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RESEARCH ARTICLE

Synthesis and NMR spectroscopy investigations of functionalized spiropyranochromenediones and their spirothiadiazole derivatives

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Abstract: This study discusses the synthesis of spiropyranoneoflavones and the modification of obtained compounds at the exocyclic oxygen atom. Kabbe cyclization of 6-acetyl-7-hydroxy-8-methyl-4-phenyl-2H-chromene-2-one with cyclohexanone or cyclopentanone in the presence of pyrrolidine provided 10'-methyl-6'-phenyl-8'*H*-spiro[cyclohexane(cyclopentane)-1,2'-pyrano[3,2-*g*]chromene]-4',8'(3'*H*)-diones. Their new functionalized derivatives with thiosemicarbazide residues were synthesized. Acetylation of obtained thiosemicarbazones with acetic anhydride proceeded via cyclization of thiosemicarbazide fragment into 1,3,4-thiadiazole ring to give corresponding *N*-[3"-acetyl-10'-methyl-8'-oxo-6'-phenyl-3'*H*,3"*H*,8'*H*-dispiro[cyclohexane-1,2'-pyrano[3,2-*g*]chromene-4',2"-[1,3,4]-thiadiazol]-5"-yl]acetamide and *N*-[3"-acetyl-10'-methyl-8'-oxo-6'-phenyl-3'*H*,3"*H*,8'*H*-dispiro[cyclopentane-1,2'-pyrano[3,2-*g*]chromene-4',2"-[1,3,4]thiadiazol]-5"-yl]acetamide. The structure of the obtained compounds was confirmed by NMR spectroscopy.

Keywords: spiro compounds; neoflavones; spiropyranochromenediones; NMR spectroscopy; heteronuclear correlation.

Introduction

Derivatives of 4-phenylcoumarin, also known as neoflavones, are a research topic of significant interest. Neoflavones are common in plants – as of today, more than 160 compounds of this class have been obtained from natural sources [1] - and both natural neoflavones and their synthetic analogs demonstrate a wide range of biological activities, including antitumor [2], cytotoxic [3], antioxidant [4], antibacterial [5, 6], insecticidal [7], and many others.

In this study, we discuss the synthesis of spiropyranoneoflavone derivatives, the modification of obtained compounds at the exocyclic oxygen atom, and the confirmation of their structure with NMR spectroscopy data.

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Two common synthetic approaches to the synthesis of pyranocoumarins exist: coumarin ring formation in a pyran system, and pyran ring formation in a coumarin system. Earlier, we have obtained spirodihydropyranocoumarins *via* Pechmann condensation of 3,4-dihydrospirocyclohexane-chomane-7-ol [8]. 4-Phenylcoumarin with a fused dimethylpyran ring was synthesized through Kabbe cyclization of 6-acetyl-7-hydroxy-8-methylneoflavone with acetone [9].

Results and Discussion

In this study, we used Kabbe cyclization of 6-acetyl-7-hydroxy-8-methyl-4-phenyl-2*H*-chromene-2-one (**1**) with cyclohexanone and cyclopentanone in the presence of pyrrolidine to obtain the corresponding linear spiropyranoneoflavones **2** and **3** (Scheme 1).

The subsequent heating of compounds **2**, **3** with thiosemicarbazide led to thiosemicarbazones **4**, **5**. The substitution takes place at the exocyclic oxygen atom of the spiropyran ring, and not the benzopyran-2-one system, even in the presence of a threefold excess of the nucleophilic reagent. Acetylation of thiosemicarbazones **4**, **5** proceeds *via* cyclization of thiosemicarbazide fragment into

