Supporting Information

Novel nitric oxide donors of phenylsulfonylfuroxan and 3-benzyl coumarin derivatives as potent antitumor agents

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I. *In vitro* anti-proliferative assay.

For MTT assay: the *in vitro* anti-proliferation of the chemical compounds was measured by the MTT reagent, as described in the literature[1]. Briefly, 3~5×10^3 cells in 100 μL of medium per well were plated in 96-well plates. After incubated for 24 h, the cells were treated with different concentration of tested compound or DMSO (as negative control) for 48 h. Then, the medium with compound or DMSO was replaced with 200 μL of fresh medium containing 10 % MTT (5 mg/mL in PBS) in each well and incubated at 37 ºC for 4 h. Last, the MTT-containing medium was removed and 150 μL of DMSO per well was added to dissolve the formazan crystals newly formed. Absorbance of each well was determined by a microplate reader (Synergy H4, Bio-Tek) at a 490 nm wavelength. The inhibition rates of proliferation were calculated with the following equation:

Inhibition ratio = (OD_{DMSO}-$OD_{compd.}$)/(OD_{DMSO}-OD_{blank})×100%.

The concentrations of the compounds that inhibited cell growth by 50% (IC_{50}) were calculated using Graph Pad Prism version 6.0.

For MTS assay: 8~20×10^3 cells of hematological tumor cell lines in 90 μL of medium per well were plated in 96-well plates. For solid tumor cell lines, 2~3×10^3 cells were plated in 96-well plates for 16h before compound treatment. Cells were treated with 0.2% DMSO or serial dilutions of compounds from 10 mM stock solutions in DMSO (0.2% final

concentration of DMSO) for 72 h. A CellTiter 96® AQueous Non-Radioactive Cell Proliferation Assay kit (Promega, Madison, USA) was employed. 20 μL of MTS reagent was added to each well and incubated at 37 ºC for 4 h. Plates were read for absorbance at 490 nm using a Spectra max Molecular Devices microplate reader. The inhibition rates of proliferation were calculated the same with MTT assay using Graph Pad Prism version 5.0

II. Method for Nitrite measurement *in vitro*.

The levels of NO released by tested compounds in the cells are presented as that of nitrite and were determined by the Griess Reagent (Beyotime, China), according to the literature with some modifications[2]. Briefly, cells (1×10^7 per 10 cm dish) were treated with a 100 μM concentration of each compound for 150 min. Subsequently, the cells were harvested and lysed with 100 μL RIPA lysis buffer (Beyotime, China) for 30 min on ice. The cell lysates were mixed with Griess for 30 min at dark place, followed by measurement by a microplate reader (Synergy H4, Bio-Tek) at a 540 nm wavelength. The cells treated with diluent were used to determine the background levels of nitrite production, while sodium nitrite at different concentrations was measured to generate standard curve.

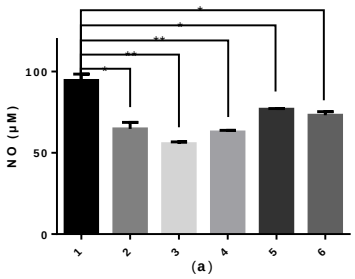


Figure S1. Incecullar NO produced by **2**-**6** in A2780 cells (**a**)Results were indicated as the Mean ± SEM of two independent experiments. (*) $p < 0.05$; (**) $p < 0.01$.

The intracellular NO releasing capability of these furoxan/3-phenyl coumarin hybrids (**2**-**4**) and their seco-B-ring derivatives (**5**-**6**) was determined and presented as that of nitrite in the cell lysates using a Griess assay with some modifications, with lead compound **1** as a reference. As expected in **Figure** S**1**, **2**-**6** could also produce various levels of nitrite intracellularly with the releasing percentage range from 55.6 to 76.8 μM, much less than that of lead compound **1** (94.6 μM).

III. Inhibition activities of colony formation.

We tested the colony formation ability of cells after treatment with different concentration of compounds **2**-**6**. As shown in **Figure S2**, the exposure of A2780 and MDA-MB-231/Gem cell lines to 1.25 ~ 40 nM of compounds **2**-**6** resulted in a significant inhibition of colony

formation. Among them, the inhibition of MDA-MB-231/Gem colony formation was almost complete at doses above 5 nM, while it didn't improve much in A2780, compared to that of lead compound **1**.

Colony formation assay. 1×10^3 cells per well were seeded in six-well plates at a single cell density. 48 h later, the cells were treated with DMSO or different concentrations of testing compounds for 48 h. Then the medium was replaced with fresh medium to allow cell growth for at least one week. The cells were fixed with methyl alcohol for 15 min and stained by gentian violet staining for 30 min.

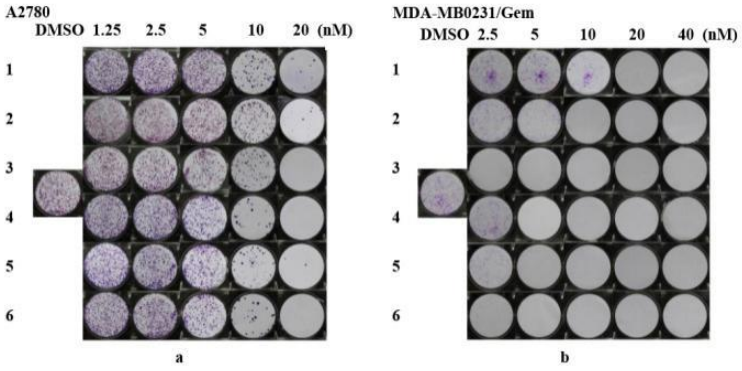


Figure S2. Treatment of ovarian cancer cell line A2780 (**a**) and drug-resistant breast cancer cell line MDA-MB-231/Gem (**b**) with the compound **2**-**6** resulted in a dose-dependent inhibition of colony formation. One colony was defined to be an aggregate of >50 cells.

IV. Method for cell apoptosis analysis.

Cell apoptosis was detected by flow cytometry according to a previously published method[3]. Briefly, cells were incubated with DMSO or different concentrations of testing compounds for 24 h. Then cells were harvested, washed twice with cold 1×PBS, and resuspended in 200 μL binding buffer at density of 1×10^5 cells /mL. The cells were then stained with 5 μL Annexin-V and 10 μL PI, for 15 min in dark condition at room temperature and subjected to analysis by flow cytometry (BD FACSAria II). The early apoptosis was evaluated based on the percentage of cells with Annexin V+/PI-, while the late apoptosis was that of Annexin V+/PI+. The results were indicated as mean values from two independent determinations.

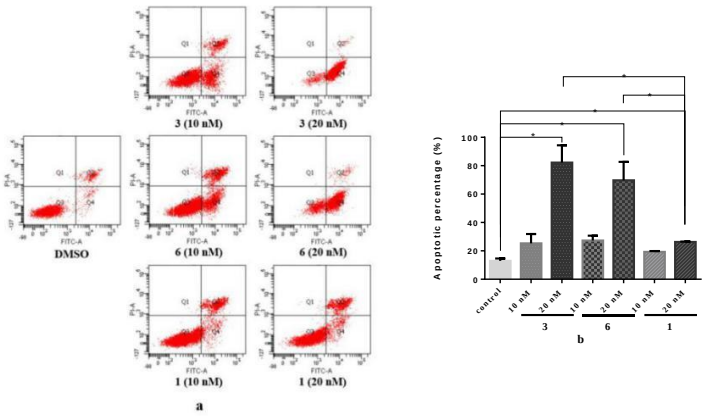


Figure S3. Apoptosis induced by compound **3** and **6** in A2780 cells. (**a**) Apoptotic cells were detected with annexin-V/PI double staining by flow cytometry after incubation with compound **3**, **6** and **1** at the concentration of 10 and 20 nM or diluent (DMSO) for 24 h. (**b**) The apoptotic percentage (%) of A2780 cells. Results represent the mean ± SEM from two independent experiments: (*) $p < 0.05$.

Compounds **3** and **6** were chosen to determine the number and stage of apoptotic cells using annexin-V/PI double staining assay and quantitating by flow cytometry. Lead compound **1** was used as a reference. As **Figure S3b** described, compounds **3** and **6** remarkably induced cell apoptosis. Unexpectedly, compounds **3** and **6** seemed to mainly induce early apoptosis, while lead compound **1** mainly induced late apoptosis (**Figure S3a**). Next, we carried out the DNA-based cell cycle analysis. A2780 cells were detected with PI staining by flow cytometry after incubation with compounds **3**, **6** and **1** at the concentration of 20, 30, 40 nM and diluent (DMSO) for 18 h (data not shown). Comparing to lead compound **1**, compound **6** only displayed relatively low G2/M arrest at concentration of 40 nM, and compound **3** hardly affect cell cycle at the tested concentrations mentioned above. Those results suggested that the newly synthesized compounds, especially 3-fluorobenzyl coumarin/furoxan hybrid **3**, might have different pharmacologic actions for anti-cancer compared to lead compound **1**.

V. Method for western-blot analysis.

Cells were harvested, washed with cold 1×PBS, and lysed with RIPA lysis buffer (Beyotime, China) for 30 min on ice, then centrifuged at 12,000 g for 15 min at 4 ºC. The total protein concentration was determined by BCA Protein Assay Kit (Beyotime, China). Equal amounts (30 μg per load) of protein samples were subjected to SDS-PAGE electrophoresis and transferred on to polyvinylidene fluoride (PVDF) membranes (Millipore) which was then blocked in 10% non-fat milk (BD Biosciences), and reacted with primary antibodies. The antibodies MDR1/ABCB1 (E1Y7B) Rabbit mAb and β-actin were purchased from Cell Signaling Technology. The secondary antibodies conjugated with horseradish

peroxidase (HRP) were also purchased from Cell Signaling Technology. The protein bands were developed by the chemiluminescent reagents (Millipore).

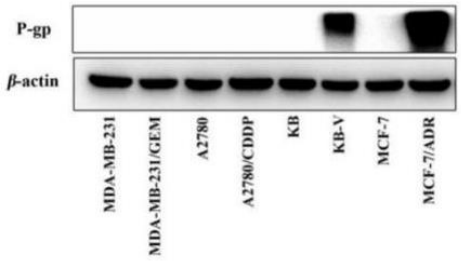


Figure S4. P-gp expression in four couples of drug-sensitive and their drug-resistant cancer cell lines.

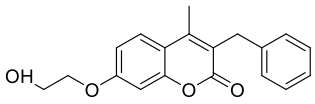
As expected (**Figure S4**), the expression of P-gp was outstanding in two drug-resistant cell lines KB-V and MCF-7/ADR, to which our newly synthesized compounds showed significant selective antiproliferation activities compared with their sensitive cell lines KB and MCF-7. While P-gp overexpression wasn't observed in two couples of the drug-resistant cell lines A2780/CDDP and MDA-MB-231/GEM *vs* their drug-sensitive ones A2780 and MDA-MB-231, which having no relevant selective antiproliferation activities for our compounds. Based on this result, we will further study the possible pharmacologic mechanism of these new compounds and the correlationship between P-gp overexpression and the remarkable anti-cancer activities in multi-drug resistant tumor cell lines.

