand, C. *Biomed. Pharmacother.* **2006**, *60*, 529.
3. Yung-Chin, H.; Wu-Hsien, K.; Pei-Ni, C.; Horng-Rong, C.; Tseng-His, L.; Wei-En, Y.; Yih-Shou, H.; Shu-Chen, C. *Chem. Biol. Interact.* **2007**, *167*, 193.
4. Eun, M. H.; Young, B. R.; Hoi, Y. K.; Dong-Gyu, K.; Seong-Geun, H.; Jin, H. L.; Curtis-Long, M. J.; Seong, H. J.; Jae-Yong, P.; Ki, H. P. *Bioorg. Med. Chem.* **2008**, *16*, 6669.
5. Eo-Jin, L.; Gi-Seong, M.; Won-Seok, C.; Wun-Jae, K.; Sung-Kwon, M. *Food Chem. Toxicol.* **2008**, *46*, 3800.
6. Wang, W.; Wang, L. *J. Shen yang Med. College* **2002**, *4*, 115.
7. Tsukasa, I.; Junichi, K.; Sadamu, M. *Biochem. Syst. Ecol.* **2006**, *34*, 14.
8. Expectorant action of farrerol. *Chin. Med. J. (Engl.).* **1977**, *4*, 259.
9. Wang, X. J. Thesis, Shanxi Medical University, May 2009.
10. Institute of Materia Medica Chinese Academy of Medical Sciences. Chinese herbal medicine, 1973, 6, 31.
11. Wang, L.; Kong, J. Acta Northwest normal university. **1984**, *7*, 64.
12. Juntned, K. Y. T.; Junte, T. S. T. European Patent, 292576, 1988.
13. Tabkaa, A. C.; Murthi, K. K.; Kollol, P. *Ogr. Por. Res. Dev.* **1999**, *26*, 256.
14. Zeng, X. H.; Yang, X. D.; Zhang, Y. L.; Qing, C.; Zhang, H. B. *Bioorg. Med. Chem. Lett.* **2010**, *20*, 1844.
15. Srinivasan, M.; Trivadi, S. G. *Clin. Biochem.* **2004**, *37*, 618.
16. Han, G. L.; Shang, X. Y.; Du, G. H. *Chin. Pharmacol. Bulletin* **2005**, *21*, 628.
17. Wang, G. X.; Liu, Y.; Yang, C. L. *Chin. J. Arterioscler* **2009**, *17*, 193.
18. Hwa-Jin, C.; Ok-Jai, J.; Mi, J. C.; Sung-Yu, H.; Kwang-Hoe, C.; Sang, K. L.; Chung-Kyu, R. *Bioorg. Med. Chem. Lett.* **2005**, *15*, 3380.
19. Ross, R. *Nature* **1993**, *362*, 801.
20. Ross, R. *N. Engl. J. Med.* **1999**, *340*, 115.