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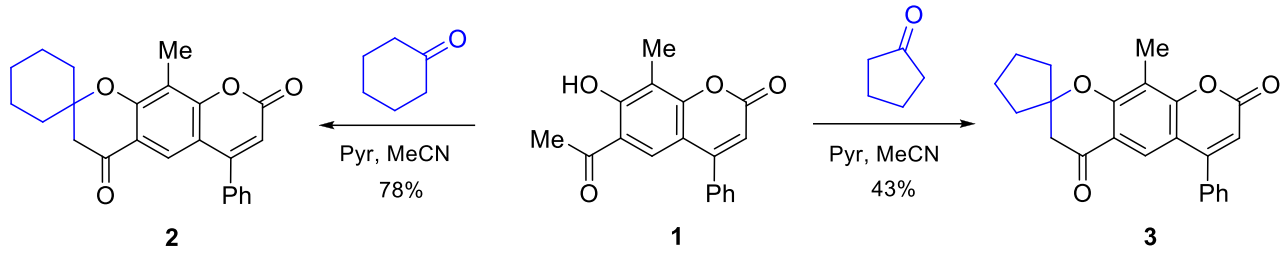
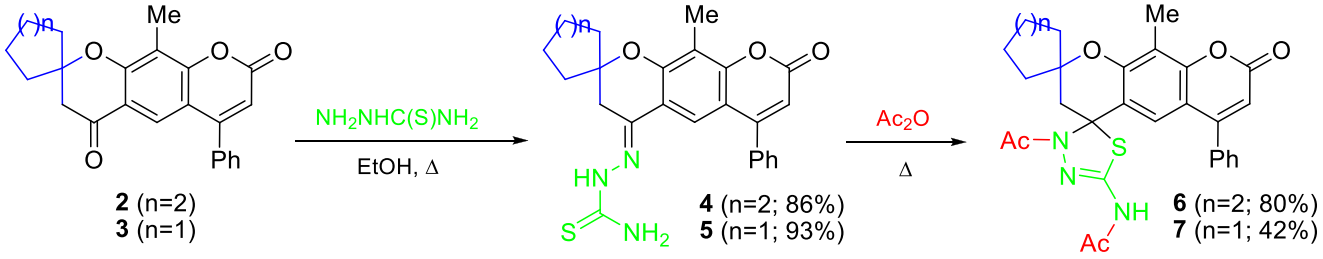


Scheme 1. The synthesis of linear spiropyranoneoflavones **2** and **3**.



Scheme 2. The synthesis of spirothiadiazole derivatives of spiropyranoneoflavones **6** and **7**.

Table 1. ^{1}H chemical shifts (δ/ppm) of compounds **2-7** in DMSO-d$_6$.

No	H-3'[a]	H-5'[a]	7'-CH_2	8'-$(CH_2)_n$[c]	10'-CH_3[a]	H-2″, 6"[b]	H-3″, 4", 5"[c]	Other signals
2	6.22	7.69	2.72[a]	1.56-1.96[j]	2.35	7.44	7.57	–
3	6.22	7.57	2.86[a]	1.69-2.01[i]	2.29	7.45	7.46	–
4	6.17	7.94	2.88[a]	1.61-1.83[j]	2.31	7.51	7.57	7.06 (s, NH), 8.22 (s, NH), 10.52 (s, NH)
5	6.18	7.95	3.00[a]	1.73-1.90[i]	2.26	7.51	7.55	7.08 (s, NH), 8.21 (s, NH), 10.48 (s, NH)
6	6.23	7.26	2.55[d], 3.16[e,f]	1.48-1.88[j]	2.25	7.39	7.53	2.06 (s, *Ac*N-*TDA*), 1.99 (s, *Ac*NH-*TDA*), 11.64 (s, NH)
7	6.13	7.25	2.40[d], 3.55[e,f]	1.75-1.85[i]	2.25	7.40	7.55	2.00 (s, *Ac*N-*TDA*), 2.08 (s, *Ac*NH-*TDA*), 11.50 (s, NH)

[a] s.; [b] d., J = 8.8 Hz; [c] m.; [d] *pro-S*; [e] *pro-R*; [f] 2×d., J = 14.0 Hz; [j] n = 5; [i] n = 4

Table 2. ^{13}C chemical shifts (δ/ppm) of compounds **2-7** in DMSO-d$_6$.

No	C-2'	C-3'	C-4'	C-4a'	C-5'	C-5a'	C-6'	C-7'	C-8'	C-9a'	C-10'	C-10a'
2	160.8	112.7	155.4	111.6	123.2	116.9	196.8	49.8	78.4	156.4	119.0	155.4
3	160.4	112.6	155.2	111.4	123.	116.7	196.5	49.5	78.6	156.1	119.1	155.4
4	159.9	112.8	155.4	111.8	123.6	113.9	146.5	36.1	78.3	156.8	119.2	153.4
5	150.0	112.7	155.4	111.6	123.9	112.9	146.5	34.6	78.1	153.3	119.2	153.4
6	160.4	112.4	155.3	113.5	123.4	122.7	74.2	44.9	77.6	153.7	113.5	152.8
7	160.8	112.5	155.4	111.6	123.8	121.7	74.2	44.8	77.4	153.5	118.9	153.2