VI. High resolution mass spectral data for target compounds

Table S1. High Resolution Mass Spectra for Target Compounds.

compounds	High resolution mass spectra				
	Chemical formula	Calculated	Calculated + Na	Measured	ppm error
2	C27H22N2O8S	534.1097	557.0989	557.0984	-0.9
3	C27H21FN2O8S	552.1003	575.0895	575.0905	1.8
4	C28H26N4O10S2	642.1090	665.0983	665.0979	-0.5
5	C26H22N2O8S	522.1097	545.0989	545.0988	-0.2
6	C26H21FN2O8S	540.1003	563.0895	563.0867	-5.0

VII. ^{1}H NMR, ^{13}C NMR and HSQC spectra



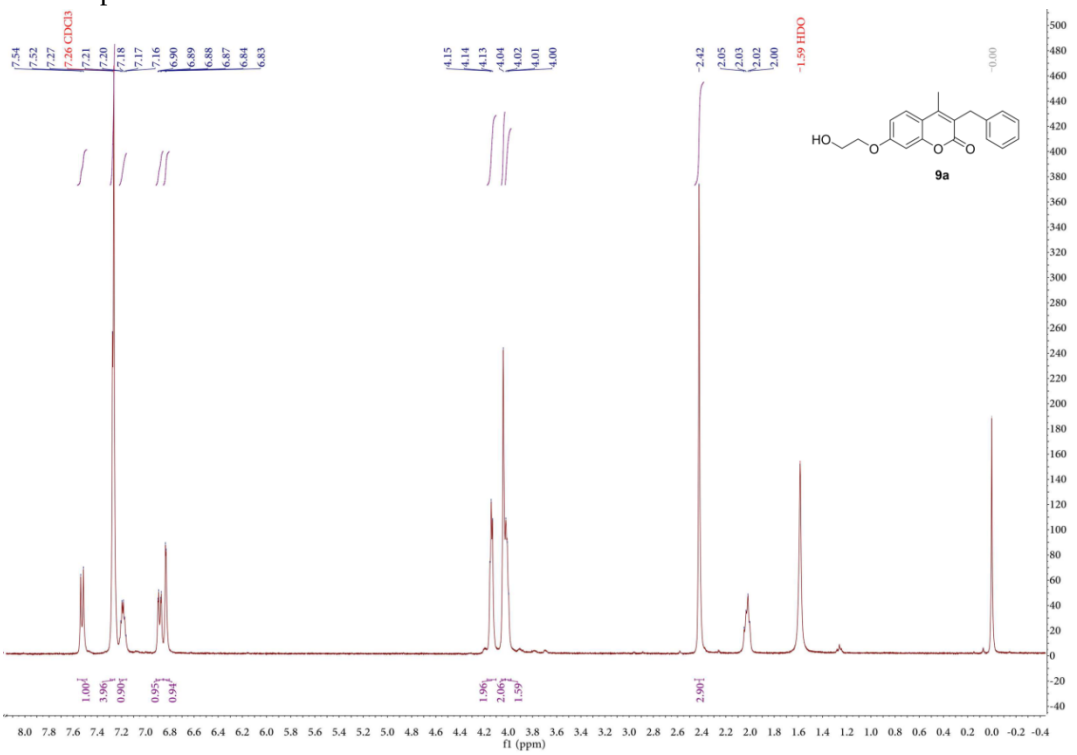
9a

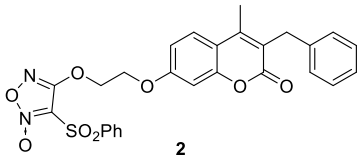
$C_{19}H_{18}O_4$

[310.1]

3-benzyl-7-(2-hydroxyethoxy)-4-methyl-2H-chromen-2-one (**9a**). m.p. 88.0-89.5 °C, MS(ESI) (m/z) 311.0 $[M+H]^+$. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.53 (d, *J* = 8.8 Hz, 1H, 5-H), 7.29 – 7.26 (m, 4H, Bn-H), 7.22 – 7.16 (m, 1H, Bn-H), 6.88 (dd, *J* = 8.8, 2.1 Hz, 1H, 6-H), 6.83 (d, *J* = 2.1 Hz, 1H, 8-H), 4.14 (t, *J* = 4.4 Hz, 2H, O$\underline{CH_2}CH_2OH$), 4.04 (s, 2H, 3'-CH_2), 4.01 (t, *J* = 4.4 Hz, 2H, $OCH_2\underline{CH_2}OH$), 2.42 (s, 3H, CH_3).

^{1}H NMR spectra:





2

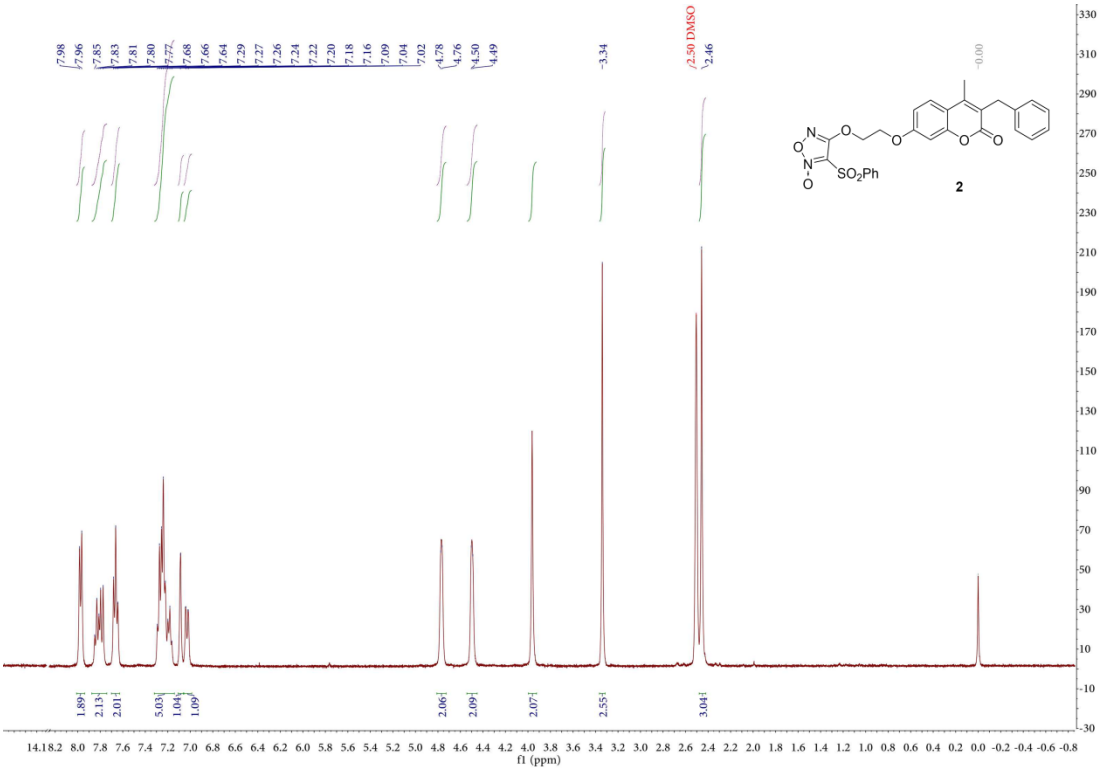
$C_{27}H_{22}N_2O_8S$

[534.1]

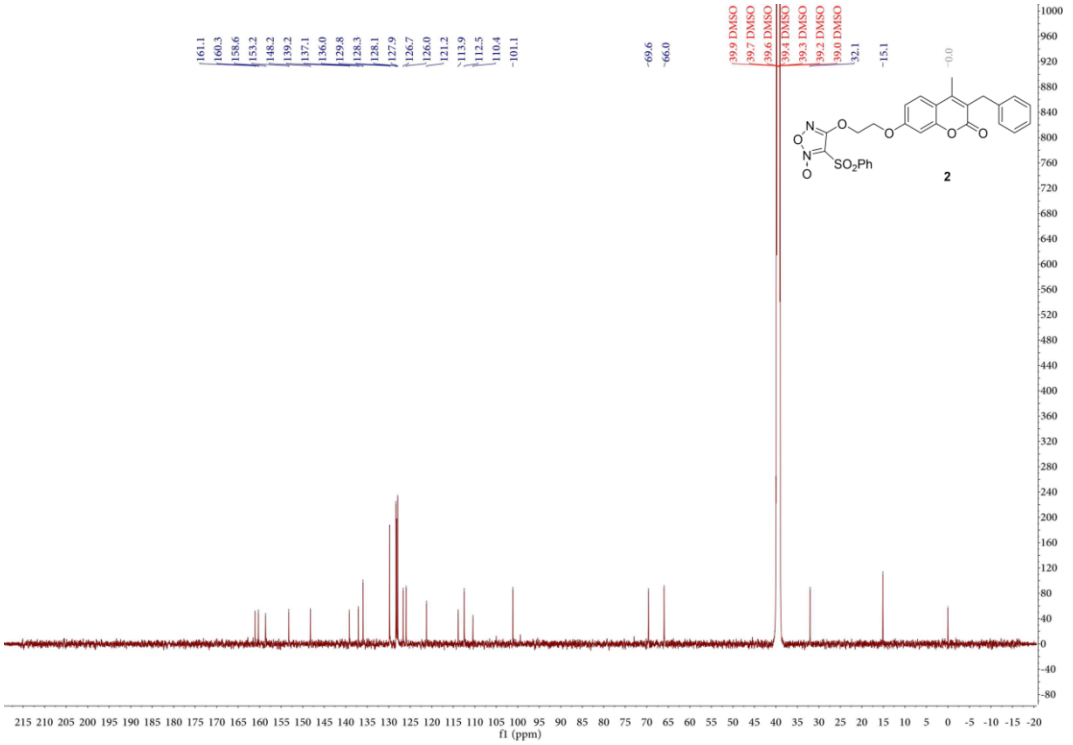
4-(2-((3-benzyl-4-methyl-2-oxo-2H-chromen-7-yl)oxy)ethoxy)-3-(phenylsulfonyl)-1,2,5-oxadiazole 2-oxide (**2**). m.p. 205.2-206.5 °C, MS(ESI) (m/z) 557.2 $[M+Na]^+$. ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.97 (d, *J* = 7.8 Hz, 2H, (2',6'-SO_2Ph-H)), 7.87 – 7.74 (m, 2H, 4'-SO_2Ph-H, 5-H), 7.66 (t, *J* = 7.8 Hz, 2H, (3',5'-SO_2Ph-H)), 7.29-7.16 (m, 5H, Bn-H), 7.09 (s, 1H, 8-H), 7.03 (d, *J* = 8.8 Hz, 1H, 6-H), 4.77 (d, *J* = 5.4 Hz, 2H, Furazanyl-O$\underline{CH_2}CH_2O$), 4.50 (d, *J* = 5.4 Hz, 2H, Furazanyl-$OCH_2\underline{CH_2}O$), 3.34 (s, 2H, 3-CH_2), 2.46 (s, 3H, 4-CH_3). ^{13}C NMR (150 MHz,

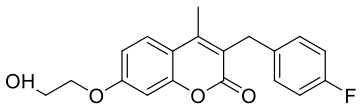
DMSO-d_6) δ 161.1, 160.3, 158.7, 153.2, 148.2, 139.2, 137.1, 136.0, 129.8, 128.3, 128.1, 127.9, 126.7, 126.0, 121.2, 113.9, 112.5, 110.4, 101.1, 69.6, 66.0, 32.1, 15.1.

