Supporting Information

Discovery of human TyrRS inhibitors by structure-based virtual screening, structural optimization, and bioassays

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Supporting Data

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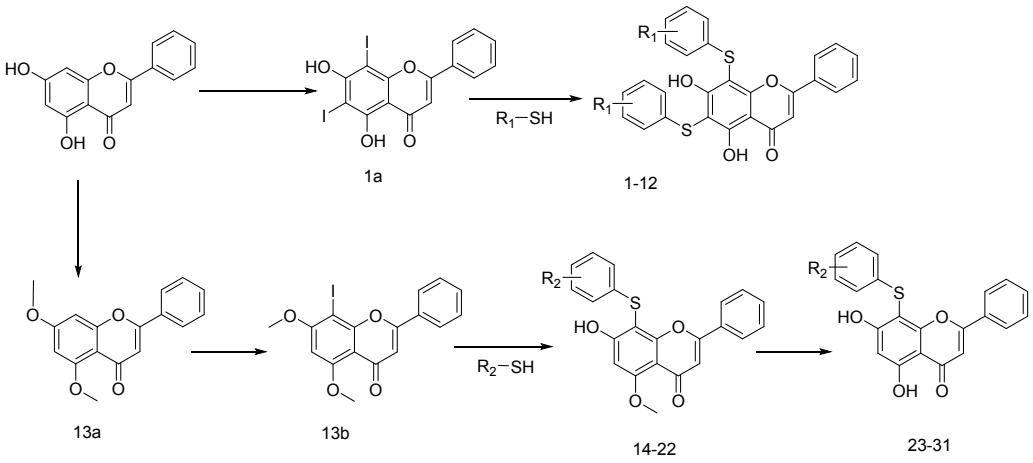
Chemical materials

All reagents were obtained from standard suppliers and used without further purification. The homogeneity of the compounds was monitored by ascending thin layer chromatography (TLC) on silica gel-G (Merck)-coated aluminum plates, visualized by UV light. NMR spectra were measured on a Bruker AM-400. The ^{1}H NMR (400 MHz) chemical shifts were given in ppm relative to the internal reference TMS. The ^{13} CNMR (100 MHz) chemical shifts were given using $CDCl_3$ and DMSO-d_6 as the internal standard. ESI-MS spectral data was recorded on a Finnigan LCQDECA and a Bruker Daltonics Bio TOF mass spectrometer.



Scheme S1 Simple synthetic route for all compounds.

Chemical synthesis experiments section

General procedure for the synthesis of compounds (1-12)

5,7-dihydroxy-6,8-diiodo-2-phenyl-4H-chromen-4-one (**1a**, 0.25 mmol, 1 equivalent), the derivative of benzenethiol (0.5 mmol, 2 equivalent), $Pd_2(dba)_3$ (0.0025 mmol, 0.01 equivalent), X-phos (0.005 mmol, 0.02 equivalent) and Cs_2CO_3 (0.75 mmol, 3 equivalent) were dissolved in *N, N*-dimethylformamide (DMF) under

inert gas and the reaction was stirred overnight at 110 °C. The filter catalysts, ligands and inorganic salts were filtered by celite to obtain black solution. After evaporation of the solvent, the sepia residue was dissolved in dichloromethane was extracted with water. The combined organic layers were dried over $MgSO_4$ and concentrated, and the residue was purified by column chromatography to give the desired product.

*5,7-dihydroxy-6,8-diiodo-2-phenyl-4H-chromen-4-one (**1a**)*

5,7-dihydroxy-2-phenyl-4H-chromen-4-one (5.08 g ,20 mmol) and iodine (5.76 g, 20 mmol) were dissolved in dichloromethane (20 mL) and glacial acetic acid (20 mL). The reaction was stirred 30 min at room temperature and the 2 g of 65 % HNO_3 acid was slowly dripped into the stirred solution using a constant pressure dropping funnel. After 2 h, the reaction was filtered, washed with 10% $Na_2S_2O_4$ solution, then the mixture of water and cold methanol and diethyl ether, and the solvent was removed in vacuo to afford the intermediate compound **1a** as a yellow solid (yield 70.0 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 14.50 (s, 1H), 8.04 (dd, J = 8.0, 1.6 Hz, 2H), 7.66-7.58 (m, 3H), 7.28 (s, 1H). ^{13}C NMR (101 MHz, DMSO) δ 181.6, 163.8, 162.8, 162.0, 155.6 , 130.4, 129.4, 126.8, 105.1, 104.7, 66.9. ESI-MS m/z: 506.04 $[M+H]^+$.

*6,8-bis((3-bromophenyl)thio)-5,7-dihydroxy-2-phenyl-4H-chromen-4-one (**1**)*

Follow the general method described above (yield 46.0 %). ^{1}H NMR (400 MHz, $CDCl_3$) δ 14.26 (s, 1H), 7.81 (d, J = 7.4 Hz, 2H), 7.53 (dt, J = 14.7, 7.1 Hz, 3H), 7.36 (d, J = 14.4 Hz, 2H), 7.33-7.27 (m, 2H), 7.20-7.06 (m, 4H), 6.79 (s, 1H). ESI-MS m/z: 626.59 $[M+H]^+$.