No	8'-$(CH_2)_n$	10'-CH_3	C-1"	C-2",6"	C-3",5"	C-4"	Other signals
2	37.7, 34.5, 23.8, 22.4, 21.2	8.6	140.0	126.4	129.5	130.1	–
3	37.9, 34.5, 23.6, 21.9	8.6	140.0	126.6	128.9	130.1	–
4	37.3, 35.6, 24.1, 22.6, 21.3	8.9	134.2	128.9	128.7	130.4	181.3 (C=S)
5	38.4, 36.5, 24.6, 21.1	8.6	136.1	128.4	128.6	130.3	181.3 (C=S)
6	37.8, 33.3, 24.7, 22.0, 21.1	8.8	135.6	129.1	129.5	130.6	23.0, 167.8 (*Ac*), 22.0, 170.2 (*Ac*NH), 143.7 (*TDA*-C-5)
7	38.6, 34.1, 23.6, 21.3	8.9	136.3	129.1	129.7	130.4	23.1, 168.3 (*Ac*), 22.7, 171.4 (*Ac*NH), 144.3 (*TDA*-C-5)

1,3,4-thiadiazole ring, and gives corresponding *N*-[3''-acetyl-10'-methyl-8'-oxo-6'-phenyl-3'*H*,3''*H*,8'*H*-dispiro[cyclohexane-1,2'-pyrano[3,2-*g*]chromene-4',2''-[1,3,4]thiadiazol]-5''-yl]acetamide (**6**) and *N*-[3''-acetyl-10'-methyl-8'-oxo-6'-phenyl-3'*H*,3''*H*,8'*H*-dispiro[cyclopentane-1,2'-pyrano[3,2-*g*]chromene-4',2''-[1,3,4]thiadiazol]-5''-yl]-acetamide (**7**) (Scheme 2).

Table 3. Heteronuclear correlation data for compound **6**.

^{1}H NMR signal, δ/ppm	^{13}C NMR signals which correlate with ^{1}H NMR signal, δ/ppm	
	HMQC	HMBC
1.55	21.7	
1.48-1.65	22.0	24.7, 44.9, 77.6, 153.7
	24.7	
1.65-1.88	33.3	143.7, 170.2
	37.8	
1.99	22.0	170.2, 143.7
2.06	23.0	167.8
2.25	8.8	153.7, 152.8, 123.4, 113.5
2.55	44.9	123.4, 74.2
3.16	44.9	77.6, 74.2, 37.8, 33.3
6.23	112.4	160.4, 135.6, 112.4, 123.4
7.26	123.4	155.3, 153.7, 152.8, 123.4, 113.5, 74.2
7.39	129.1	155.3, 130.6, 129.1
7.53	129.5, 130.6	135.6, 129.5, 129.1
11.64	–	170.2, 143.7

^{1}H and ^{13}C NMR spectra were measured to confirm the structures of obtained compounds. ^{1}H NMR spectra (Table 1) of compounds **2**-**7** match the assumed structures; however, due to their relative complexity and instability of lactone derivatives toward nucleophilic reagent, ^{13}C NMR spectra were measured (Table 2).

In ^{13}C NMR spectra of compounds **2**, **3**, signals at 196.8-196.5 ppm are attributed to the C-6 atom; this carbon atom is observed at 146.5 ppm for compounds **4**, **5**, and at 77.4-78.6 ppm for compounds **2**-**7**, which is typical for a spirocarbon (C-8). The second spirocyclic carbon in compounds **6**, **7** gives a signal at 74.2 ppm. Cyclohexane or cyclopentane ring gives a wide signal in ^{1}H NMR spectra at 1.48-2.01 ppm. A less expressed correlation of C-7 atom's chemical shift with the nature of the exocyclic substituent at C-6 can also be observed (Table 2). ^{1}H and ^{13}C NMR spectra of remaining elements of molecules **2**-**7** are only slightly affected by the substituent at C-6, but even though the spectra do not contradict the proposed structures, a reliable correlation of signals cannot be established. It becomes possible after 2D heteronuclear ^{1}H-^{13}C correlation experiments: *via* one bond (HMQC) and 2-3 bonds (HMBC). Table 3 summarizes cross-peak coordinates, found in HMQC and HMBC spectra of the compound **6**.

A scheme below shows the assignments and the most important HMBC correlations upon which the assignments were based (Figure 1).

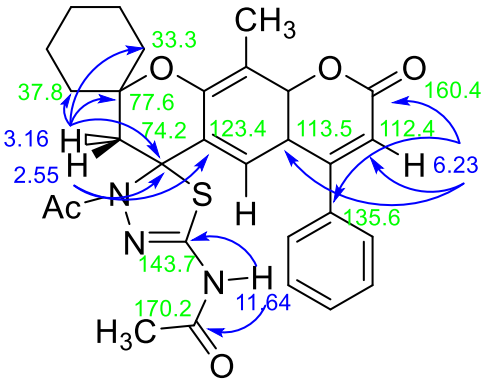


Figure 1. The principal HMBC correlations for compound **6**.