^{1}H NMR spectra:



^{13}C NMR spectra:



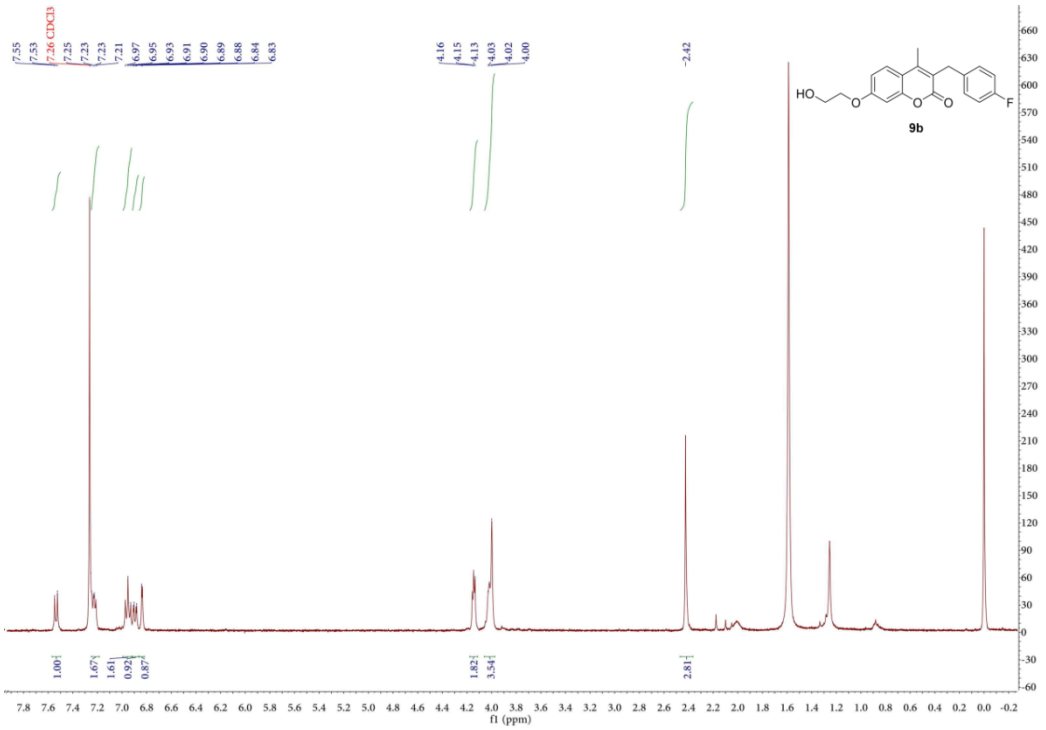


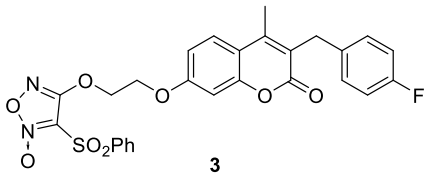
9b

$C_{19}H_{17}FO_4$
[328.1]

3-(4-fluorobenzyl)-7-(2-hydroxyethoxy)-4-methyl-2H-chromen-2-one (**9b**). m.p. 126.0-128.1 °C, MS(ESI) (m/z) 329.0 $[M+H]^+$. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.54 (d, *J* = 8.9 Hz, 1H, 5-H), 7.25-7.18 (m, 2H, (2',6'-Bn-H)), 6.95 (t, *J* = 8.6 Hz, 2H, (3',5'-Bn-H)), 6.90 (dd, *J* = 8.9, 2.5 Hz, 1H, 6-H), 6.84 (d, *J* = 2.5 Hz, 1H, 8-H), 4.18 – 4.11 (m, 2H, OCH_2CH_2OH), 4.06 – 3.97 (m, 4H, OCH_2CH_2OH, 3-CH_2), 2.42 (s, 3H, 4-CH_3).

^{1}H NMR spectra:





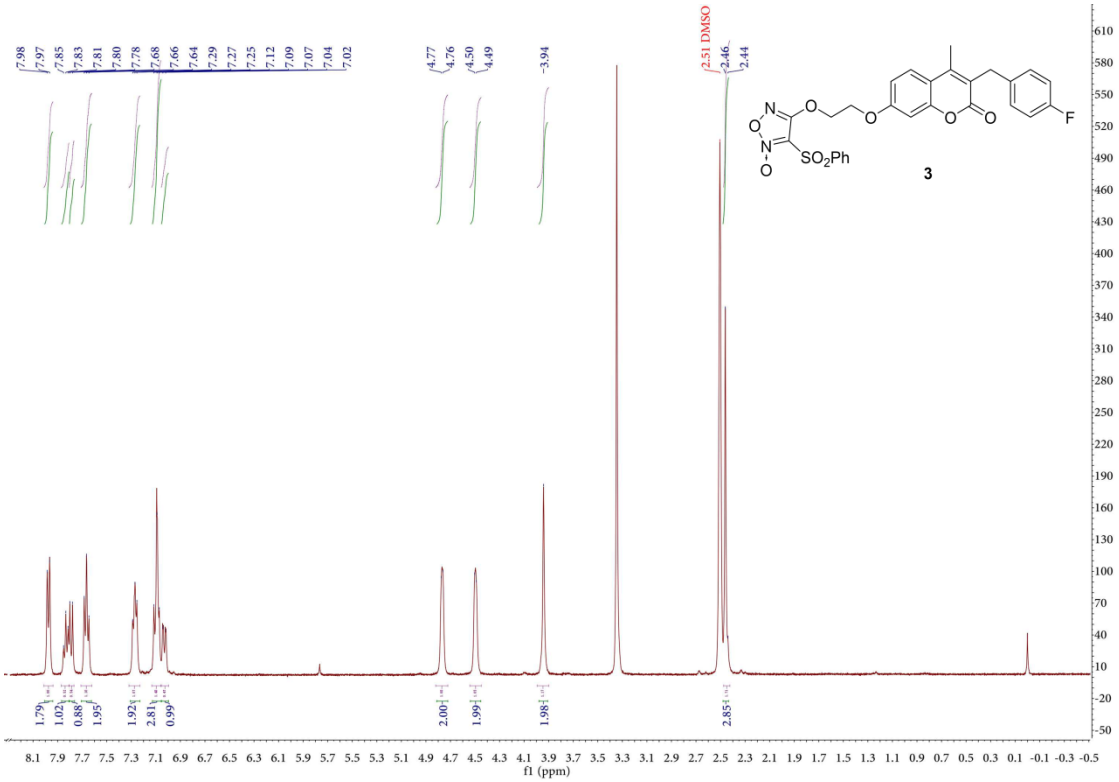
3

$C_{27}H_{21}FN_2O_8S$
[552.1]

4-(2-((3-(4-fluorobenzyl)-4-methyl-2-oxo-2H-chromen-7-yl)oxy)ethoxy)-3-(phenylsulfonyl)-1,2,5-oxadiazole 2-oxide (**3**). m.p. 216.4-218.5 °C, MS(ESI) (m/z) 553.2 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.97 (d, *J* = 7.8 Hz, 2H, (2',6'-SO_2Ph-H)), 7.83 (t, *J* = 7.4 Hz, 1H, 4'-SO_2Ph-H), 7.79 (d, *J* = 8.8 Hz, 1H, 5-H), 7.66 (t, *J* = 7.8 Hz, 2H, (3',5'-SO_2Ph-H)), 7.27 (t, *J* = 6.8 Hz, 2H, (2",6"-Bn-H)), 7.13 – 7.06 (m, 3H, (3",5"-Bn-H), 8-H), 7.03 (d, *J* = 8.8 Hz,

1H, 6-H), 4.77 (d, *J* = 5.5 Hz, 2H, Furazanyl-O$\underline{CH_2}CH_2O$), 4.50 (d, *J* = 5.5 Hz, 2H, Furazanyl-$OCH_2\underline{CH_2}O$), 3.94 (s, 2H, 3-CH_2), 2.46 (s, 3H, 4-CH_3). ^{13}C NMR (150 MHz, DMSO-d_6) δ 161.1, 160.3, 158.7, 153.2, 148.3, 137.1, 136.0, 135.3, 129.8, 129.8, 129.7, 128.2, 126.7, 121.2, 115.1, 114.9, 113.9, 112.5, 110.4, 101.2, 69.6, 66.0, 31.3, 15.1.

^{1}H NMR spectra:



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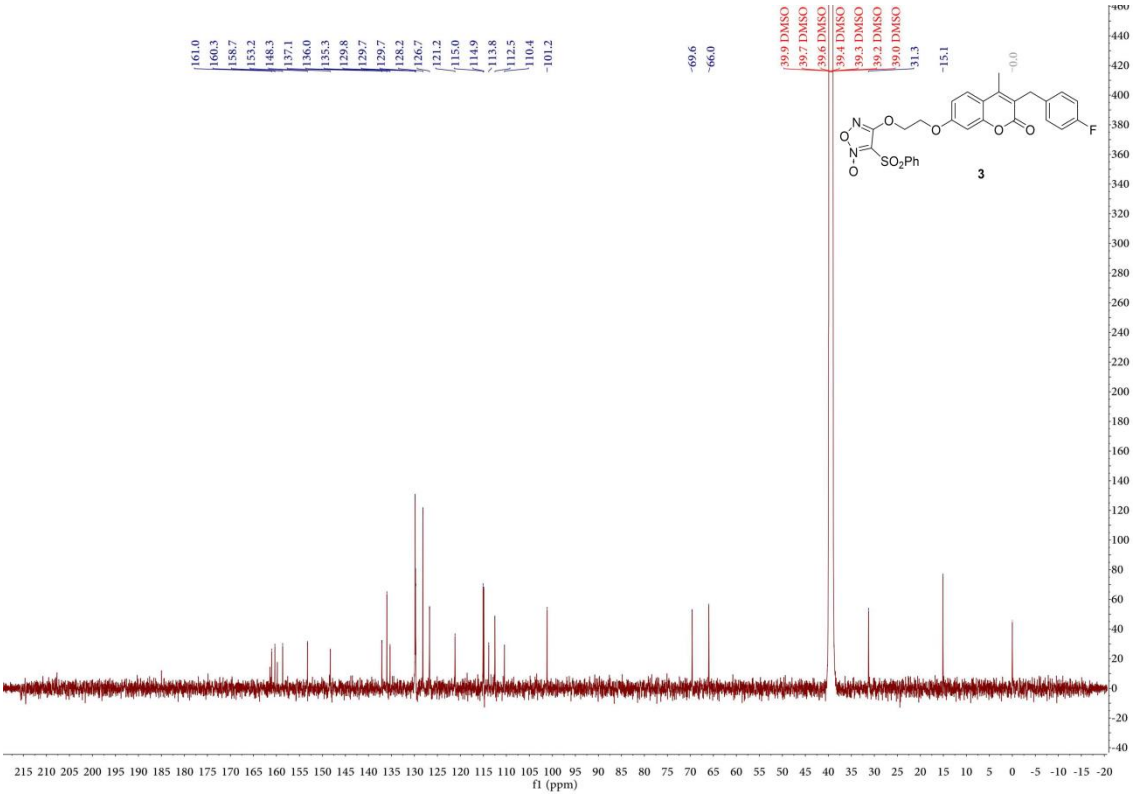
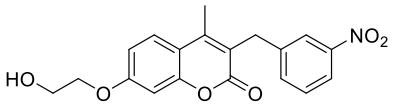


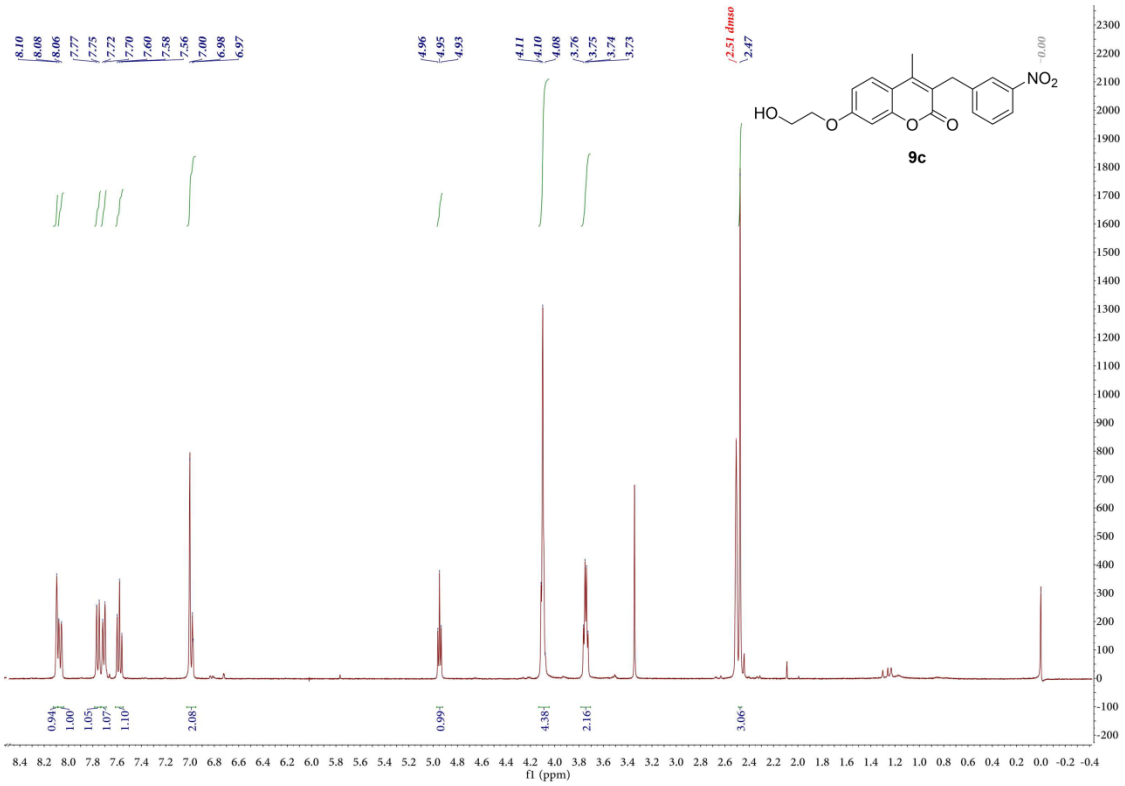


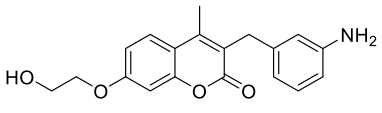
9c

$C_{19}H_{17}NO_6$
[355.1]