*6,8-bis((2-bromophenyl)thio)-5,7-dihydroxy-2-phenyl-4H-chromen-4-one (**2**)*

Follow the general method described above (yield 48.1 %). ^{1}H NMR (400 MHz, DMSO) δ 14.28 (s, 1H), 11.29 (s, 1H), 7.90 (s, 2H), 7.56 (d, J = 41.7 Hz, 5H), 7.23 (s, 3H), 7.07 (s, 2H), 6.79 (d, J = 33.6 Hz, 2H). ESI-MS m/z: 626.60 $[M+H]^{+}$.

*5,7-dihydroxy-6,8-bis((2-methoxyphenyl)thio)-2-phenyl-4H-chromen-4-one (**3**)*

Follow the general method described above (yield 51.9 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 14.16 (s, 1H), 10.76 (s, 1H), 7.89 (d, J = 6.5 Hz, 2H), 7.62-7.43 (m, 3H), 7.19 (s, 1H), 7.15-6.97 (m, 4H), 6.80 (d, J = 8.6 Hz, 2H), 6.70 (d, J = 6.8 Hz, 1H), 6.58 (d, J = 7.1 Hz, 1H), 3.93 (s, 3H), 3.88 (s, 3H). ESI-MS m/z: 528.84 $[M-H]^{-}$.

*5,7-dihydroxy-6,8-bis((3-methoxyphenyl)thio)-2-phenyl-4H-chromen-4-one (**4**)*

Follow the general method described above (yield 54.2 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 14.26 (s, 1H), 11.02 (s, 1H), 7.96 (s, 2H), 7.56 (d, J = 20.7 Hz, 3H), 7.19 (d, J = 14.6 Hz, 3H), 6.67 (dd, J = 25.1, 16.5 Hz, 6H), 3.67 (d, J = 6.6 Hz, 6H). ^{13}C NMR (101 MHz, DMSO) δ 182.62, 165.57, 163.94, 160.20, 160.11, 159.19, 138.35, 138.24, 132.89, 130.62, 130.41, 129.67, 126.85, 118.61, 118.48, 111.93, 111.89, 111.79, 111.29, 106.00, 101.13, 96.24, 55.56, 55.54. ESI-MS m/z: 528.89 $[M-H]^{-}$.

*6,8-bis((4-fluorophenyl)thio)-5,7-dihydroxy-2-phenyl-4H-chromen-4-one (**5**)*

Follow the general method described above (yield 42.0 %). ^{1}H NMR (400 MHz, $CDCl_3$) δ 14.18 (s, 1H), 7.82 (dd, J = 18.6, 6.3 Hz, 2H), 7.63-7.41 (m, 3H), 7.31 (s, 2H), 7.11 (s, 2H), 6.97 (m, 4H), 6.77 (s, 1H). ESI-MS m/z: 504.89 $[M-H]^{-}$.

*5,7-dihydroxy-2-phenyl-6,8-bis(p-tolylthio)-4H-chromen-4-one (**6**)*

Follow the general method described above (yield 48.1 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 14.20 (s, 1H), 10.91 (s, 1H), 7.95 (d, J = 7.5 Hz, 2H), 7.64-7.58 (m, 1H),

7.53 (t, *J* = 7.4 Hz, 2H), 7.19 (s, 1H), 7.09 (s, 4H), 7.03 (d, *J* = 8.2 Hz, 2H), 2.23 (s, 3H), 2.20 (s, 3H). ESI-MS m/z: 497.89 $[M-H]^-$.

6,8-bis((3-fluorophenyl)thio)-5,7-dihydroxy-2-phenyl-4H-chromen-4-one (7)

Follow the general method described above (yield 48.0 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 14.34 (s, 1H), 11.17 (s, 1H), 7.96 (d, *J* = 6.6 Hz, 2H), 7.56 (dd, *J* = 20.0, 6.3 Hz, 3H), 7.30 (d, *J* = 6.3 Hz, 2H), 7.23 (s, 1H), 6.95 (dd, *J* = 42.7, 25.2 Hz, 6H). ^{13}C NMR (101 MHz, DMSO) δ 182.7, 177.5, 163.4, 161.8, 161.0, 138.0, 131.1, 130.5, 130.1, 128.3, 127.3, 122.9, 113.6, 109.2, 104.8, 101.3.ESI-MS m/z: 504.85 $[M-H]^-$.

*5,7-dihydroxy-2-phenyl-6,8-bis(o-tolylthio)-4H-chromen-4-one (**8**)*

Follow the general method described above (yield 45.9 %). ^{1}H NMR (400 MHz, $CDCl_3$) δ 14.14 (s, 1H), 7.91 (s, 1H), 7.72 (d, *J* = 7.5 Hz, 2H), 7.51 (d, *J* = 6.9 Hz, 1H), 7.44 (t, *J* = 7.3 Hz, 2H), 7.19 (t, *J* = 7.1 Hz, 2H), 7.14-6.93 (m, 4H), 6.86 (d, *J* = 7.5 Hz, 1H), 6.79 (d, *J* = 7.6 Hz, 1H), 6.76 (s, 1H), 2.55 (s, 3H), 2.53 (s, 3H). ESI-MS m/z: 497.86 $[M-H]^-$.

