A Platinum Open Access Journal for Organic Chemistry

Paper

Free to Authors and Readers | DOAJ Seal | *Arkivoc* **2024** (8) 202412244

Synthesis bis(arylidenes) containing heterocyclic chromones and α-pyronochromones

Thanh Ngoc Nguyen[1*], Giang Thu Thi Pham[1], and Van Thuy Ngo[1]

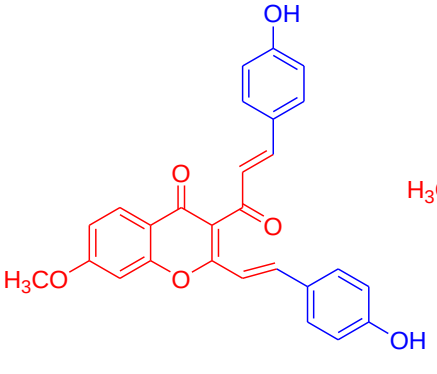
[1]*Faculty of Chemical Technology, Hanoi University of Industry, Bac Tu Liem District, Hanoi 10000, Vietnam*
Email: thanhnn@haui.edu.vn

Received mm-dd-yyyy **Accepted** mm-dd-yyyy **Published on line** mm-dd-yyyy

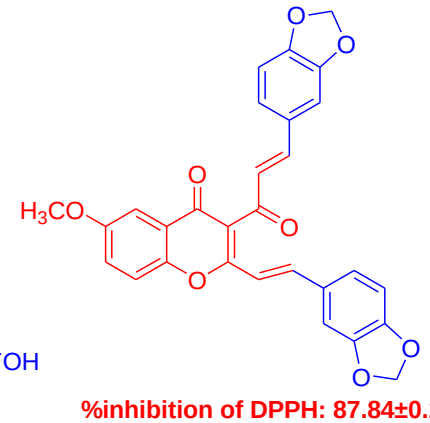
Dates to be inserted by editorial office

Abstract

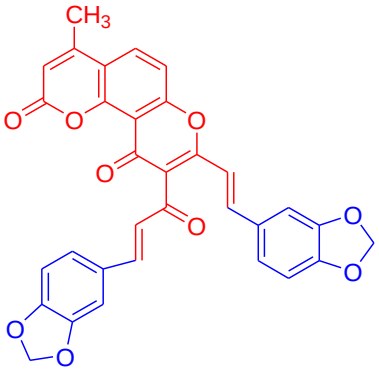
Five chromone and *α*-pyronochromone derivatives have been synthesized by reaction of *o*-hydroxyacetophenone and *o*-acetylhydroxycoumarin with anhydride acetic and sodium acetate as a catalyst. Then, these derivatives were reacted with some aromatic aldehyde and received fourteen new *bis*(arylidene). Furthermore, the intermediate product resulting from the reaction of 3-acetyl-7-methoxy-2-methylchromone and *p*-hydroxybenzaldehyde was successfully isolated and identified as 7-methoxy-2-methyl-3-[(2'*E*)-3'-(*p*-hydroxyphenyl)-prop-2'-enoyl]chromone. The antioxidant activity of all compounds was evaluated. The results show that 2-(4-hydroxystyryl)-3-((*E*)-3-(4-hydroxyphenyl)prop-2-enoyl)-7-methoxychromone has the highest antioxidant activity in tested compounds with inhibition of 88.48% at 50 µg/mL.



%inhibition of DPPH: 88.48±0.21
Ascorbic acid: 97.65±0.1



%inhibition of DPPH: 87.84±0.22
Ascorbic acid: 97.65±0.1



%inhibition of DPPH: 86.92±0.22
Ascorbic acid: 97.65±0.1

Keywords: Bis(arylidene); *α*,*β*-unsaturated ketones, chromone, *α*-pyronochromone, antioxidant activity

Introduction

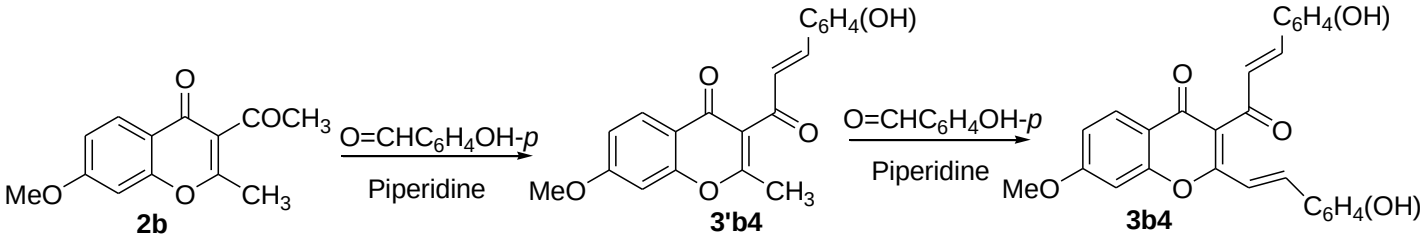
Compounds containing chromone heterocyclic are widespread and have many important biological functions in nature [1, 2]. These compounds have biological activity and the application is very diverse, These compounds have diverse biological activities and applications, such as anticancer [3], HIV-1 inhibition [4] antiplatelet [5], anti-inflammatory [6], antibacterial, antifungal [7], antioxidant [8], inhibits the enzyme acetylcholinesterase [9, 10]. Several chromone-containing derivatives have been used as drugs such as khelin, sodium cromoglycate [11, 12], crodimyl [13], diosmin [14, 15], and flavoxate [16].

Because compounds containing the chromone framework have a wide range of biological activities, therefore, synthetic studies to create new compounds containing these frameworks attract further research interest. This paper reports the synthesis results and antioxidant evaluation of *bis*(arylidene) compounds containing heterocyclic chromone and *α*-pyronochromone.

Results and Discussion

The Claisen-Schmidt condensation reaction of acetylchromone derivatives with aromatic aldehydes give *α,β*-unsaturated ketones. Weak base catalysts, for example, piperidine, trimethylamine, and pyridine are used for this reaction in chloroform or ethanol solvents. Meanwhile, strong inorganic catalysts, such as KOH and NaOH, produce lower yields of main product bis(arylidene) because chromone and *α*-pyronochromone undergo ring-opening under strong base conditions.