7-(2-hydroxyethoxy)-4-methyl-3-(3-nitrobenzyl)-2H-chromen-2-one (**9c**). m.p. 144.5-146.9 °C, MS(ESI) (m/z) 356.0 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.10 (s, 1H, 2'-Bn-H), 8.07 (d, J = 8.4 Hz, 1H, 4'-Bn-H), 7.76 (d, J = 8.6 Hz, 1H, 5'-Bn-H), 7.71 (d, J = 7.8 Hz, 1H, 6'-Bn-H), 7.58 (t, J = 7.8 Hz, 1H, 5-H), 7.03 – 6.95 (m, 2H, 6-H,8-H), 4.95 (t, J = 5.5 Hz, 1H, OH), 4.13 – 4.05 (m, 4H, O$\underline{CH_2}CH_2$OH, 3-CH_2), 3.74 (q, J = 5.1 Hz, 2H, OCH$_2\underline{CH_2}$OH), 2.47 (s, 3H, 4-CH_3).

^{1}H NMR spectra:



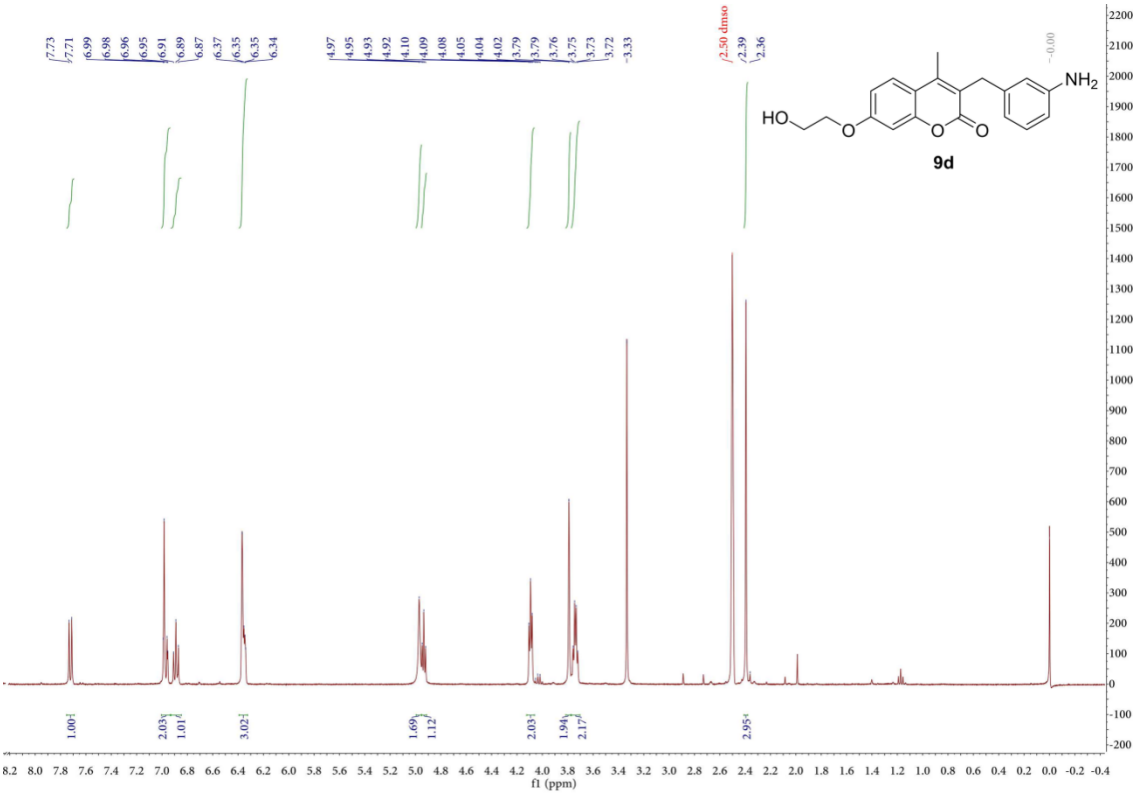


9d

$C_{19}H_{19}NO_4$
[325.1]

3-(3-aminobenzyl)-7-(2-hydroxyethoxy)-4-methyl-2H-chromen-2-one (**9d**). m.p. 158.1-160.0 °C, MS(ESI) (m/z) 326.2 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.72 (d, J = 8.6 Hz, 1H, 5-H), 7.00 – 6.93 (m, 2H, 6-H,8-H), 6.89 (t, J = 7.8 Hz, 1H, 2'-Bn-H), 6.39 – 6.32 (m, 3H, (4',5',6'-Bn-H)), 4.97 (s, 2H, NH_2), 4.93 (t, J = 5.5 Hz, 1H, OH), 4.09 (t, J = 4.8 Hz, 2H, O$\underline{CH_2}CH_2OH$) , 3.79 (s, 2H 3-CH_2), 3.74 (q, J = 4.8 Hz, 2H, $OCH_2\underline{CH_2}OH$), 2.39 (s, 3H, 4-CH_3).

^{1}H NMR spectra:

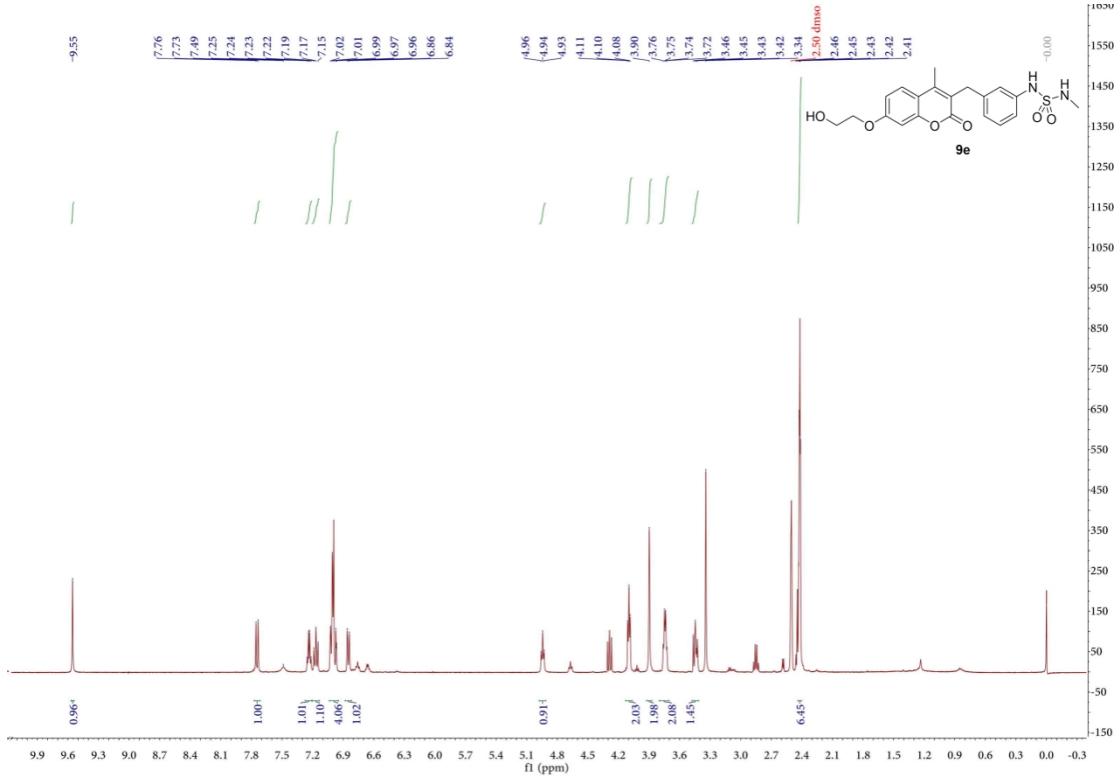


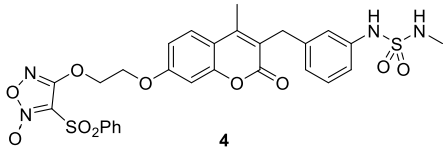
9e

$C_{20}H_{22}N_2O_6S$
[418.1]

3-(3-(methylsulfamic)aminobenzyl)-7-(2-hydroxyethoxy)-4-methyl-2H-chromen-2-one (**9e**). MS(ESI) (m/z) 419.0 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.55 (s, 1H, $\underline{NH}SO_2NHCH_3$), 7.75 (d, J = 8.5 Hz, 1H, 5-H), 7.23 (q, J = 5.0 Hz, 1H, $\underline{NH}CH_3$), 7.17 (t, J = 8.1 Hz, 1H, 2'-Bn-H), 7.04 – 6.95 (m, 4H, 6-H, 8-H, (4',5'-Bn-H)), 6.85 (d, J = 7.6 Hz, 1H, 6'-Bn-H), 4.94 (t, J = 5.5 Hz, 1H, OH), 4.10 (t, J = 4.8 Hz, 2H, $O\underline{CH_2}CH_2OH$), 3.90 (s, 2H, 3-CH_2), 3.74 (q, J = 4.8 Hz, 2H, $OCH_2\underline{CH_2}OH$), 2.44 – 2.40 (m, 6H, 4-CH_3, $NH\underline{CH_3}$).