*6,8-bis((4-(tert-butyl)phenyl)thio)-5,7-dihydroxy-2-phenyl-4H-chromen-4-one (**9**)*

Follow the general method described above (yield 36.0 %). ^{1}H NMR (400 MHz, $CDCl_3$) δ 14.13 (s, 1H), 8.09 (s, 1H), 7.79 (d, *J* = 7.5 Hz, 2H), 7.58 -7.50 (m, 1H), 7.47 (d, *J* = 7.6 Hz, 2H), 7.28 (m, 3H), 7.25-7.13 (m, 5H), 6.75 (s, 1H), 1.27 (s, 9H), 1.25 (s, 9H). ESI-MS m/z: 581.03 $[M-H]^-$.

*5,7-dihydroxy-2-phenyl-6,8-bis(pyridin-4-ylthio)-4H-chromen-4-one (**10**)*

Follow the general method described above (yield 48.0 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 14.35 (s, 1H), 11.94 (s, 1H), 8.27 (s, 4H), 7.87 (d, J = 7.3 Hz, 2H), 7.50

(dd, J = 12.9, 7.1 Hz, 3H), 7.13 (s, 2H), 7.05 (s, 2H), 6.91 (s, 1H). ESI-MS m/z: 410.88 $[M-H]^-$.

*5,7-dihydroxy-6,8-bis((3-hydroxyphenyl)thio)-2-phenyl-4H-chromen-4-one (**11**)*

Follow the general method described above (yield 42.0 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 14.19 (s, 1H), 10.96 (s, 1H), 9.34 (s, 1H), 9.31 (s, 1H), 7.90 (d, J = 7.4 Hz, 2H), 7.54 (d, J = 7.3 Hz, 1H), 7.49 (t, J = 7.3 Hz, 2H), 7.01 (dd, J = 16.0, 8.0 Hz, 3H), 6.61 (d, J = 8.0 Hz, 1H), 6.54-6.49 (m, 2H), 6.44 (s, 3H). ^{13}C NMR (101 MHz, DMSO) δ 182.6, 176.5, 163.1, 161.9, 159.8, 138.7, 132.1, 130.5, 130.1, 129.3, 127.3, 122.8, 112.4, 109.3, 104.5, 101.2. ESI-MS m/z: 500.85 $[M-H]^-$.

*5,7-dihydroxy-6,8-bis((2-hydroxyphenyl)thio)-2-phenyl-4H-chromen-4-one (**12**)*

Follow the general method described above (yield 42.2 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 14.13 (s, 1H), 10.36 (s, 3H), 7.99 (d, J = 7.3 Hz, 2H), 7.60 (s, 1H), 7.54 (d, J = 7.6 Hz, 2H), 7.14 (s, 1H), 6.98 (d, J = 6.8 Hz, 2H), 6.90-6.57 (m, 6H). ESI-MS m/z: 500.88 $[M-H]^-$.

General procedure for the synthesis of compounds (14-22)

Compounds **14-22** were prepared in a manner similar to that described for **1-12**.

*5,7-dimethoxy-2-phenyl-4H-chromen-4-one (**13a**)*

5,7-dihydroxy-2-phenyl-4H-chromen-4-one (2.54 g, 10 mmol), iodomethane (1.87 mL, 30 mmol) and K_2CO_3 (4.10 g, 30 mmol) were dissolved in dry DMF under inert gas and the reaction was stirred overnight at room temperature. Dichloromethane (100 mL) and HCl (0.1M, 50 mL) were added into reaction mixture to obtain the organic layers by filtration, and the solvent was removed in vacuo to afford the

intermediate compound **13a** as a yellow solid (yield 80 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.05 (dd, J = 7.6, 1.9 Hz, 2H), 7.57 (dd, J = 5.8, 4.8 Hz, 3H), 6.87 (d, J = 2.2 Hz, 1H), 6.78 (s, 1H), 6.52 (d, J = 2.2 Hz, 1H), 3.91 (s, 3H), 3.84 (s, 3H).

*8-iodo-5,7-dimethoxy-2-phenyl-4H-chromen-4-one (**13b**)*

13a (1.41 g, 5 mmol) and NIS (1.35 g, 6 mmol) were dissolved in Trifluoroacetic acid (TFA) and the reaction was stirred overnight at 80 °C. Then, the solid was separated out by adding the superfluous ethyl acetate. After filtration of the solvent, the residue was washed by saturated sodium bicarbonate, water, ethyl alcohol and diethyl ether to obtained the intermediate compound **13b** as a white solid (yield 70 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.18 (s, 2H), 7.59 (s, 3H), 6.88 (s, 1H), 6.72 (s, 1H), 4.03 (s, 3H), 3.95 (s, 3H).