In many cases, *α,* and *β*-unsaturated ketones are not readily soluble under reaction conditions, they separated as solids and the reaction is stopped. With some of *α*, and *β*-unsaturated ketones dissolved in the reaction mixture, a condensation reaction can occur between the methyl group at position 2 on the *γ*-pyrone ring with aromatic aldehyde to form bis(arylidene). To support this hypothesis, 7-methoxy-2-methyl-3-[(2'*E*)-3'-(*p*-hydroxyphenyl)-prop-2'-enoyl]chromone (**3'b4)** was isolated and its structure was confirmed. Then, it was condensed with *p*-hydroxybenzaldehyde in a 1:1 mol ratio using Claisen-Schmidt condensation reaction conditions, to form **3b4** (see Scheme 1).



Scheme 1. Synthetic path to *α,β*-unsaturated ketone intermediate (**3'b4**) and bis(arylidene) **3b4**

In our opinion, the 2-methyl group is activated by the electron attraction of the 3-acetyl group. On the other hand, it also has a hyperconjugation effect with the vinyl ketone group, so it is highly activated. In addition, quantum chemical calculations by HyperChem Release 8.0 software have also shown that the charge density of the carbon atoms in the 2-methyl and 3-acetyl groups in the *γ*-pyrone ring is quite similar: **2a,** -0.265, and -0.201 respectively, whereas, with **2b**, the values are -0.264 and -0.201, with **2c**, -0.265 and -0.203, with **5**, -0.267 and -0.199, and with **8**, -0.265 and -0.204. As a result, the *γ*-pyrone ring's 2-methyl and 3-acetyl groups can react with aromatic aldehydes to form bis(arylidene) compounds.

Synthesis of bis(arylidene) is relatively slow, with the reaction times of up to 30-40 hrs, which was recorded by TLC and a molar ratio of **2a-c**, **5**, **8,** and aromatic aldehydes was 1:2. In this reaction, chloroform and 0.5 mol% of piperidine were used as a solvent and weak base catalyst, respectively. The reaction mixture was heated under reflux in a water bath. Based on the difference in solubility of the initial reactants and products, the reaction progress was monitored through the precipitate separated during the reaction. Bis(arylidene) derivatives containing chromone and α-pyronochromone are solids, have high melting points, and are difficult to dissolve in organic solvents such as ethanol, acetone, and DMF. The successful formation of bis(arylidene) having chromone and α-pyronochromone components from acetylmethylchromone derivatives and aromatic aldehyde was determined by IR, NMR, and MS spectral data.

The IR spectra of compounds bis(arylidene) showed the absorption peaks characterized by the carbonyl ketone group and carbonyl pyrone appearing in the 1621.83-1762.90 cm^{-1} region. The trans-configuration of α,β-unsaturated ketones, and vinyl was confirmed by IR absorption bands in the 958.50–997.89 cm^{-1} range.

The ^{1}H-NMR spectra of compounds bis(arylidene) showed two pairs of doublets-doublets with roof effect at 6.688-8.064 ppm with coupling constant in the range of 15.5-16.0 Hz. This showed the two vinyl groups have a trans configuration. Besides, the ^{1}H-NMR also showed resonance signals of other protons in chromone, α-pyronochromone, and substituted benzene. The identification of aromatic proton signals is based on spin-spin splitting (s, d, t,..), signal intensity (1H, 2H, 3H,..), and the effect of substituted groups attached to the aromatic ring.

The ^{13}C-NMR, HSQC, and HMBC spectra of **3a2, 3b2,** and **3b3** showed that they had the structures proposed for the compounds. In the ^{13}C-NMR, HSQC and HMBC spectra of **3a2, 3b2,** and **3b3** compounds also gave signals as expected. However, compound **9a** does not dissolve well in DMSO-d6 solvent, which is used for taking spectra so signals appeared in the low-resolution spectra.

Mass spectrometry of some bis(arylidene) gives ion-molecule peaks corresponding to the calculated formula. It is easy to detect the presence of chlorine or bromine in a molecule or ion by observing the intensity ratios of ions that differ by 2 amu. Such as **3b2** the molecular ion consists of three peaks at 476.0540 amu (100%); 478.0532 amu (69.70%); 480.0513 amu (14.33%). From the spectrum, it is evident that natural chlorine is made up of a combination of isotopes with atomic masses of 35 and 37 amu in the ratio of 3:1, as shown by the three peaks.