^{1}H NMR spectra:



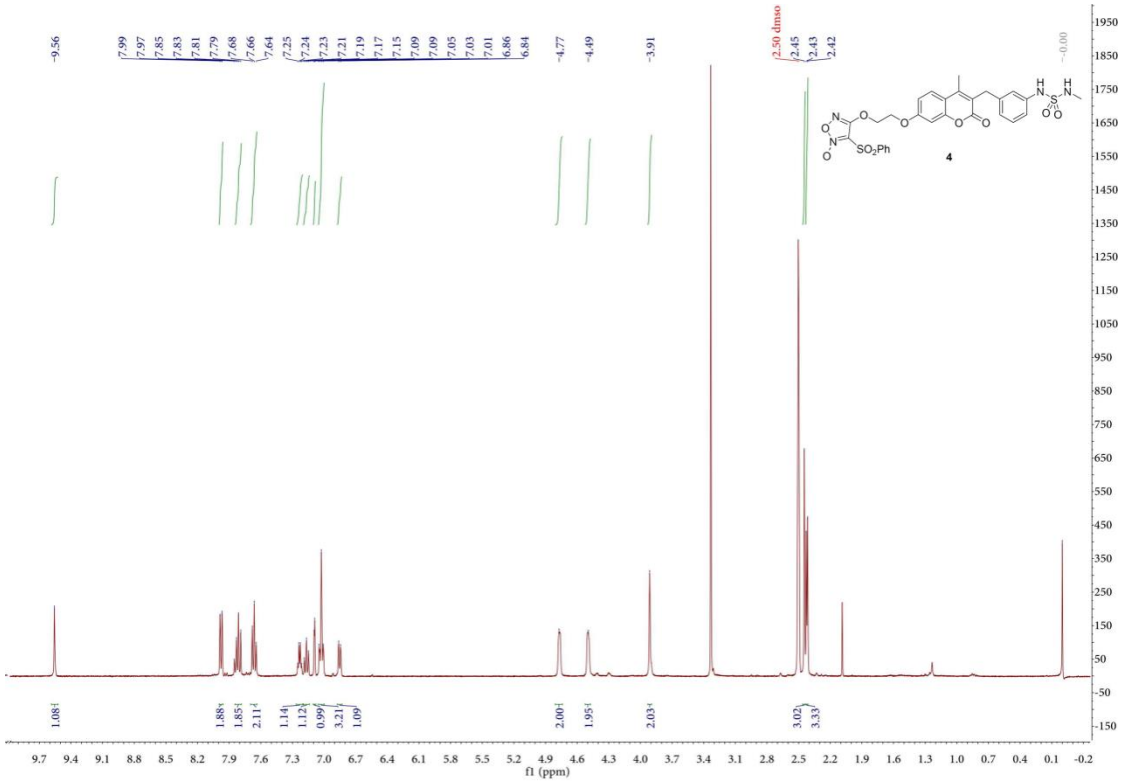


4

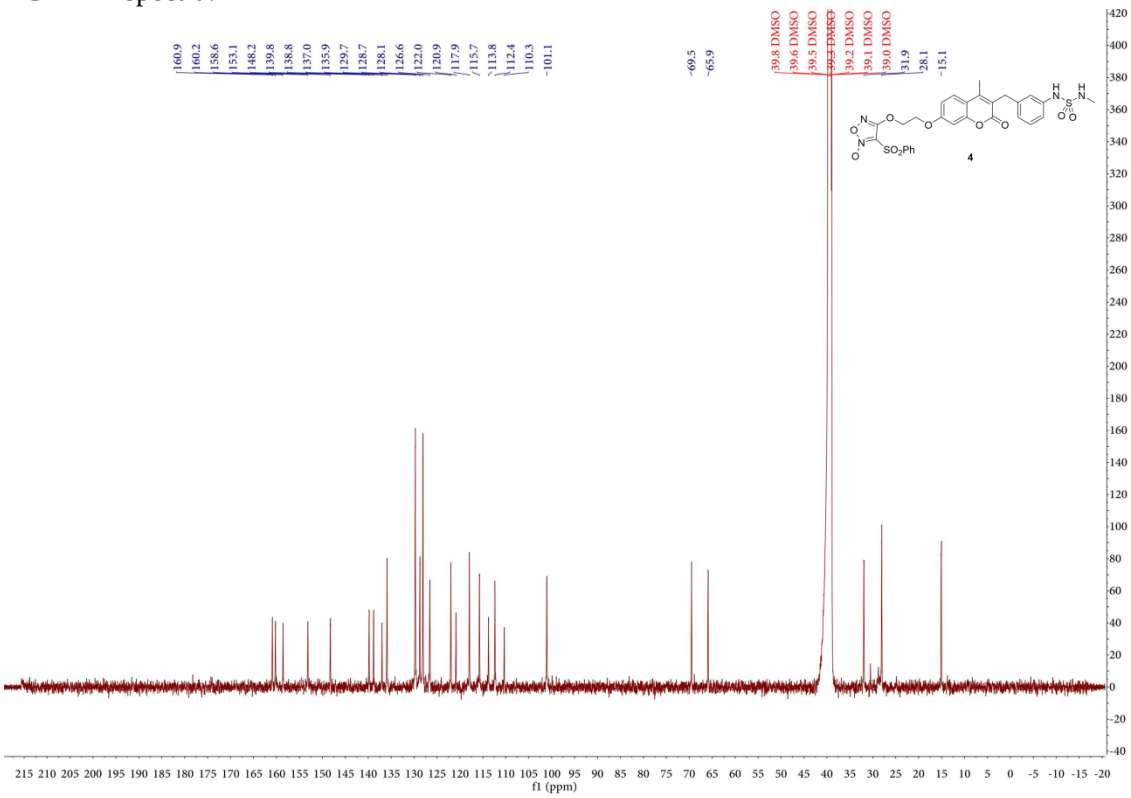
$C_{28}H_{26}N_4O_{10}S_2$
[642.1]

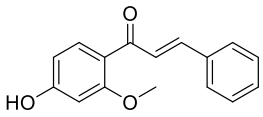
4-(2-((3-(3-(methylsulfamic)aminobenzyl)-4-methyl-2-oxo-2H-chromen-7-yl)oxy)ethoxy)-3-(phenylsulfonyl)-1,2,5-oxadiazole 2-oxide (**4**). m.p. 175.0-176.5 °C, MS(ESI) (m/z) 643.2 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.56 (s, 1H, $\underline{NH}SO_2NHCH_3$), 7.98 (d, J = 7.9 Hz, 2H, (2',6'-SO_2Ph-H)), 7.88 – 7.77 (m, 2H, 4'-SO_2Ph-H, 5-H), 7.66 (t, J = 7.7 Hz, 2H, (3',5'-SO_2Ph-H)), 7.23 (q, J = 5.0 Hz, 1H, $\underline{NH}CH_3$), 7.17 (t, J = 7.8 Hz, 1H, 2"-Bn-H), 7.09 (d, J = 2.5 Hz, 1H, 8-H), 7.07 - 6.98 (m, 3H, 6-H, (4",5"-Bn-H)), 6.85 (d, J = 7.6 Hz, 1H, 6"-Bn-H), 4.77 (brs, 2H, Furazanyl-O$\underline{CH_2}CH_2O$), 4.49 (brs, 2H, Furazanyl-O$CH_2\underline{CH_2}O$), 3.91 (s, 2H, 3-CH_2), 2.45 (s, 3H, 4-CH_3), 2.42 (d, J = 4.9 Hz, 3H, NH$\underline{CH_3}$). ^{13}C NMR (150 MHz, DMSO-d_6) δ 160.9, 160.2, 158.6, 153.2, 148.3, 139.8, 138.8, 137.0, 135.9, 129.7, 128.7, 128.1, 126.6, 122.0, 120.9, 117.9, 115.8, 113.8, 112.4, 110.3, 101.1, 69.5, 65.9, 28.1, 15.1.

^{1}H NMR spectra:



^{13}C NMR spectra:



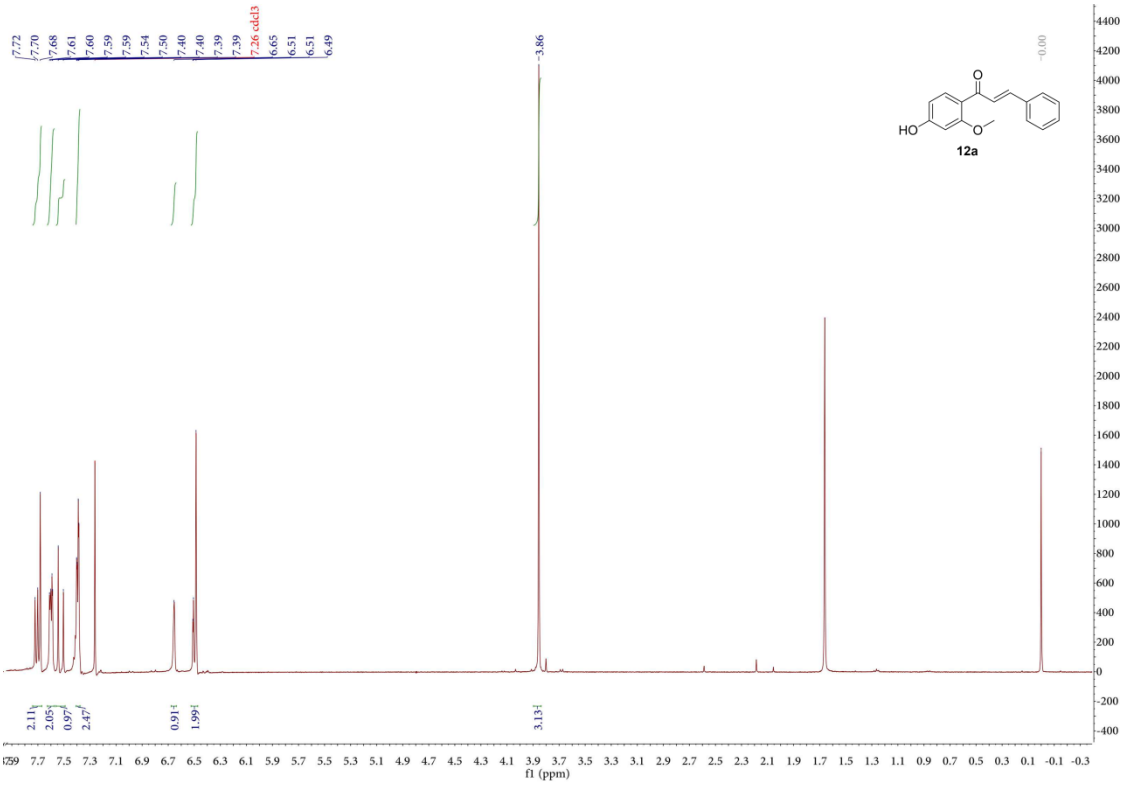


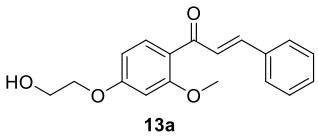
12a

$C_{16}H_{14}O_3$
[254.1]

(*E*)-1-(4-hydroxy-2-methoxyphenyl)-3-phenylprop-2-en-1-one (**12a**). m.p. 144.2-145.8 °C, MS(ESI) (m/z) 255.1 $[M+H]^+$. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.74 – 7.67 (m, 2H, 6-Ar-H, 3-H), 7.63 – 7.58 (m, 2H, (2',6'-Ph-H)), 7.52 (d, *J* = 15.8 Hz, 1H, 2-H), 7.42 – 7.38 (m, 2H, (3',5'-Ph-H)), 6.65 (s, 1H, 4'-Ph-H), 6.54 -6.46 (m, 2H, (3,5-Ar-H)), 3.86 (s, 3H, OCH_3).