*8-((3-bromophenyl)thio)-7-hydroxy-5-methoxy-2-phenyl-4H-chromen-4-one (**14**)*

Follow the general method described above (yield 30.2 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.40 (s, 1H), 7.84 (d, J = 7.1 Hz, 2H), 7.53-7.43 (m, 3H), 7.29 (dd, J = 7.2, 0.9 Hz, 2H), 7.19 (t, J = 8.1 Hz, 1H), 7.10 (d, J = 7.7 Hz, 1H), 6.82 (s, 1H), 6.65 (s, 1H), 3.89 (s, 3H). ESI-MS m/z: 452.98 $[M-H]^-$.

*8-((3-fluorophenyl)thio)-7-hydroxy-5-methoxy-2-phenyl-4H-chromen-4-one (**15**)*

Follow the general method described above (yield 28.0 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.38 (s, 1H), 7.88-7.81 (m, 2H), 7.52-7.42 (m, 3H), 7.28 (s, 1H), 6.97-6.88 (m, 3H), 6.83 (s, 1H), 6.65 (s, 1H), 3.89 (s, 3H). ESI-MS m/z: 392.85 $[M-H]^-$.

*8-((4-fluorophenyl)thio)-7-hydroxy-5-methoxy-2-phenyl-4H-chromen-4-one (**16**)*

Follow the general method described above (yield 33.1 %). ^{1}H NMR (400 MHz,

DMSO-d_6) δ 11.21 (s, 1H), 7.99-7.76 (m, 2H), 7.65-7.41 (m, 4H), 7.24 (d, J = 9.2 Hz, 2H), 6.93 (d, J= 9.1 Hz, 2H), 6.80 (s, 1H), 6.60(s, 1H), 3.83 (s, 3H), 3.64 (s, 3H). ESI-MS m/z: 392.90 $[M-H]^-$.

*7-hydroxy-5-methoxy-8-((3-methoxyphenyl)thio)-2-phenyl-4H-chromen-4-one (**17**)*

Follow the general method described above (yield 43.1 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.25 (s, 1H), 7.85 (d, J = 7.4 Hz, 2H), 7.56-7.41 (m, 3H), 7.15 (t, J = 8.2 Hz, 1H), 6.81 (s, 1H), 6.66 (d, J = 12.9 Hz, 4H), 3.88 (s, 3H), 3.66 (s, 3H). ESI-MS m/z: 405.02 $[M-H]^-$.

*8-((2-fluorophenyl)thio)-7-hydroxy-5-methoxy-2-phenyl-4H-chromen-4-one (**18**)*

Follow the general method described above (yield 28.9 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.39 (s, 1H), 7.89-7.79 (m, 2H), 7.48 (dt, J = 22.0, 7.1 Hz, 3H), 7.24 (dd, J = 9.9, 8.7 Hz, 1H), 7.19-7.09 (m, 1H), 7.07-6.97 (m, 1H), 6.82 (s, 1H), 6.78 (td, J = 7.9, 1.5 Hz, 1H), 6.65 (s, 1H), 3.88 (s, 3H). ESI-MS m/z: 405.06 $[M-H]^-$.

*8-((2-bromophenyl)thio)-7-hydroxy-5-methoxy-2-phenyl-4H-chromen-4-one (**19**)*

Follow the general method described above (yield 31.1 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.43 (s, 1H), 7.84-7.75 (m, 2H), 7.64 (dd, J = 7.9, 1.1 Hz, 1H), 7.49 (d, J = 7.3 Hz, 1H), 7.42 (t, J = 7.5 Hz, 2H), 7.20-7.11 (m, 1H), 7.06-6.97 (m, 1H), 6.82 (s, 1H), 6.67 (s, 1H), 6.63 (dd, J = 8.0, 1.4 Hz, 1H), 3.90 (s, 3H). ESI-MS m/z: 452.96 $[M-H]^-$.

*7-hydroxy-5-methoxy-8-((2-methoxyphenyl)thio)-2-phenyl-4H-chromen-4-one (**20**)*

Follow the general method described above (yield 35.0 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.16 (s, 1H), 7.77 (d, J = 7.5 Hz, 2H), 7.49 (t, J = 7.2 Hz, 1H), 7.42 (t, J

= 7.5 Hz, 2H), 7.11-6.99 (m, 2H), 6.79 (s, 1H), 6.76-6.69 (m, 1H), 6.65 (s, 1H), 6.52 (d, *J* = 7.7 Hz, 1H), 3.94 (s, 3H), 3.88 (s, 3H). ESI-MS m/z: 405.12 $[M-H]^-$.

*7-hydroxy-5-methoxy-8-((4-methoxyphenyl)thio)-2-phenyl-4H-chromen-4-one (**21**)*

Follow the general method described above (yield 38.1 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.19 (s, 1H), 7.97-7.89 (m, 2H), 7.57-7.46 (m, 3H), 7.18-7.10 (m, 2H), 6.83 (d, *J* = 8.9 Hz, 2H), 6.81 (s, 1H), 6.62 (s, 1H), 3.85 (s, 3H), 3.66 (s, 3H). ESI-MS m/z: 405.03 $[M-H]^-$.