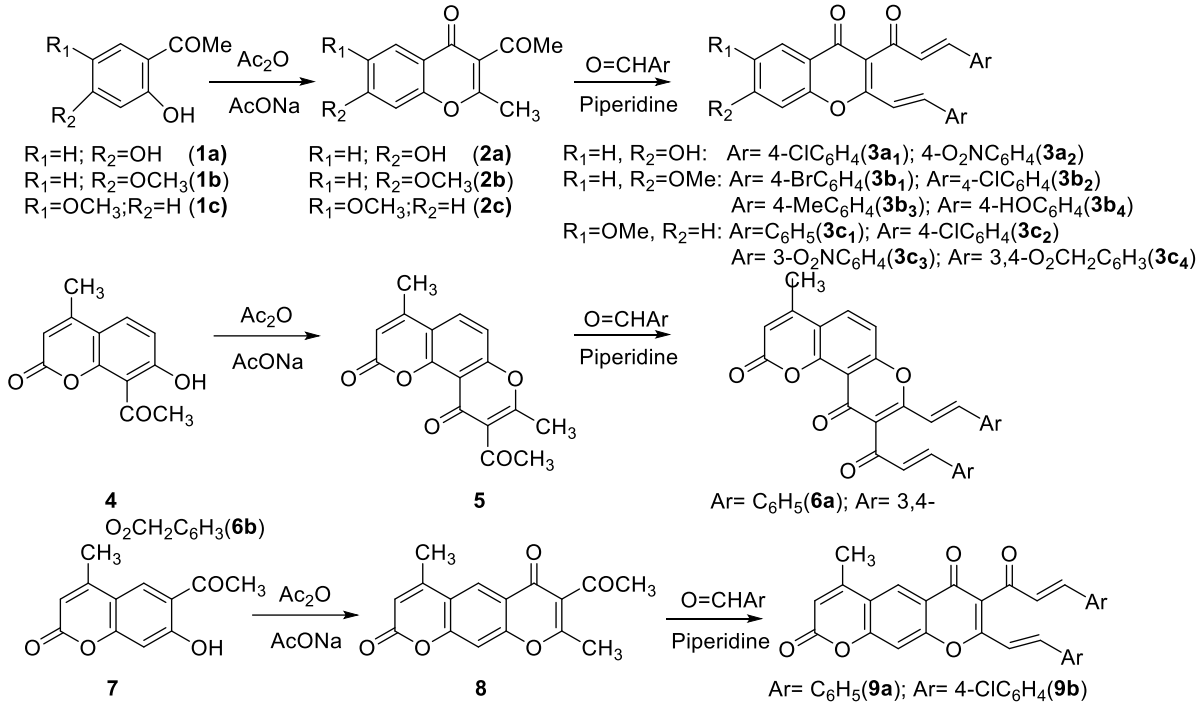
The antioxidant activity of synthesized compounds was tested using the DPPH radical scavenging method. The details of the results are given in Table 1. All of the test compounds exhibited good antioxidant properties, with the strongest being observed in compounds **3b4**, **3c4**, and **6b**. However, all the synthesized compounds were less potent than ascorbic acid as the reference. The potencies for the antioxidant activity of the strongest test compounds to the reference drug are in the following order: Ascorbic acid > **3b4** > **6b**> **3c4**.

Table 1. Inhibition (%) of test compounds

Comp.	Concentration (µg/mL)	% Inhibition	Comp.	Concentration (µg/mL)	% Inhibition
3a1	10	27.65±1.27	3c2	10	27.81±1.34
	50	60.34±0.24		50	56.82±0.22
3a2	10	30.25±1.41	3c3	10	29.26±1.28
	50	62.10±0.18		50	56..82±0.19
3b1	10	23.52±1.31	**3c4**	**10**	**37.62±1.26**

	50	54.13±0.14		**50**	**86.92±0.22**
3b2	10	28.58±1.27	6a	10	32.52±1.28
	50	59.55±0.22		50	62.56±0.18
3b3	10	26.63±1.28	**6b**	**10**	**38.28±1.30**
	50	51.45±0.24		**50**	**87.84±0.22**
3b4	**10**	**38.56±1.28**	9a	10	30.04±1.32
	50	**88.48±0.21**		50	60.85±0.19
3c1	10	25.92±1.28	9b	10	33.46±1.32
	50	50.45±0.20		50	58.88±0.21
Ascorbic acid*	**10**	**40.49±2.27**	**Ascorbic acid***	**10**	**40.49±2.27**
	50	**97.65±0.1**		**50**	**97.65±0.1**

Synthesis. Chemical reagents with high purity were bought from Merck Chemical Company. All reagents were of a grade for organic synthesis. Compounds **4** and **7** were synthesized according to literature procedures [18, 19]. Chromone and bis(arylidene) have been synthesized according to Scheme 2.



Scheme 2. Synthetic path to chromone, α-pyronochromone and bis(arylidene)

Conclusions

Fourteen new bis(arylidene) derivatives containing chromone and *α*-pyronochromone have been successfully synthesized. Their structures have been clarified by IR, NMR, and MS spectra data.

The intermediate product of synthesis of **3b4** has been isolated, which is 7-methoxy-2-methyl-3-[(2'*E*)-3'-(*p*-hydroxyphenyl)-prop-2'-enoyl]chrom-one. The structure of this product has been verified through the use of IR, ^{1}H-NMR, and MS spectra data.

The antioxidant activity of all compounds was evaluated. 2-(4-hydroxystyryl)-3-((*E*)-3-(4-hydroxyphenyl)prop-2-enoyl)-7-methoxychromone (**3b4**) has the highest antioxidant activity in tested compounds with inhibition of 88.48% at 50 µg/mL.

Experimental Section

General. Instrumentation Stuart SMP3 was used to confirm the melting point of the synthetic compounds. IR spectrum was analyzed by an Impact 410-Nicolet Spectrometer using KBr pellets. NMR spectra were obtained at 500 MHz for ^{1}H and 125 MHz for ^{13}C by an Avance AV500 Spectrometer made by Bruker, a company based in Germany., using d6- DMSO as the solvent and TMS as the internal standard. LC-MS data was recorded by LC-MS-ORBITRAP-XL, MS data was recorded by 5989B Hewlett–Packard Mass spectrometer, and HR-MS data was recorded by Micromass AutoSpec Premier Instrument (WATER, USA). We use Merck Kieselgel 60F254 pre-coated plates for conducting thin-layer chromatography (TLC).

The charge density was calculated by the HyperChem Release 8.0 software using the Semi-Empirical methods. All compounds are built and geometrically optimized by RM1 [17] with a convergence limit of 10^{-4} and an iteration limit of 50. The Polak-Ribiere algorithm was used with a termination condition of RMS gradient = 0.1000 kcal/(A mol), and RHF calculation.

General procedure 2a-c, 5, and 8. 0.01 mol of **1a-c**, **4,** and **7** was dissolved in 0.10 mol (9.5 mL) of anhydride acetic. A catalyst of 3.0 g sodium acetate was added to the solution mentioned above. The reaction was conducted with reflux conditions at a temperature range of 130-140^{o}C for 8 hours. Afterward, the solution's reaction was cooled, and the mixture was poured into a cup with 100 g of ice water. Once the separated product was filtered, washed with distilled water, and then dried. The final products **2a-c**, **5**, and **8** were obtained by recrystallizing them in an appropriate solvent. Physical, IR, NMR, and MS spectra data of compounds **2a-e** are reported as follows:

3-Acetyl-7-hydroxy-2-methylchromone (2a). From **1a** (0.01 mol, 1.52 gram): Yield 0.654 g (30%) of **2a**, crystallized from EtOH as white crystals. Mp 185-187^{o}C. IR: $\upsilon_{C=O}$ (γ-pyrone) at 1686.95 cm^{-1}, $\upsilon_{C=O}$ (acetyl) at 1621.82 cm^{-1}, υ_{OH} at 3492.92 cm^{-1}, and 3393.00 cm^{-1}. ^{1}H-NMR: 2.367 (3H, s, CH_3), 2.480 (3H, s, $COCH_3$); 6.826 (1H, d, *J* 2.0 Hz, H_{benzo}); 6.928 (1H, dd, *J* 8.5 and 2.0 Hz, H_{benzo}); 7.891 (1H, d, *J* 8.5 Hz, H_{benzo}). MS (*m/z*): 218 (M^+, 70.0%), 203(82.3%), 137(100%)… Anal. Calcd for $C_{12}H_{10}O_4$.