^{1}H NMR spectra:





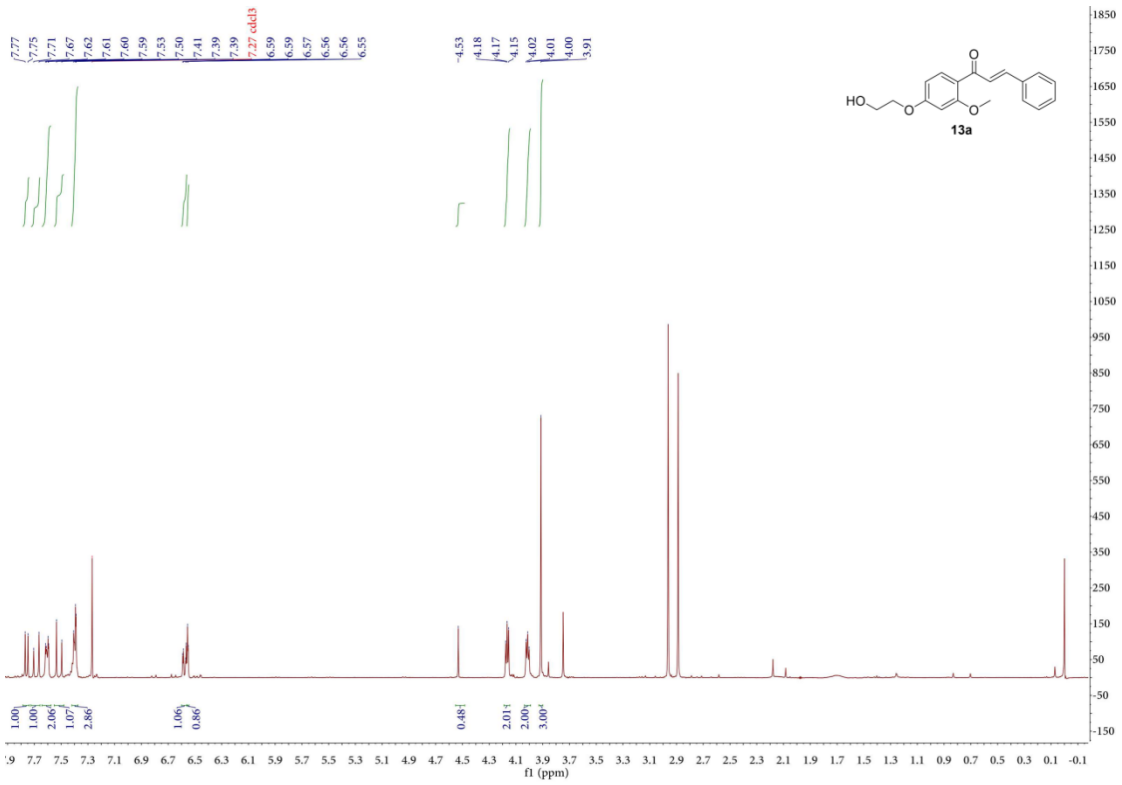
13a

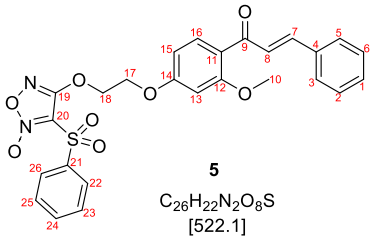
$C_{18}H_{18}O_4$
[298.1]

(*E*)-1-(4-(2-hydroxyethoxy)-2-methoxyphenyl)-3-phenylprop-2-en-1-one (**13a**). MS(ESI) (m/z) 299.1 $[M+H]^+$. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.76 (d, *J* = 8.5 Hz, 1H, 6-Ar-H), 7.69 (d, *J* = 15.8 Hz, 1H, 3-H), 7.60 (dd, *J* = 7.2, 2.2 Hz, 2H, (2',6'-Ph-H)), 7.51 (d, *J* = 15.8 Hz, 1H, 2-H), 7.42 – 7.37 (m, 3H, (3',4',5'-Ph-H)), 6.58 (dd, *J* = 8.5, 2.2 Hz, 1H, 5-Ar-H), 6.55 (d, *J* = 2.1 Hz, 1H, 3-Ar-H), 4.53 (s, 1H, OH), 4.19 – 4.15 (m, 2H, Ar-O$\underline{CH_2}CH_2OH$),

4.04 – 3.99 (m, 2H, Ar-$OCH_2\underline{CH_2}OH$), 3.91 (s, 3H, OCH_3).

^{1}H NMR spectra:





5

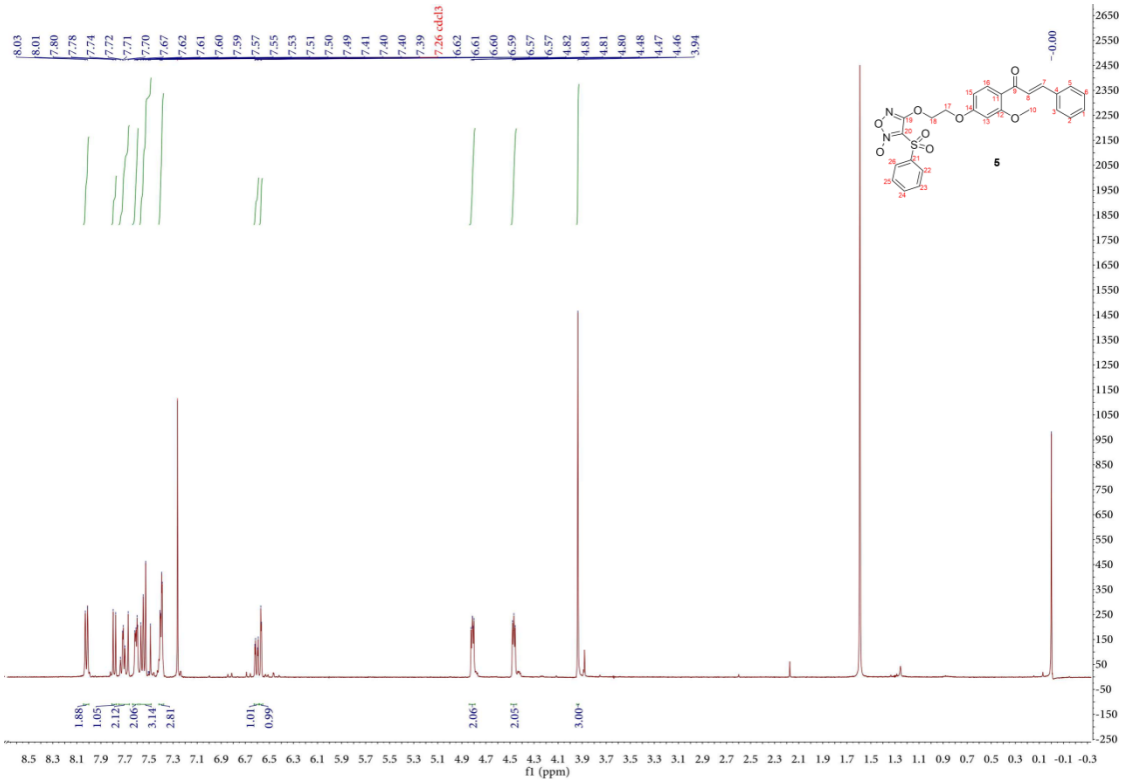
$C_{26}H_{22}N_2O_8S$

[522.1]

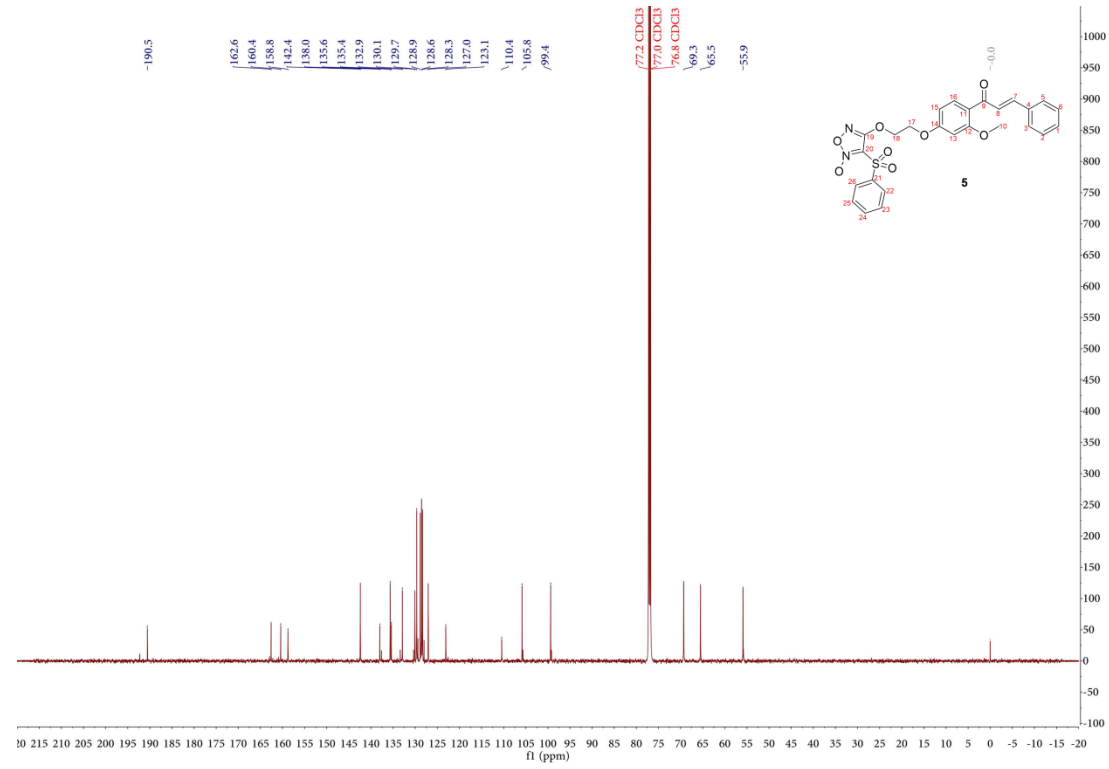
Note: The numbers around each carbon in compound **5** are listed manually just for easier clarification for each hydrogen's chemical shifts. It may be inconsistent with the IUPAC nomenclature of chemical compounds.

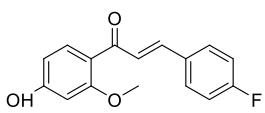
4-(2-(4-cinnamoyl-3-methoxyphenoxy)ethoxy)-3-(phenylsulfonyl)-1,2,5-oxadiazole 2-oxide (**5**). m.p. 123.5-125.0 °C, MS(ESI) (m/z) 523.1 $[M+H]^+$. ^{1}H NMR (600 MHz, Chloroform-*d*) δ 8.02 (d, *J* = 7.9 Hz, 2H, H_{22}, H_{26}), 7.79 (d, *J* = 8.6 Hz, 1H, H_{16}), 7.75 – 7.66 (m, 2H, H_{24}, H_7), 7.61 (dd, *J* = 6.8, 2.8 Hz, 2H, H_{23}, H_{25}), 7.58 – 7.48 (m, 3H, H_3, H_5, H_8), 7.40 (dd, *J* = 5.1, 1.9 Hz, 3H, H_2, H_6, H_1), 6.61 (dd, *J* = 8.6, 2.2 Hz, 1H, H_{15}), 6.57 (d, *J* = 2.2 Hz, 1H, H_{13}), 4.81 (dd, *J* = 5.6, 3.4 Hz, 2H, H_{18A}, H_{18B}), 4.49 – 4.45 (m, 2H, H_{17A}, H_{17B}), 3.94 (s, 3H, H_{10A}, H_{10B}, H_{10C}). ^{13}C NMR (150 MHz, Chloroform-*d*) δ 190.6, 162.6, 160.4, 158.8, 142.4, 138.0, 135.6, 135.4, 132.9, 130.1, 129.7, 128.9, 128.6, 128.3, 127.1, 123.1, 110.4, 105.8, 99.4, 69.3, 65.5, 55.9.