*8-((2-chlorophenyl)thio)-7-hydroxy-5-methoxy-2-phenyl-4H-chromen-4-one (**22**)*

Follow the general method described above (yield 30.0 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.42 (s, 1H), 7.78 (d, *J* = 7.5 Hz, 2H), 7.53-7.45 (m, 2H), 7.42 (t, *J* = 7.5 Hz, 2H), 7.17-7.04 (m, 2H), 6.81 (s, 1H), 6.69 – 6.63 (m, 2H), 3.89 (s, 3H). ESI-MS m/z: 409.02 $[M-H]^-$.

General procedure for the synthesis of compounds (23-31)

14 or **15-22** (0.1 mmol, 1 equivalent) were dissolved in dichloromethane (20 ml). Boron tribromide (5.38mmol, 53.8 equivalents) was added and the reaction was stirred overnight at room temperature and then quenched with cold methanol. After regulation of pH value about 10, the water was added for extraction and washed by saturated salt solution. The combined organic layers were dried over $MgSO_4$ and concentrated, and the solvent was removed in vacuo to afford the desired product.

*8-((3-bromophenyl)thio)-5,7-dihydroxy-2-phenyl-4H-chromen-4-one (**23**)*

Follow the general method described above (yield 68.2 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 13.11 (s, 1H), 10.47 (s, 1H), 7.91 (d, *J* = 7.0 Hz, 2H), 7.52 (dd, *J* = 13.6,

7.4 Hz, 3H), 7.19 (dd, *J* = 20.1, 12.5 Hz, 3H), 7.13 (d, *J* = 1.7 Hz, 1H), 6.86 (s, 1H), 6.05 (s, 1H). ESI-MS m/z: 438.92 $[M-H]^-$.

*8-((3-fluorophenyl)thio)-5,7-dihydroxy-2-phenyl-4H-chromen-4-one (**24**)*

Follow the general method described above (yield 65.0 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 13.24 (s, 1H), 11.58 (s, 1H), 7.91 (d, *J* = 7.6 Hz, 2H), 7.62-7.54 (m, 1H), 7.50 (t, *J* = 7.5 Hz, 2H), 7.29 (dd, *J* = 14.4, 8.0 Hz, 1H), 7.10 (s, 1H), 7.01-6.88 (m, 3H), 6.48 (s, 1H). ESI-MS m/z: 378.72 $[M-H]^-$.

*8-((4-fluorophenyl)thio)-5,7-dihydroxy-2-phenyl-4H-chromen-4-one (**25**)*

Follow the general method described above (yield 75.1 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 13.22 (s, 1H), 11.51 (s, 1H), 7.95 (s, 2H), 7.53 (s, 3H), 7.21 (s, 2H), 7.10 (s, 3H), 6.47 (s, 1H). ESI-MS m/z: 378.78 $[M-H]^-$.

*5,7-dihydroxy-8-((3-hydroxyphenyl)thio)-2-phenyl-4H-chromen-4-one (**26**)*

Follow the general method described above (yield 44.9 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.21 (s, 1H), 9.38 (s, 1H), 7.91 (d, *J* = 7.0 Hz, 2H), 7.57-7.44 (m, 3H), 7.20 (s, 1H), 7.01 (s, 1H), 6.83 (s, 1H), 6.57 (d, *J* = 8.0 Hz, 1H), 6.45 (d, *J* = 6.9 Hz, 1H), 6.00 (s, 1H). ESI-MS m/z: 376.85 $[M-H]^-$.

*8-((2-fluorophenyl)thio)-5,7-dihydroxy-2-phenyl-4H-chromen-4-one (**27**)*

Follow the general method described above (yield 55.3 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 13.23 (s, 1H), 11.66 (s, 1H), 7.92 (d, *J* = 7.3 Hz, 2H), 7.56 (d, *J* = 7.2 Hz, 1H), 7.51 (t, *J* = 7.4 Hz, 2H), 7.23 (d, *J* = 9.3 Hz, 1H), 7.15 (d, *J* = 7.1 Hz, 1H), 7.11 (s, 1H), 7.02 (t, *J* = 7.6 Hz, 1H), 6.86 (dd, *J* = 7.9, 1.3 Hz, 1H), 6.48 (s, 1H). ESI-MS m/z: 378.98 $[M-H]^-$.

*8-((2-bromophenyl)thio)-5,7-dihydroxy-2-phenyl-4H-chromen-4-one (**28**)*

Follow the general method described above (yield 66.5 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 13.22 (s, 1H), 11.64 (s, 1H), 7.91 – 7.83 (m, 2H), 7.64 (dd, J = 7.9, 1.1 Hz, 1H), 7.55 (d, J = 7.3 Hz, 1H), 7.48 (t, J = 7.6 Hz, 2H), 7.21-7.13 (m, 1H), 7.11 (s, 1H), 7.02 (td, J = 7.7, 1.5 Hz, 1H), 6.69 (dd, J = 8.0, 1.4 Hz, 1H), 6.51 (s, 1H). ESI-MS m/z: 438.96 $[M-H]^-$.