3-Acetyl-7-methoxy-2-methyl chromone (2b). From **1b** (0.01 mol, 1.66 gram): Yield 0.696 g (30%) of **2b**, crystallized from EtOH as pale-yellow crystals. Mp 164-166^{o}C. IR: $\upsilon_{C=O}$ (γ-pyrone, acetyl) at 1699.52 cm^{-1} (br). ^{1}H-NMR: 2.403 (3H, s, CH_3); 2.502 (3H, s, $COCH_3$); 3.901 (3H, s, OCH_3); 7.085(1H, dd, *J* 8.0 and 2.5 Hz, H_{benzo}); 7.144 (1H, d, *J* 3.0 Hz, H_{benzo}); 7.961 (1H, d, *J* 8.5 Hz, H_{benzo}). HR-MS (*m/z*): 231.9436 (M^+, 92.6%); 213.9506 (100%); 150.9495 (91.8%),… Anal. Calcd for $C_{13}H_{1.}$

3-Acetyl-6-methoxy-2-methylchromone (2c). From **1c** (0.01 mol, 1.66 gram): Yield 0.464 g (20%) of **2c**, crystallized from EtOH as brown crystals. Mp 130-132^{o}C. IR: $\upsilon_{C=O}$ (γ-pyrone) at 1690.24 cm^{-1}, $\upsilon_{C=O}$ (acetyl) at 1647.10 cm^{-1}. ^{1}H-NMR: 2.453 (3H, s, CH_3); 2.535 (3H, s, $COCH_3$); 3.920 (3H, s, OCH_3); 7.366 (1H, dd, *J* 9.0 Hz and 3.0 Hz, H_{benzo}); 7.502 (1H, d, *J* 3.0 Hz, H_{benzo}); 7.521 (1H, d, *J* 9.0 Hz, H_{benzo}). HRMS (*m/z*): 232.0728 (M^+, 90.2%); 217.0473 (89.4%); 151.0417 (100%),.. Anal. Calcd for $C_{13}H_{12}O_4$.

9-Acetyl-4,8-dimethylpyrano[6,5-*f*]chromene -2,10-dione (5). From **4** (0.01 mol, 2.18 gram)**:** Yield: 1.287 gram (45%) of **5**, crystallized from DMF as yellow-brown crystals. Mp 245-246^{o}C. IR: $\upsilon_{C=O}$ (pyrone) at 1726.38 cm^{-1}, 1697.65 cm^{-1} and CO-acetyl at 1648.51 cm^{-1}. ^{1}H-NMR: 2.400 (3H, s, $CH_{3\gamma\text{-pyrone}}$); 2.484 (3H, d, *J* 1.5 Hz, $CH_{3\alpha}$-

$_{pyrone}$); 2.496 (3H, s, $COCH_3$); 6.494 (1H, d, *J* 1.0 Hz, $H_{\alpha\text{-pyrone}}$); 7.564 (1H, d, *J* 9.0 Hz, H_{benzo}); 8.130 (1H, d, *J* 9.0 Hz, H_{benzo}). HRMS (*m/z*): 284.0687 (M^+, 100%), 269.0514 (52.4%), 241.0584 (40.2%),… Anal. Calcd for $C_{16}H_{12}O_5$.

3-Acetyl-2,6-dimethyl-4*H*-pyrano[3,2-*g*]chromone-8-one (8). From **7** (0.01 mol, 2.18 gram)**:** Yield: 1.221 gram (43%) of **8**, crystallized from acetonitrile as pale-yellow crystals. Mp 246-247°C. IR: CO-lactone, γ-pyrone, acetyl at 1734.73 cm^{-1} and 1695.78 cm^{-1}. ^{1}H-NMR: 2.446 (3H, s, $CH_{3\gamma\text{-pyrone}}$); 2.507(3H, s, $CH_{3\alpha\text{-pyrone}}$); 2.518(3H, s, $COCH_3$); 6.483(1H, s, $H_{\alpha\text{-pyrone}}$); 7.619(1H, s, H_{benzo}), and 8.301(1H, s, H_{benzo}). MS (*m/z*):· 284 (M^+, 75.3%), 269 (100%), 203 (56.2%),… Anal. Calcd for $C_{16}H_{12}O_5$.

Synthesis of intermediates 7-methoxy-2-methyl-3-[(2'*E*)-3'-(*p*-hydroxyphenyl)-prop-2'-enoyl]chromone (**3'b4**). A mixture of 3-acetyl-7-methoxy-2-methylchromone **2b** (5 mmol, 1.16 g) and *p*-hydroxybenzaldehyde (5 mmol, 0.61 g) in 30 mL ethanol, piperidine (0.5 mL; 0.5 mol%) was heated under reflux conditions for 20 hrs. After the reaction is complete, the mixture is cooled using ice. Next, the resulting precipitate is filtered and washed with distilled water followed by cold ethanol. The obtained product was air-dried at room temperature and recrystallized from ethanol to get the compound **3'b4**. Physical, IR, ^{1}H-NMR (supporting data 1), and HR-MS spectra data of compound **3'b4** are reported as follows: Yield 0.588 g (35%) of **3b'4** as pale-yellow crystals. Mp 287-289°C. IR: ν_{CO} (γ-pyrone, ketone) at 1684.15 cm^{-1}; $\gamma_{CH=trans}$ at 975.81 cm^{-1}. ^{1}H-NMR: 2.575 (3H, s, CH_3), 3.820 (3H, s, OCH_3), 6.940 (1H, d, *J* 8.5 and 2.5 Hz, H_{benzo}), 6.961 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$), 6.981 (1H, d, *J* 2.5 Hz, H_{benzo}), 7.014 (2H, d, *J* 9.0 Hz, H_{phenyl}), 7.643 (2H, d, *J* 8.5 Hz, H_{phenyl}), 7.777 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$), 7.899 (1H, d, *J* 8.5 Hz, H_{benzo}). HR-MS: *m/z* 336.1212 (M^+, 82.5%), 293.0979 (100%), 243.0785 (28.8%),… Anal. Calcd for $C_{20}H_{16}O_5$.