^{1}H NMR spectra:



^{13}C NMR spectra:



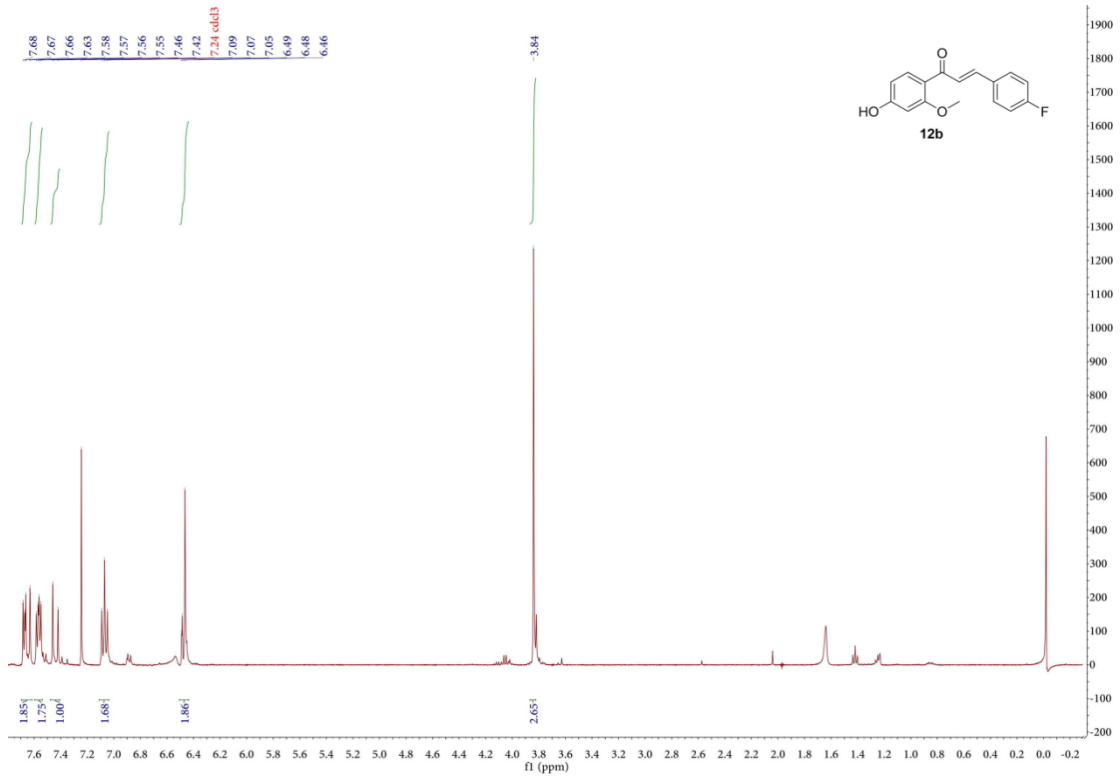


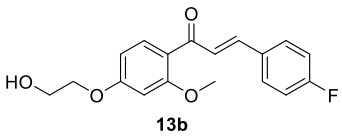
12b

$C_{16}H_{13}FO_3$
[272.1]

(*E*)-3-(4-fluorophenyl)-1-(4-hydroxy-2-methoxyphenyl)prop-2-en-1-one (**12b**). m.p. 125.8-127.4 °C, MS(ESI) (m/z) 273.0 $[M+H]^+$. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.70 – 7.62 (m, 2H, 6-Ar-H, 3-H), 7.59 – 7.54 (m, 2H, (2',6'-Ph-H)), 7.44 (d, *J* = 15.8 Hz, 1H, 2-H), 7.07 (t, *J* = 8.6 Hz, 2H, (3',5'-Ph-H)), 6.51 – 6.44 (m, 2H, (3,5-Ar-H)), 3.84 (s, 3H, OCH_3).

^{1}H NMR spectra:



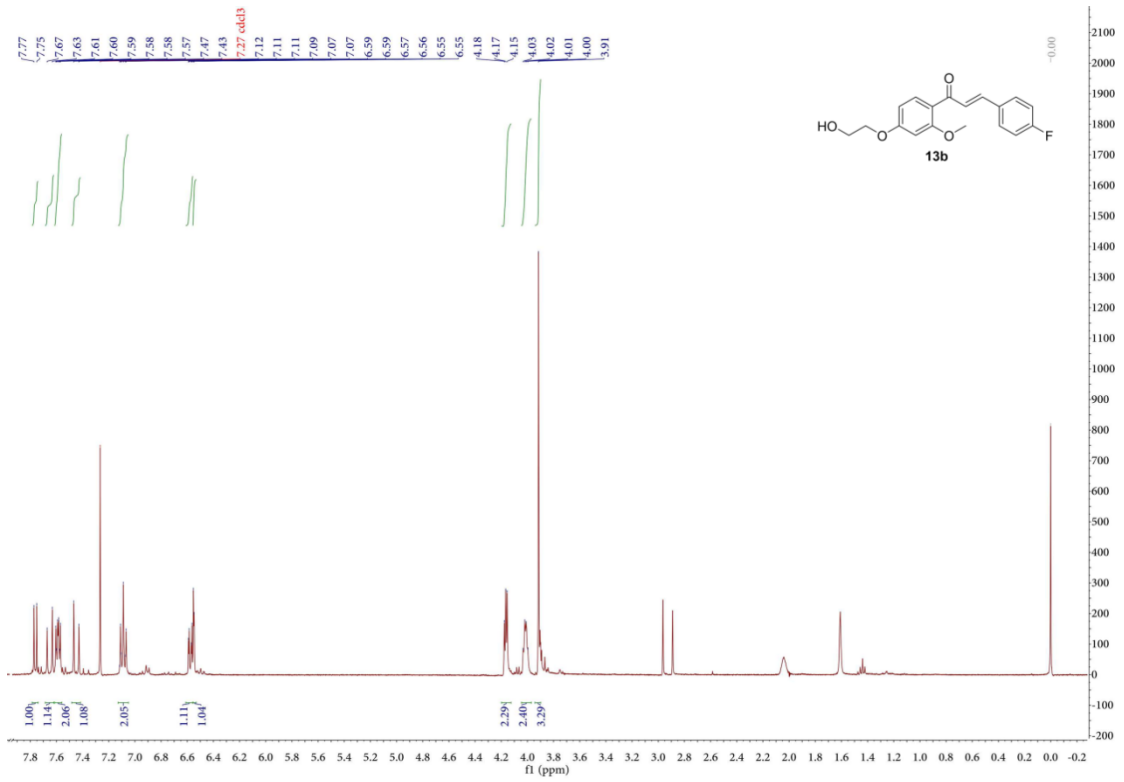


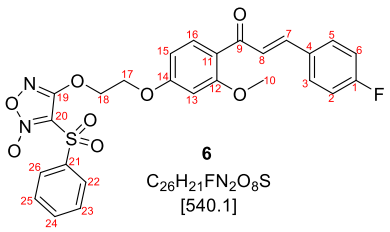
13b

$C_{18}H_{17}FO_4$
[316.1]

(*E*)-3-(4-fluorophenyl)-1-(4-(2-hydroxyethoxy)-2-methoxyphenyl)prop-2-en-1-one (**13b**). MS(ESI) (m/z) 317.1 $[M+H]^+$. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.76 (d, *J* = 8.5 Hz, 1H, 6-Ar-H), 7.65 (d, *J* = 15.8 Hz, 1H, 3-H), 7.61 – 7.56 (m, 2H, (2',6'-Ph-H)), 7.45 (d, *J* = 15.8 Hz, 1H, 2-H), 7.09 (t, *J* = 8.5 Hz, 2H, (3',5'-Ph-H)), 6.58 (dd, *J* = 8.5, 2.3 Hz, 1H, 5-Ar-H), 6.55 (d, *J* = 2.2 Hz, 1H, 3-Ar-H), 4.20 – 4.13 (m, 2H, Ar-O$\underline{CH_2}CH_2OH$), 4.01 (q, *J* = 5.0 Hz, 2H, Ar-$OCH_2\underline{CH_2}OH$), 3.91 (s, 3H, OCH_3).

^{1}H NMR spectra:

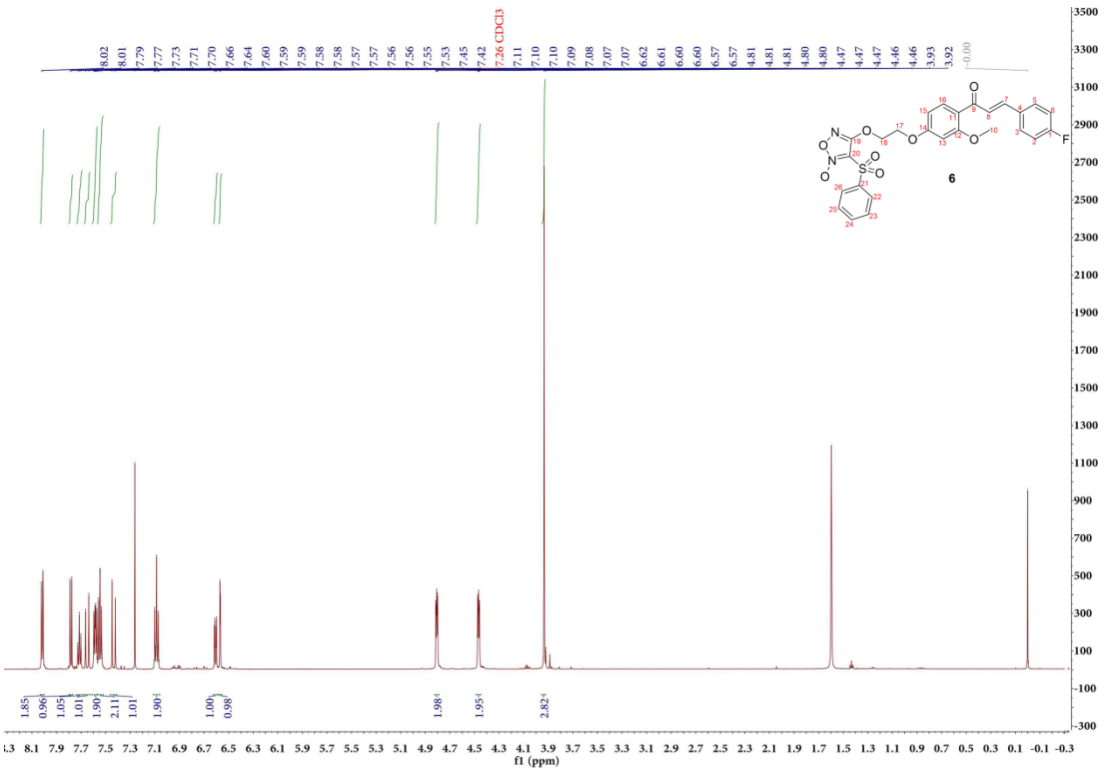




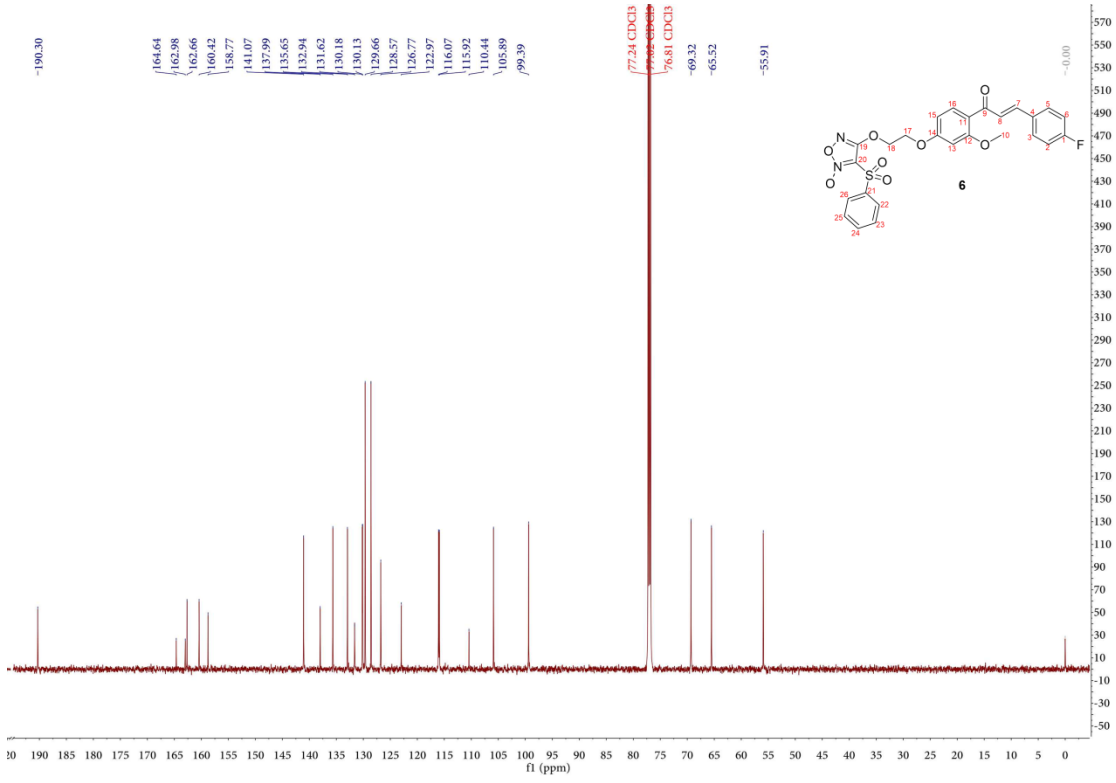
Note: The numbers around each carbon in compound **6** are listed manually just for easier clarification for each hydrogen's chemical shifts in ^{1}H NMR and each carbon's chemical shifts in ^{13}C NMR. It may be inconsistent with the IUPAC nomenclature of chemical compounds.