*5,7-dihydroxy-8-((2-hydroxyphenyl)thio)-2-phenyl-4H-chromen-4-one (**29**)*

Follow the general method described above (yield 34.6 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 13.12 (s, 1H), 11.34 (s, 1H), 9.28 (s, 1H)，7.91 (d, J = 7.1 Hz, 2H), 7.64 (dd, J = 7.9, 1.1 Hz, 1H), 7.55 (d, J = 7.3 Hz, 1H), 7.28 (t, J = 7.6 Hz, 2H), 7.11-7.03 (m, 1H), 6.91 (s, 1H), 6.82 (td, J = 7.9, 1.6 Hz, 1H), 6.49 (dd, J = 8.0, 1.5 Hz, 1H), 6.45 (s, 1H). ESI-MS m/z: 376.93 $[M-H]^-$.

*5,7-dihydroxy-8-((4-hydroxyphenyl)thio)-2-phenyl-4H-chromen-4-one (**30**)*

Follow the general method described above (yield 42.0 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 13.16 (s, 1H), 11.37 (s, 1H), 9.41 (s, 1H), 8.01 (d, J = 7.0 Hz, 2H), 7.61-7.52 (m, 3H), 7.12-7.04 (m, 3H), 6.66 (d, J = 8.6 Hz, 2H), 6.45 (s, 1H). ESI-MS m/z: 376.96 $[M-H]^-$.

*8-((2-chlorophenyl)thio)-5,7-dihydroxy-2-phenyl-4H-chromen-4-one (**31**)*

Follow the general method described above (yield 48.0 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 13.26 (s, 1H), 11.57 (s, 1H), 10.48 (s, 1H), 7.93-7.79 (m, 2H), 7.50 (dd, J = 14.8, 12.3 Hz, 4H), 7.18-7.00 (m, 2H), 6.85 (s, 1H), 6.67 (d, J = 7.3 Hz, 1H), 6.06 (s, 1H). ESI-MS m/z: 394.83 $[M-H]^-$.

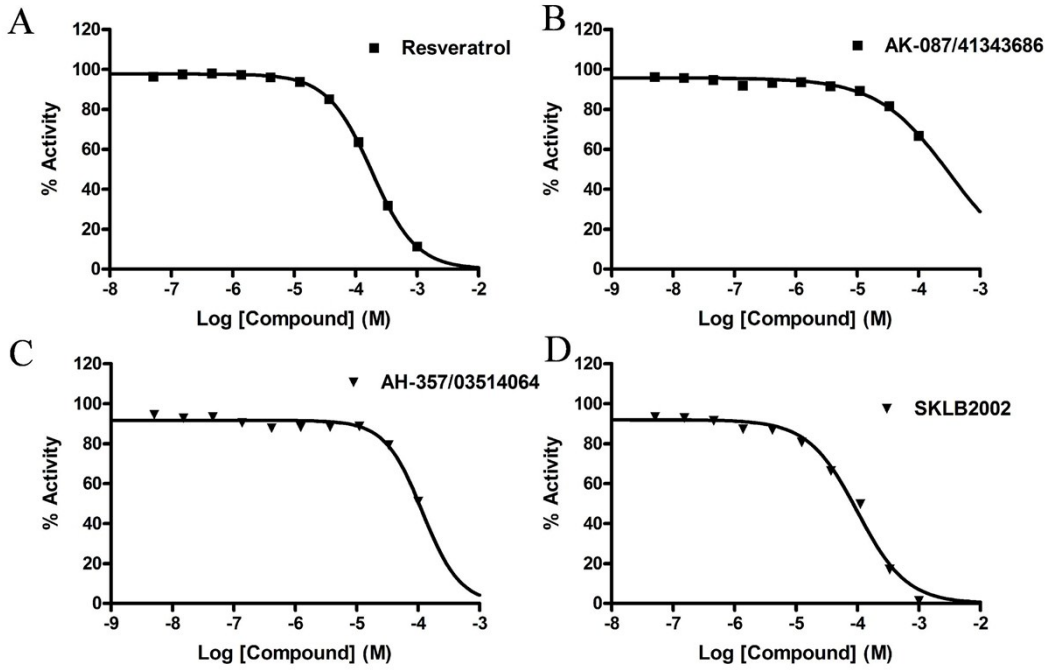


Fig. S1 Activity data against TyrRS of **resveratrol** and hit compounds selected by virtual screening. (A-D) The Ki values of **resveratrol**, **AK-087/41343686**, **AH-357/03514064**, and **SKLB2002**, respectively.