Conclusions

To summarize, we have demonstrated an easy and effective approach to the synthesis of 10-methyl-4-phenyl-2*H*-spiro[cyclohexane(or cyclopentane)-1',8-pyrano-[3,2-*g*]chromene]-2,6(7*H*)-diones *via* condensation of 6-acetyl-7-hydroxyneoflavone with cyclohexanone and cyclopentanone. The further reaction of spiropyranoneoflavones with thiosemicarbazide led to the obtainment of thiosemicarbazones. It is worth noting that only C-6 position, and not C-2 position, was affected in the course of the reaction. Acetylation of thiosemicarbazones led to the synthesis of dispiro[cyclohexane(or cyclopentane)-1',8-pyrano[3,2-*g*]chromene-4',6-[1,3,4]thiadiazol]-5'-yl]acetamides. The structure of obtained compounds was confirmed by ^{1}H and ^{13}C NMR spectra data of two-dimensional ^{1}H-^{13}C shift correlation experiments *via* one bond and multiple bonds.

Experimental section

Reaction flow and identity of obtained compounds were controlled with TLC on Merck F_{254} plates using chloroform:methanol (9:1, *v*/*v*) and (95:5, *v*/*v*) systems as eluents. Melting points were determined using a *Kofler*-type Leica Galen III micro hot stage microscope. NMR spectra were recorded on a Mercury-400 spectrometer (spectrometer frequency for ^{1}H: 400 MHz, ^{13}C: 100 MHz) from DMSO-d_6 solutions. The TMS signal was used as an internal standard. HMQC spectra were acquired as 128×32 data matrices with spectral ranges: for ^{1}H – 4 kHz, for ^{13}C – 21 kHz; mixing time corresponds to $^1J_{CH}$ = 140 Hz. HMBC spectra were acquired as 400×32 data matrices with spectral ranges: for ^{1}H – 4 kHz, for ^{13}C – 21 kHz; mixing time corresponds to $^{2\text{-}3}J_{CH}$ = 8 Hz. Spectra were measured with detection on protons and gradient selection of signals.

Elemental analyses for C, H, and N were conducted using Perkin-Elmer C, H, N Analyzer. Their results were found to be in good agreement (± 0.2%) with the calculated values. Mass spectra were recorded on Agilent 1100 LC/MSD instrument with chemical ionization (CI).

*6-Acetyl-7-hydroxy-8-methyl-4-phenyl-2H-chromene-2-one (**1**)* was synthesized earlier [10].

*General procedure for the preparation of spiropyranoneoflavones (**2, 3**).*

To a solution of 2.94 g of **1** (10 mmol) in 20 ml acetonitrile, 2.0 ml pyrrolidine (25 mmol) and 7 mmol of a corresponding ketone (cyclohexanone, 7.2 ml; cyclopentanone, 5.8 ml) were added. The reaction mixture was kept at 45 °C for 8 h (the reaction completeness was monitored with TLC). The resulting solution was diluted with ice H_2O, acidified to pH 5 and filtered. Recrystallization from acetonitrile afforded the product.

*10'-Methyl-6'-phenyl-8'H-spiro[cyclohexane-1,2'-pyrano[3,2-g]chromene]-4',8'(3'H)-dione (**2**, $C_{24}H_{22}O_4$).*

Yield 2.93 g (78.2%), mp 246-248 °C.
^{1}H NMR spectra are shown in Table 1.
^{13}C NMR spectra are shown in Table 2.
MS *m/z* 375.4 $[M+H]^+$.

*10'-Methyl-6'-phenyl-8'H-spiro[cyclopentane-1,2'-pyrano[3,2-g]chromene]-4',8'(3'H)-dione (**3**, $C_{23}H_{20}O_4$).*

Yield 1.56 g (43.3%), mp 201-202 °C.
^{1}H NMR spectra are shown in Table 1.
^{13}C NMR spectra are shown in Table 2.
MS *m/z* 361.4 $[M+H]^+$.

*General procedure for the preparation of 10-methyl-4-phenyl-2H-spiro[cyclohexane(cyclopentane)-1',8-pyrano-[3,2-g]chromene]-2,6(7H)-dione thiosemicarbazones (**4, 5**).*

To a solution of a corresponding compound **2** or **3** (3 mmol) in 10 ml ethanol, an alcoholic solution of 3.5 mmol of a thiosemicarbazide with 1 ml HCl was added dropwise. The reaction mixture was kept on a water bath for 3-5 h. The formed precipitate was filtered and crystallized from propan-2-ol.