(*E*)-4-(2-(4-(3-(4-fluorophenyl)acryloyl)-3-methoxyphenoxy)ethoxy)-3-(phenylsulfonyl)-1,2,5-oxadiazole 2-oxide (**6**). m.p. 128.3-130.2 °C, MS(ESI) (m/z) 541.2 $[M+H]^{+}$. ^{1}H NMR (600 MHz, Chloroform-*d*) δ 8.02 (d, *J* = 7.8 Hz, 2H, H_{22}, H_{26}), 7.78 (d, *J* = 8.6 Hz, 1H, H_{16}), 7.71 (t, *J* = 7.5 Hz, 1H, H_{24}), 7.65 (d, *J* = 15.8 Hz, 1H, H_{7}), 7.61 – 7.57 (m, 2H, H_{23}, H_{25}), 7.57 – 7.52 (m, 2H, H_{3}, H_{5}), 7.43 (d, *J* = 15.8 Hz, 1H, H_{8}), 7.09 (t, *J* = 8.6 Hz, 2H, H_{2}, H_{6}), 6.61 (dd, *J* = 8.6, 2.3 Hz, 1H, H_{15}), 6.57 (d, *J* = 2.3 Hz, 1H, H_{13}), 4.82 – 4.79 (m, 2H, H_{18A}, H_{18B}), 4.49 – 4.45 (m, 2H, H_{17A}, H_{17B}), 3.93 (s, 3H, H_{10A}, H_{10B}, H_{10C}). ^{13}C NMR (150 MHz, Chloroform-*d*) δ 190.3 (C_{9}), 164.6 (C_{1}), 163.0 (C_{1}), 162.7 (C_{14}), 160.4 (C_{12}), 158.8 (C_{19}), 141.1 (C_{8}), 138.0 (C_{4}), 135.7 (C_{24}), 132.9 (C_{16}), 131.7 (C_{21}), 130.2, 130.1 (C_{3}, C_{5}), 129.7 (C_{23}, C_{25}), 128.6 (C_{22}, C_{26}), 126.8 (C_{7}), 123.0 (C_{11}), 116.1, 115.92 (C_{2}, C_{6}), 110.4 (C_{20}), 105.9 (C_{15}), 99.4 (C_{13}), 69.3 (C_{17}), 65.5 (C_{18}), 55.9 (C_{10}).

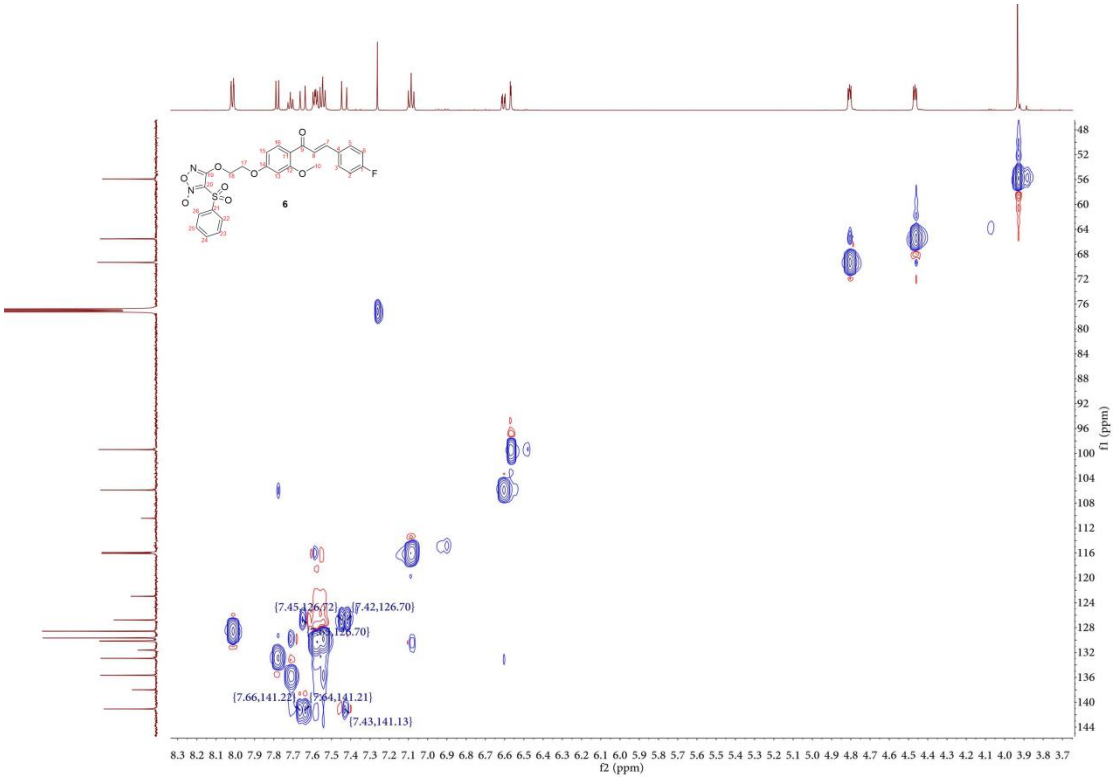
^{1}H NMR spectra:

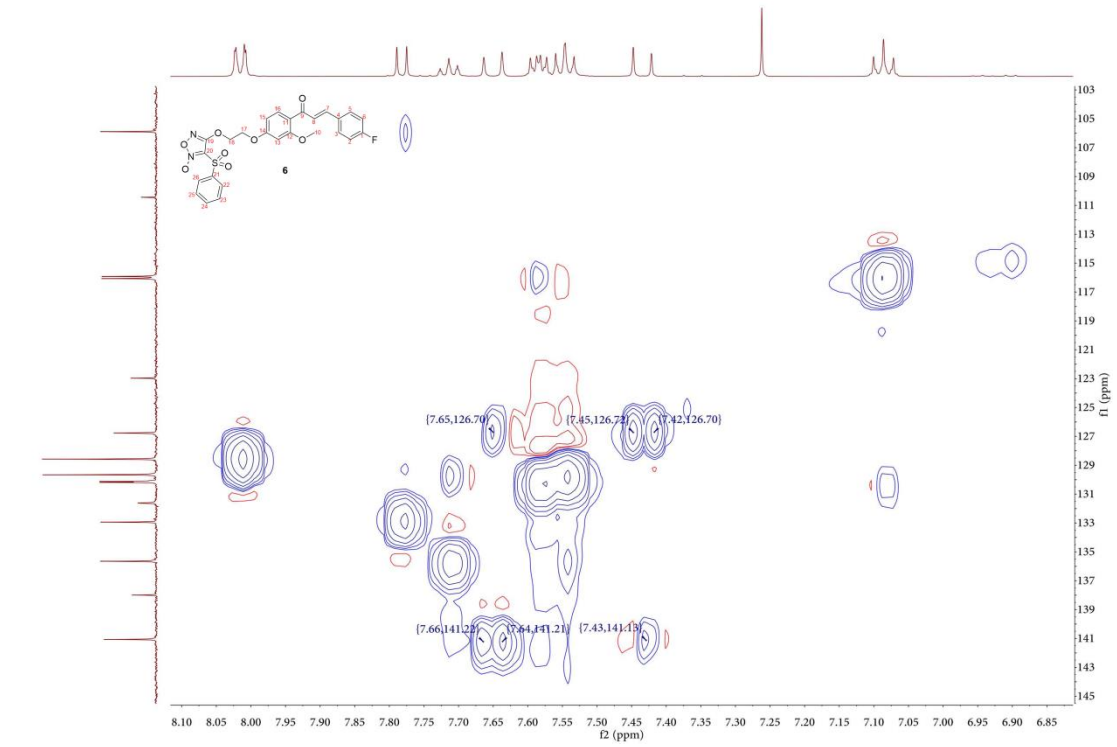


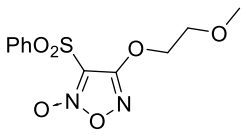
^{13}C NMR spectra:



HSQC spectra:





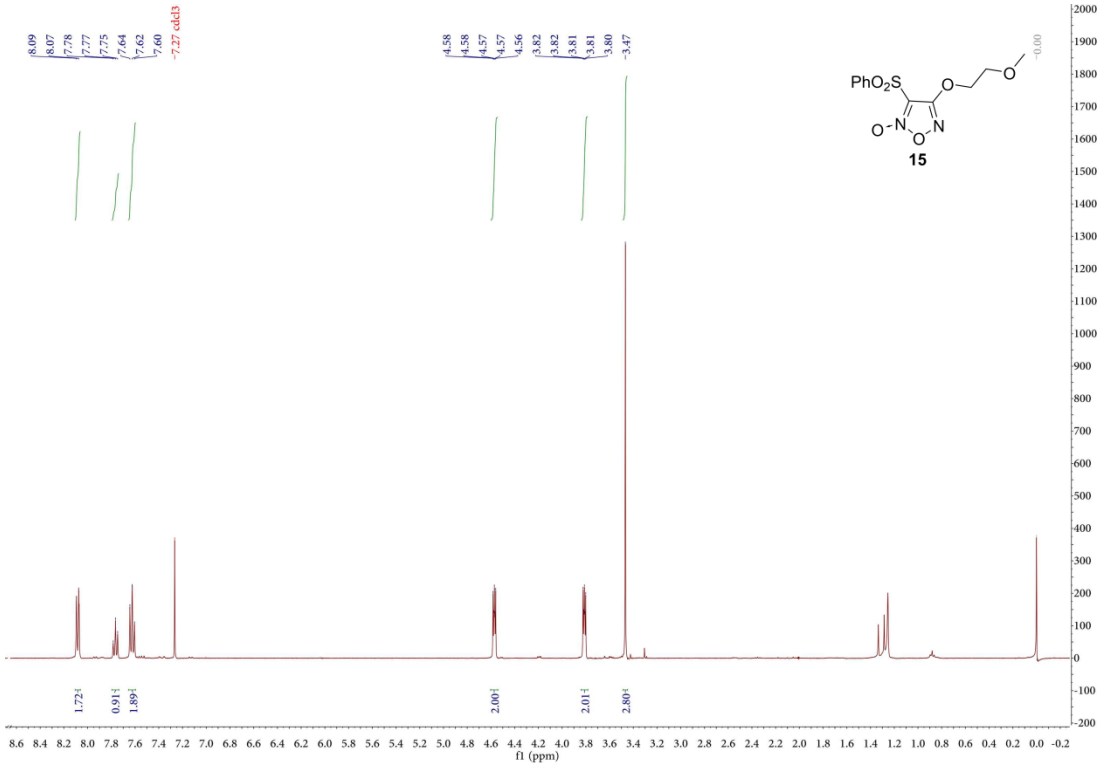


15

$C_{11}H_{12}N_2O_6S$
[300.0]

3-(2-methoxyethoxy)-4-(phenylsulfonyl)-1,2,5-oxadiazole 2-oxide (**15**). m.p. 99.5-101.2 °C, MS(ESI) (m/z) 301.1 $[M+H]^+$. ^1H NMR (400 MHz, Chloroform-*d*) δ 8.08 (d, *J* = 7.9 Hz, 2H, (2', 6'-SO_2Ph-H)), 7.77 (t, *J* = 7.5 Hz, 1H, 4'-SO_2Ph-H), 7.62 (t, *J* = 7.9 Hz, 2H, (3', 5'-SO_2Ph-H)), 4.60 – 4.54 (m, 2H, Furazanyl-O$\underline{CH_2}CH_2$), 3.84 – 3.78 (m, 2H, Furazanyl-$OCH_2\underline{CH_2}$), 3.47 (s, 3H, OCH_3).

^1H NMR spectra:



VIII. References

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