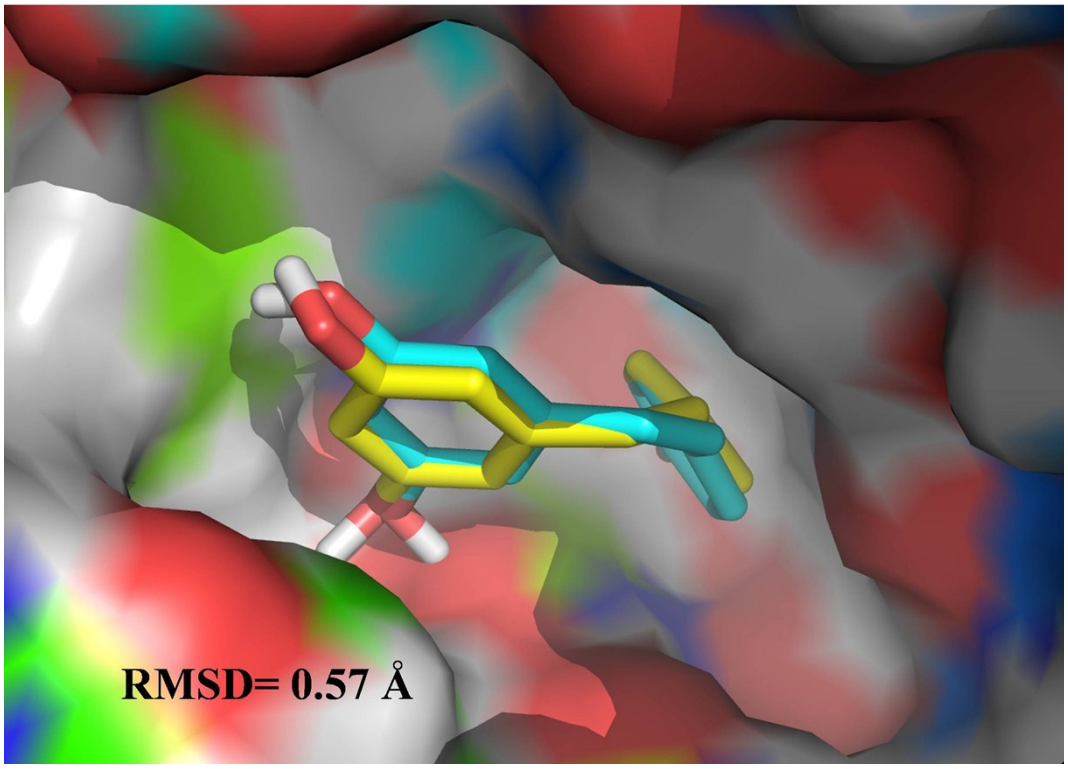


Fig. S2 The molecular docking method was validated. Resveratrol was docked into the TyrRS structure (PDB entry 4D93) according to the described protocol and was found in the same conformation as the Resveratrol ligand (yellow) in the crystal structure.

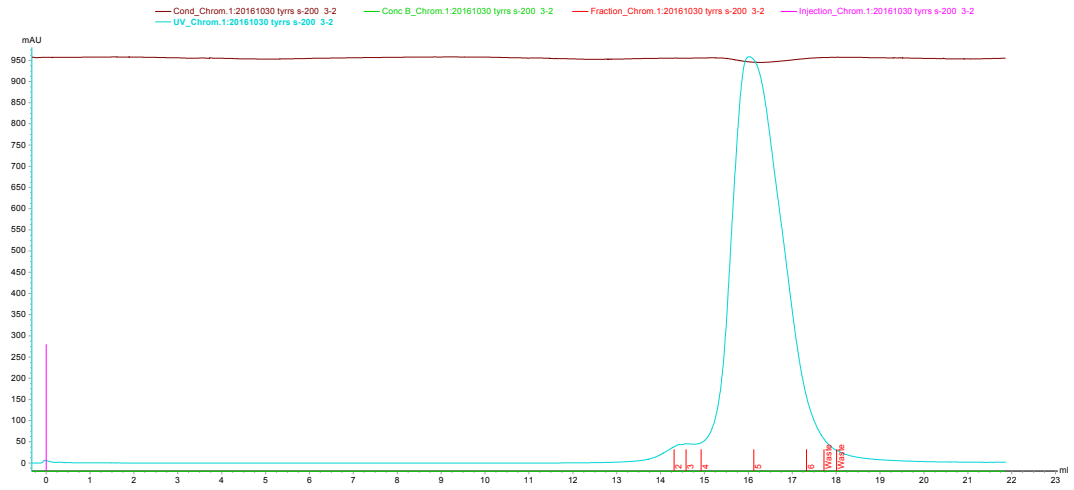


Fig. S3 The protein TyrRS peak corresponding to homogenous protein in buffer (20 mM Hepes-Na, pH = 7.5, 50 mM NaCl, 2 mM BME) by gel-filtration (Superdex 200; GE-Healthcare) chromatography.