General procedure for preparing bis(arylidene). A mixture of **2a-c, 5, and 8** (5 mmol) and aromatic aldehydes (10 mmol) in chloroform (30 mL), piperidine (0.5 mL; 0.5 mol%) was heated under reflux conditions for 30-40 hrs. At first, when heating the reaction mixture the substances dissolve, then products are formed and separated in the form of a precipitate while heating. Filter the precipitate, and wash it several times with hot chloroform. The obtained products were air-dried at room temperature, and recrystallized from DMF to get the compounds **3a1-2**; **3b1-4**; **3c1-4**; **6a**; **6b**; **9a,** and **9b**. Physical and IR, MS, and NMR spectra data of *bis*(arylidene) derivatives are reported as follows.

2-(4-Chlorostyryl)-3-((*E*)-3-(4-chlorophenyl) prop-2-enoyl)-7-hydroxychromone (3a1). Yield 0.962 g (40%) of **3a1** as yellow crystals. Mp 295-297°C. IR: ν_{CO} (γ-pyrone, ketone) 1667.58 cm^{-1}, $\gamma_{CH=trans}$ at 960.82 cm^{-1}. ^{1}H-NMR: 6.944 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$); 6.955 (1H, dd, *J* 8.0 and 3.0 Hz, H_{benzo}); 7.030 (1H, d, *J* 2.0 Hz, H_{benzo}); 7.204 (1H, d, *J* 16.5 Hz, H_{vinyl}); 7.485 (2H, d, *J* 8.0 Hz, H_{phenyl}); 7.492 (2H, d, *J* 8.0 Hz, H_{phenyl}); 7.620 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.713 (2H, d, *J* 8.0 Hz, H_{phenyl}); 7.767 (2H, d, *J* 8.5 Hz, H_{phenyl}); 7.843 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$); 7.896 (1H, d, *J* 8.0 Hz, H_{benzo}). MS (*m/z*): M^+ 462 (4.8%); 464 (3.1%); 463 (1.3%),.. Anal. Calcd for $C_{26}H_{16}Cl_2O_4$.

2-(4-Nitrostyryl)-3-((*E*)-3-(4-nitrophenyl) prop-2-enoyl)-7-hydroxychromone (3a2). Yield 0.962 g (35%) of **3a2** as yellow crystals. Mp 297-299°C. IR: ν_{CO} (γ-pyrone, ketone) 1663.07 cm^{-1}; $\gamma_{CH=trans}$ at 978.82 cm^{-1}. ^{1}H-NMR: 6.974 (1H, d, *J* 8.5 Hz, H_{benzo}); 7.055 (1H, s, H_{benzo}); 7.179 (1H, *J* 16.0 Hz, $H_{vinylketone}$); 7.401 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.759 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.911 (1H, d, *J* 8.5 Hz, H_{benzo}); 7.964 (1H, *J* 16.0 Hz, $H_{vinylketone}$); 7.972 (2H, d, *J* 8.0 Hz, H_{phenyl}); 8.016 (2H, d, *J* 9.0 Hz, H_{phenyl}); 8.248 (2H, d, *J* 8.0 Hz, H_{phenyl}); 8.259 (2H, d, *J* 8.0 Hz, H_{phenyl}). ^{13}C-NMR: 102.924; 116.016; 116.363; 122.341; 123.086; 124.472; 124.608; 127.400; 129.621; 130.212; 131.887; 136.702; 141.405; 141.586; 141.857; 148.217; 148.632; 157.336; 159.288; 164.113; 175.403; 192.573.

2-(4-Bromostyryl)-3-((*E*)-3-(4-bromophenyl) prop-2-enoyl)-7-methoxychromone (3b1). Yield 0.991 g (35%) of **3b1** as yellow crystals. Mp 196-198°C. IR: ν_{CO} (γ-pyrone, ketone) 1621.83 (br) cm^{-1}; $\gamma_{CH=trans}$ at 978.62 cm^{-1}. ^{1}H-

NMR: 3.957 (3H, s, OCH_3); 6.989 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$); 7.107 (1H, d, *J* 9.0 Hz, H_{benzo}); 7.231 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.337 (s, 1H, H_{benzo}); 7.708-7.617 (8H, m, H_{phenyl}); 7.685 (1H, d, *J* 15.5 Hz, H_{vinyl}); 7.864 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$); 7.958 (1H, d, *J* 9.0 Hz, H_{benzo}).

2-(4-Chlorostyryl)-3-((*E*)-3-(4-chlorophenyl) prop-2-enoyl)-7-methoxychromone (3b2). Yield 0.904 g (38%) of **3b2** as yellow crystals. M.p: 200-202°C. IR: ν_{CO} (γ-pyrone, ketone) 1668.78 cm^{-1}; $\gamma_{CH=trans}$ at 977.86 cm^{-1}. ^{1}H-NMR: 3.966 (3H, s, OCH_3); 6.995 (1H, d, *J* 15.5 Hz, $H_{vinylketone}$); 7.098 (1H, dd, *J* 9.0 and 2.0 Hz, H_{benzo}); 7.199 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.285 (1H, d, *J* 2.0 Hz, H_{benzo}); 7.467 (4H, t, *J* 8.0 and 8.0 Hz, H_{phenyl}); 7.601 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.724 (2H, d, *J* 8.5 Hz, H_{phenyl}); 7.665 (2H, d, *J* 8.5 Hz, H_{phenyl}); 7.843 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$); 7.978 (1H, d, *J* 9.0 Hz, H_{benzo}). ^{13}C-NMR: 55.839; 100.605; 114.351; 116.719; 118.216; 121.785; 126.196; 128.300; 128.577; 128.712; 129.182; 129.869; 133.073; 133.386; 134.390; 134.908; 137.012; 142.321; 156.476; 159.032; 164.170; 174.418; 191.384. HR-MS (*m/z*): M^+ 476.0540 (100%); 478.0523 (69.7%); 80.0513 (14.33%),.. Anal. Calcd for $C_{27}H_{18}Cl_2O_4$.