*(E/Z)-2-(10'-methyl-8'-oxo-6'-phenyl-8'H-spiro[cyclohexane-1,2'-pyrano[3,2-g]chromen]-4'(3'H)-ylidene)-hydrazine-1-carbothioamide (**4**, $C_{25}H_{25}N_3O_3S$).*

Yield: 1.24 g (86.8%), mp 258-259 °C.
^{1}H NMR spectra are shown in Table 1.
^{13}C NMR spectra are shown in Table 2.
MS *m/z* 448.6 $[M+H]^+$.

*(E/Z)-2-(10'-methyl-8'-oxo-6'-phenyl-8'H-spiro[cyclopentane-1,2'-pyrano[3,2-g]chromen]-4'(3'H)-ylidene)-hydrazine-1-carbothioamide (**5**, $C_{24}H_{23}N_3O_3S$).*

Yield: 1.14 g (93.4%), mp 244-246 °C.
^{1}H NMR spectra are shown in Table 1.
^{13}C NMR spectra are shown in Table 2.
MS *m/z* 434.5 $[M+H]^+$.

*General procedure for the preparation of N-[3'-acetyl-10-methyl-2-oxo-4-phenyl-3'H,2H-dispiro[cyclohexane(cyclopentane)-1',8-pyrano[3,2-g]chromene-4',6-[1,3,4]thiadiazol]-5'-yl]acetamides (**6, 7**).*

A solution of 1 mmol of a corresponding thiosemicarbazone **4, 5** in 10 ml acetic anhydride was refluxed at a water bath for 6-7 h. The reaction mixture was left to cool to room temperature and then poured on ice. The formed precipitate was filtered and crystallized from aqueous propan-2-ol (20:80, *v/v*).

*N-(3''-acetyl-10'-methyl-8'-oxo-6'-phenyl-3'H,3''H,8'H-dispiro[cyclohexane-1,2'-pyrano[3,2-g]chromene-4',2''-[1,3,4]thiadiazol]-5''-yl)acetamide (**6**, $C_{29}H_{29}N_3O_5S$).*

Yield: 0.45 g (80.8%), mp 204-206 °C.
^{1}H NMR spectra are shown in Table 1.
^{13}C NMR spectra are shown in Table 2.
MS *m/z* 532.6 $[M+H]^+$.

*N-(3''-acetyl-10'-methyl-8'-oxo-6'-phenyl-3'H,3''H,8'H-dispiro[cyclopentane-1,2'-pyrano[3,2-g]chromene-4',2''-[1,3,4]thiadiazol]-5''-yl)acetamide (**7**, $C_{28}H_{27}N_3O_5S$).*

Yield: 0.16 g (42.0%), mp 196-198 °C.
^{1}H NMR spectra are shown in Table 1.
^{13}C NMR spectra are shown in Table 2.
MS *m/z* 518.5 $[M+H]^+$.

Notes

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The authors declare no conflict interest.

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Синтез та дослідження ЯМР спектроскопії функціоналізованих спіропіранохромендіонів та їх спіротіадіазольних похідних

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Резюме: Дослідження присвячено синтезу спіропіранонеофлавонів та проведенню модифікації отриманих сполук за екзоциклічним атомом Оксигену. Циклізація Каббе 6-ацетил-7-гідрокси-8-метил-4-феніл-2*H*-хромен-2-ону з циклогекссаноном або циклопентаноном в присутності піролідину завершувалась отриманням 10'-метил-6'-феніл-8'*H*-спіро[циклогексан(циклопентан)-1,2'-пірано-[3,2-*g*]хромен]-4',8'(3'*H*)-діонів. Також отримані їх нові функціоналізовані похідні з залишком тіосемікарбазиду. Наступне ацетилювання отриманих тіосемікарбазонів в оцтовому ангідриді супроводжувалось циклізацією тіосемікарбазидного фрагменту в 1,3,4-тіадіазольний цикл та завершувалось отриманням *N*-[3"-ацетил-10'-метил-8'-оксо-6'-феніл-3'*H*,3"*H*,8'*H*-диспіро[циклогексан-1,2'-пірано[3,2-*g*]хромен-4',2"-[1,3,4]тіадіазол]-5"-іл]ацетаміду та *N*-[3"-ацетил-10'-метил-8'-оксо-6'-феніл-3'*H*,3"*H*,8'*H*-диспіро[циклопентан-1,2'-пірано[3,2-*g*]хромен-4',2"-[1,3,4]тіадіазол]-5"-іл]ацетаміду відповідно. Будова отриманих сполук доведена методами ЯМР спектроскопії.

Ключові слова: спіропохідні, неофлавоноїди, спіропіранохромендіони, спектроскопія ЯМР, гетероядерна кореляція.