2-(4-Methylstyryl)-3-((*E*)-3-(4-methylphenyl) prop-2-enoyl)-7-methoxychromone (3b3). Yield 0.915 g (43%) of **3b3** as pale yellow crystals. Mp 150-152°C. IR: ν_{CO} (γ-pyrone, ketone) 1665.08 cm^{-1}; $\gamma_{CH=trans}$ at 958.50 cm^{-1}. ^{1}H-NMR: 2.327 (3H, s, CH_3); 2.351 (3H, s, CH_3); 3.956 (3H, s, OCH_3); 6.886 (1H, d, *J* 15.5 Hz, $H_{vinylketone}$); 7.092 (1H, dd, *J* 8.5 and 1.5 Hz, H_{benzo}); 7.145 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.245 (4H, t, *J* 7.5 and 7.0 Hz, H_{phenyl}); 7.342 (1H, d, *J* 1.5 Hz, H_{benzo}); 7.573 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.635-7.541 (4H, m, H_{phenyl}); 7.848 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$); 7.951 (1H, d, *J* 8.5 Hz, H_{benzo}). ^{13}C-NMR: 20.979; 21.037; 56.183; 100.732; 114.806; 116.403; 116.850; 121.648; 126.457; 127.028; 127.959; 128.783; 129.599; 129.751; 131.555; 131.919; 138.637; 140.351; 141.018; 145.038; 156.851; 159.347; 164.322; 174.849; 192.383. HR-MS (*m/z*): M^+ 436.1267 (100%); 345.0842 (37.5%); 317.0919 (18.3%),… Anal. Calcd for $C_{29}H_{24}O_4$.

2-(4-Hydroxystyryl)-3-((*E*)-3-(4-hydroxyphenyl)prop-2-enoyl)-7-methoxychromone (3b4). Yield 0.858 g (39%) of **3b4** as yellow crystals. Mp 239-241°C. IR: ν_{CO} (γ-pyrone, ketone) 1697.24 cm^{-1}; $\gamma_{CH=trans}$ at 968.42 cm^{-1}, ν_{OH} at 3169.99 (*br*) cm^{-1}. ^{1}HNMR: 3.953 (3H, s, OCH_3); 6.688 (1H, d, *J* 15.5 Hz, $H_{vinylketone}$); 6.795 (2H, d, *J* 8.5 Hz, H_{phenyl}); 6.861 (2H, d, *J* 8.5 Hz, H_{phenyl}); 6.968 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.078 (1H, dd, *J* 8.5 and 2.5 Hz, H_{benzo}); 7.322 (1H, d, *J* 2.5 Hz, H_{benzo}); 7.493 (2H, *J* 8.5 Hz, H_{phenyl}); 7.520 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.580 (2H, d, *J* 9.0 Hz, H_{phenyl}); 7.775 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$); 7.938 (1H, d, *J* 8.5 Hz, H_{benzo}); 10.073 (2H, s, br, OH).

6-Methoxy-3-((*E*)-3-phenylprop-2-enoyl)-2-styrylchromone (3c1). Yield 0.714 g (35%) of **3c1** as pale yellow crystals. Mp 185-187°C. IR: ν_{CO} (γ-pyrone, ketone) 1664.76 cm^{-1}; $\gamma_{CH=trans}$ at 975.22 cm^{-1}. ^{1}H-NMR: 3.883 (3H, s, OCH_3); 6.978 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$); 7.220 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.463-7.418 (7H, m, H_{phenyl}); 7.499 (1H, dd, *J* 9.0 and 3.0 Hz, H_{benzo}); 7.665 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.674 (2H, d, *J* 7.0 Hz, H_{phenyl}); 7.750-7.731 (2H, m, $H_{phenyl,\ benzo}$); 7.790 (1H, d, *J* 9.0 Hz, H_{benzo}); 7.892 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$).

2-(4-Chlorostyryl)-3-((*E*)-3-(4-chlorophenyl) prop-2-enoyl)-6-methoxychromone (3c2). Yield 0.954 g (40%) of **3c2** as pale yellow crystals. Mp 210-212°C. IR: ν_{CO} (γ-pyrone, ketone) 1671 cm^{-1}; $\gamma_{CH=trans}$ at 978 cm^{-1}. ^{1}H-NMR: 3.883 (3H, s, OCH_3); 6.991 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$); 7.225 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.447 (1H, d, *J* 3.0 Hz, H_{benzo}); 7.496 (5H, dd, *J* 8.5 and 8.5 Hz, H_{phenyl}); 7.653 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.720 (2H, d, *J* 8.5 Hz, H_{phenyl}); 7.774 (3H, d, *J* 9.0 Hz; $H_{benzo,\ phenyl}$); 7.886 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$).

2-(3-Nitrostyryl)-6-methoxy-3-((*E*)-3-(3-nitrophenyl)prop-2-enoyl)chromone (3c3). Yield 1.120 g (45%) of **3c3** as pale yellow crystals. Mp 266-267°C. IR: ν_{CO} (γ-pyrone, ketone) 1665.93 cm^{-1}; $\gamma_{CH=trans}$ at 968.22 cm^{-1}; $\nu_{s,\ as}$ (NO_2) 1532.07, 1354.51 cm^{-1}. ^{1}H-NMR: 3.892 (3H, s, OCH_3); 7.186 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$); 7.405 (1H, d, *J* 16.5 Hz, H_{vinyl}); 7.463 (1H, s, H_{benzo}); 7.533 (1H, dd, *J* 9.0 and 2.5 Hz, H_{benzo}); 7.725 (2H, t, *J* 8.0 and 7.5 Hz, H_{phenyl}); 7.787 (1H, d, *J* 9.0 Hz, H_{benzo}); 7.845 (1H, d, *J* 16.0 Hz, H_{vinyl}); 8.064 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$); 8.257-8.168 (4H, m, H_{phenyl}); 8.521 (1H, s, H_{phenyl}); 8.558 (1H, s, H_{phenyl}).

3-((*E*)-3-(3,4-Methylenedioxyphenyl)prop-2-enoyl)-2-((*E*)-3,4-methylenedioxystyryl)-6-methoxychromone (3c4). Yield 0.620 (25%) of **3c4** as yellow crystals. Mp: 180-182°C. IR: ν_{CO} (γ-pyrone, ketone) 1662.23 cm^{-1}; $\gamma_{CH=trans}$ at 989.42 cm^{-1}. ^{1}H-NMR: 3.880 (3H, s, OCH_3); 6.081 (2H, s, O_2CH_2); 6.084 (2H, s, O_2CH_2); 6.774 (1H, d, *J* 16.5 Hz, $H_{vinylketone}$); 6.953 (1H, d, *J* 8.0 Hz, H_{phenyl}); 6.985 (1H, d, *J* 8.0 Hz, H_{phenyl}); 7.046 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.198 (1H, d, *J* 8.0 Hz, H_{phenyl}); 7.225 (1H, d, *J* 8.0 Hz, H_{phenyl}); 7.299 (1H, s, H_{phenyl}); 7.402 (1H, s, H_{phenyl}); 7.436 (1H, d, *J* 3.0 Hz, H_{benzo}); 7.482 (1H, dd, *J* 9.0 and 3.0 Hz, H_{benzo}); 7.536 (d, 1H, *J* 16.0 Hz, H_{vinyl}); 7.749 (1H, d, *J* 9.0 Hz, H_{benzo}); 7.786 (1H, *J* 16.0 Hz, $H_{vinylketone}$). LC-MS: $[M+H]^+$ 497.15 (100%); $[M+Na]^+$ 519.06 (36.2%),… Anal. Calcd for $C_{29}H_{20}O_8$.

4-Methyl-9-((*E*)-3-phenylprop-2-enoyl)-8-styrylpyrano[6,5-*f*]chromene-2,10-dione (6a). Yield 1.037 g (45%) of **6a** as pale yellow crystals. Mp 310-312°C. IR: ν_{CO} 1715.77, 1641.99 cm^{-1}; $\gamma_{CH=trans}$ at 997.89 cm^{-1}. ^{1}H-NMR: 2.503 (3H, s, CH_3); 6.507 (1H, d, *J* 1.0 Hz, $H_{\alpha\text{-pyrone}}$); 6.919 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$); 7.195 (1H, d, *J* 16.5 Hz, H_{vinyl}); 7.454 (6H, m, H_{phenyl}); 7.758-7.689 (4H, m, H_{phenyl}); 7.724 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.798 (1H, d, *J* 9.0 Hz, H_{benzo}); 7.946 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$); 8.245 (1H, d, *J* 9.0 Hz, H_{benzo}). HR-MS (*m/z*): M^+ 460.1456 (24.6%); 372.1292 (100%); 243.1067 (48.6%),... Anal. Calcd for $C_{30}H_{20}O_5$.

9-((*E*)-3-(3,4-Methylenedioxyphenyl)prop-2-enoyl)-8-((*E*)-3,4-methylenedioxystyryl)-4-methylpyrano[6,5-*f*]chromene -2,10-dione (6b). Yield 1.072 g (39%) of **6b** as yellow crystals. Mp 309-311°C. IR: ν_{CO} 1723.73, 1640.43 cm^{-1}, $\gamma_{CH=trans}$ at 970.46 cm^{-1}. ^{1}H-NMR: 2.503 (3H, s, CH_3); 6.084 (4H, s, $2O_2CH_2$); 6.494 (1H, d, *J* 1.0 Hz, $H_{\alpha\text{-pyrone}}$); 6.718 (1H, d, *J* 15.5 Hz, $H_{vinylketone}$); 6.956 (1H, d, *J* 8.0 Hz, H_{phenyl}); 6.989 (1H, *J* 8.0 Hz, H_{phenyl}); 7.025 (1H, d, *J* 16.5 Hz, H_{vinyl}); 7.216 (2H, td, *J* 7.5 and 1.5 Hz, H_{phenyl}); 7.329 (1H, d, *J* 1,5 Hz, H_{phenyl}); 7.419 (1H, d, *J* 1.5 Hz, H_{phenyl}); 7.593 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.743 (d, 1H, *J* 9.0 Hz, H_{benzo}); 7.824 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$); 8.222 (1H, d, *J* 9.0 Hz, H_{benzo}).

6-Methyl-3-((*E*)-3-phenylprop-2-enoyl)-2-styryl-4*H*-pyrano[3,2-*g*]chromone-8-one (9a). Yield 0.874 g (38%) of **9a** as pale yellow crystals. Mp 286-287°C. IR: ν_{CO} 1744.99, 1627.21 (br) cm^{-1}, $\gamma_{CH=trans}$ at 986.80 cm^{-1}. ^{1}H-NMR: 2.540 (3H, s, CH_3); 6.530 (1H, d, *J* 1.5 Hz, $H_{\alpha\text{-pyrone}}$); 6.979 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$); 7.211 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.460-7.431 (7H, m, H_{phenyl}); 7.711 (1H, *J* 16.0 Hz, H_{vinyl}); 77.743-7.678 (3H, m, H_{phenyl}); 7.849 (1H, s, H_{benzo}); 7.958(1H, d, *J* 16.0 Hz, $H_{vinylketone}$); 8.355 (1H, s, H_{benzo}). MS (*m/z*) M^+ 460 (32.4%); 383 (26.2%); 127 (72.4%); 77 (100%),… Anal. Calcd for $C_{30}H_{20}O_5$.

3-((*E*)-3-(4chlorophenyl)prop-2-enoyl)-2-(4-chlorostyryl)-6-methyl-4*H*-pyrano[3,2-*g*]chromone-8-one (9b). Yield 1.190 g (45%) of **9a** as yellow crystals. Mp 303-304°C. IR: ν_{CO} 1762.90, 1626.98 (br) cm^{-1}, $\gamma_{CH=trans}$ at 977.75 cm^{-1}. ^{1}H-NMR: 2.541 (3H, s, CH_3); 6.514 (1H, s, $H_{\alpha\text{-pyrone}}$); 7.003 (1H, d, *J* 16.0 Hz, $H_{vinylketone}$); 7.207 (1H, d, *J* 16.0 Hz, H_{vinyl}); 7.494 (4H, t, *J* 9.0 Hz, H_{phenyl}); 7.810-7.662 (6H, m, H_{vinyl}, $H_{vinylketone}$, H_{phenyl}); 7.951 (1H, s, H_{benzo}); 8.365 (1H, s, H_{benzo}).

Antioxidant testing. The free radical scavenging activity was determined by the DPPH assay [20, 21]. Test compounds are dissolved in DMSO solution to specific concentrations. DPPH was diluted in MeOH to the appropriate concentration. 10 µL of test compounds were incubated with 190 µL of DPPH solution, incubated at 37°C for 20 minutes then the absorbance was measured at 517 nm using the UV-vis spectrophotometer (ELISA). Ascorbic reference was used to control stability and evaluate equivalent inhibitory activity. Tests were repeated 3 times. The optical density was recorded and % inhibition was calculated using the formula given below: Inhibition of DPPH activity (%)= $100 - [(OD_s) / (OD_c) \times 100]$ (1)

Where OD_s is the average optical density of the test sample and OD_c is the average optical density of the control sample.

Supplementary Material

Copies of NMR spectra are provided in supplementary information

References

1. S.K. Sharma, S. Kumar, K. Chand, A. Kathuria, A. Gupta1, and R. Jain. *Current Medicinal Chemistry*. **2011**, 18, 3825-3852.
https://doi.org/10.2174/092986711803414359

2. Gaspar A, Matos MJ, Garrido J, Uriarte E, Borges F. *Chem Rev*. **2014**, 114:4960–4992.
https://doi.org/10.1021/cr400265z

3. Agullo G, Gamet-Payrastre L, Manenti S, Viala C, Rémésy C, Chap H, Payrastre B. *Biochem Pharmacol*. **1997,** 53:1649–1657.
https://doi.org/10.1016/S0006-2952(97)82453-7

4. Ungwitayatorn J., Samee W., Pimthon J. *J. Mol. Struct*. **2004**, 689, 99.
http://dx.doi.org/10.1016/j.molstruc.2003.10.036

5. Jackson SP, Schoenwaelder SM. *Nat Rev Drug Discov*. **2003**, 2:775–789.
https://doi.org/10.1038/nrd1198

6. Khan KM, Ambreen N, Mughal UR, Jalil S, Perveen S, Choudhary MI. *Eur J Med Chem*. **2010**, 45:4058–064.
https://doi.org/10.1016/j.ejmech.2010.05.065

7. Khan KM, Ahmad A, Ambreen N, Amyn A, Perveen S, Khan SA, Choudhary MI. *Lett Drug Des Discov*. **2009**, 6:363–373.
https://doi.org/10.2174/1570180810906050363

8. Miean KH, Mohamed S. *J Agric Food Chem.* **2001**, 49:3106–3112.
https://doi.org/10.1021/jf000892m

9. Viayna E, Sabate R, Muñoz-Torrero D. *Curr Top Med Chem.* **2014**, 13:1820–1842.
https://doi.org/10.2174/15680266113139990139

10. Shaikh, S., Dhavan, P., Ramana, M. M. V., & Jadhav, B. L. *Molecular Diversity.* **2021**, 25, 811–825.
https://doi.org/10.1007/s11030-020-10060-y

11. Edwards, A.M.; Howell, J.B.L. *Clin. Exp. Allergy*. **2000**, 30, 756-774.
https://doi.org/10.1046/j.1365-2222.2000.00879.x

12. Brown FJ, Bernstein PR, Cronk LA, Dosset DL, Hebbel KC, Maduskuie TP Jr, Shapiro HS, Vacek EP, Yee YK, Willard AK, Krell RD, Snyder DW. *J Med Chem.* **1989**, 32: 807–826.
https://doi.org/10.1021/jm00124a014

13. Bruneton, J. Pharmacognosy, Phytochemistry, and Medicinal Plants; English Translation by Hatton, C. K.; *Lavoisier Publishing: Paris*. **1995**; pp. 265.

14. Silvestro L, Tarcomnicu I, Dulea C, et al. *Anal Bioanal Chem*. **2013**; 405(25):8295-8310.
DOI: 10.1007/s00216-013-7237-y

15. Freag MS, Elnaggar YS, Abdallah OY. *Int J Nanomedicine*. **2013**; 8:2385-2397.
https://doi.org/10.2147/IJN.S45231

16. https://www.drugs.com/pro/flavoxate.html

17. Rocha, G.B., R.O. Freire, A.M. Simas, and J.J.P. Stewart. *J. Comp. Chem*. **2006**, 27(10): 1101-1111.
https://doi.org/10.1002/jcc.20425

18. J. Trykowska Konc, E. Hejchman, H. Kruszewska, I. Wolska, D. Maciejewska. *Eur.J.Med. Chem.* **2011**, 46, 2252-2263.
https://doi.org/10.1016/j.ejmech.2011.03.006
19. Hejchman, E.; Kruszewska, H.; Maciejwska, D.; Sowirka-Taciak, B.; Tomczyk, M.; Sztokfisz-Ignasiak, A.; Jankowski, J.; Mlynarczuk-Biały, I. *Monats. Chem*. **2019**, 150, 255–266.
https://doi.org/10.1007/s00706-018-2325-5
20. Shahat, A. A., et al. *Planta Med.* **2003**, 69(12): 1153-1156.
https://doi.org/10.1055/s-2003-818009
21. Okawa, M., et al. *Biol Pharm Bull.* **2001**, 24(10): 1202-1205.
https://doi.org/10.1248/bpb.24.1202