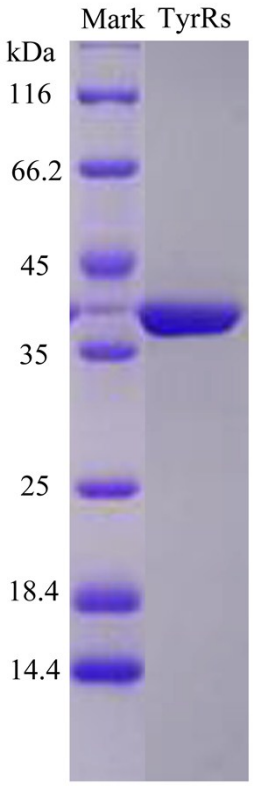
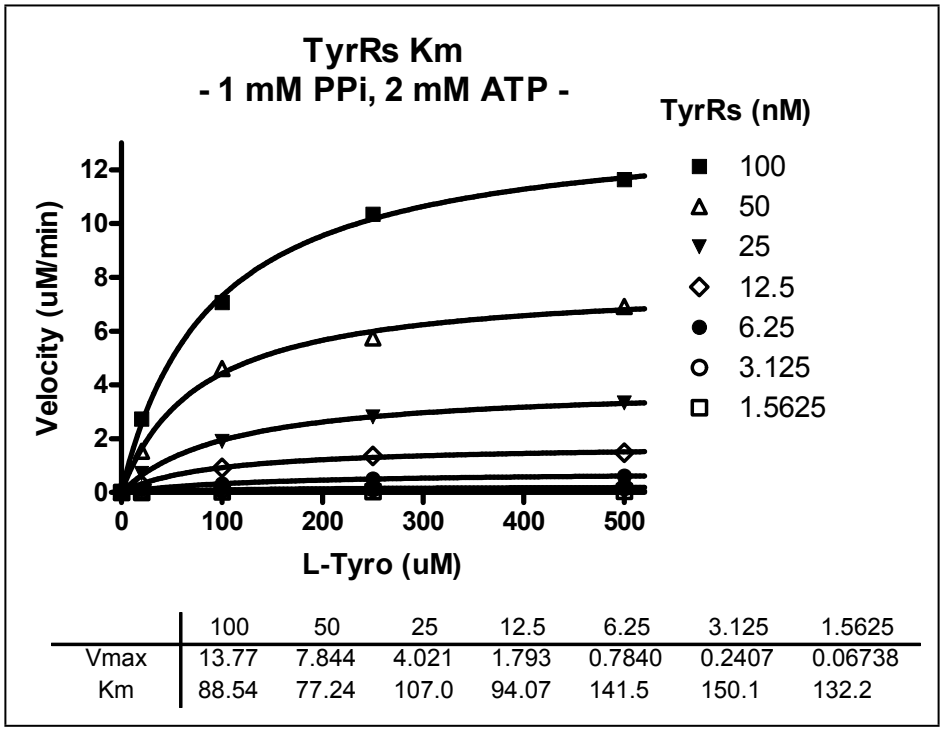
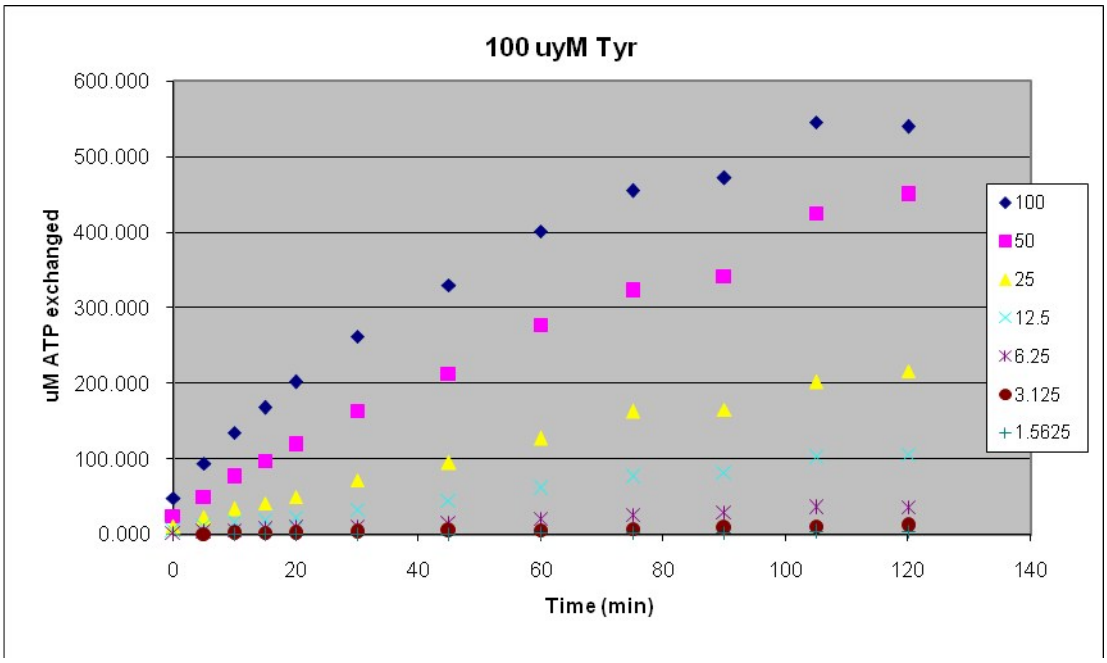


Fig. S4 The quality of protein purification was validated by SDS–PAGE analysis.



	100	50	25	12.5	6.25	3.125	1.5625
Vmax	13.77	7.844	4.021	1.793	0.7840	0.2407	0.06738
Km	88.54	77.24	107.0	94.07	141.5	150.1	132.2

Fig. S5 L-tyrosine K_m from time course data.



Slope	7.074	4.605	1.898	0.917	0.320	0.096	0.029

Fig. S6 Enzyme Titration from time course data.

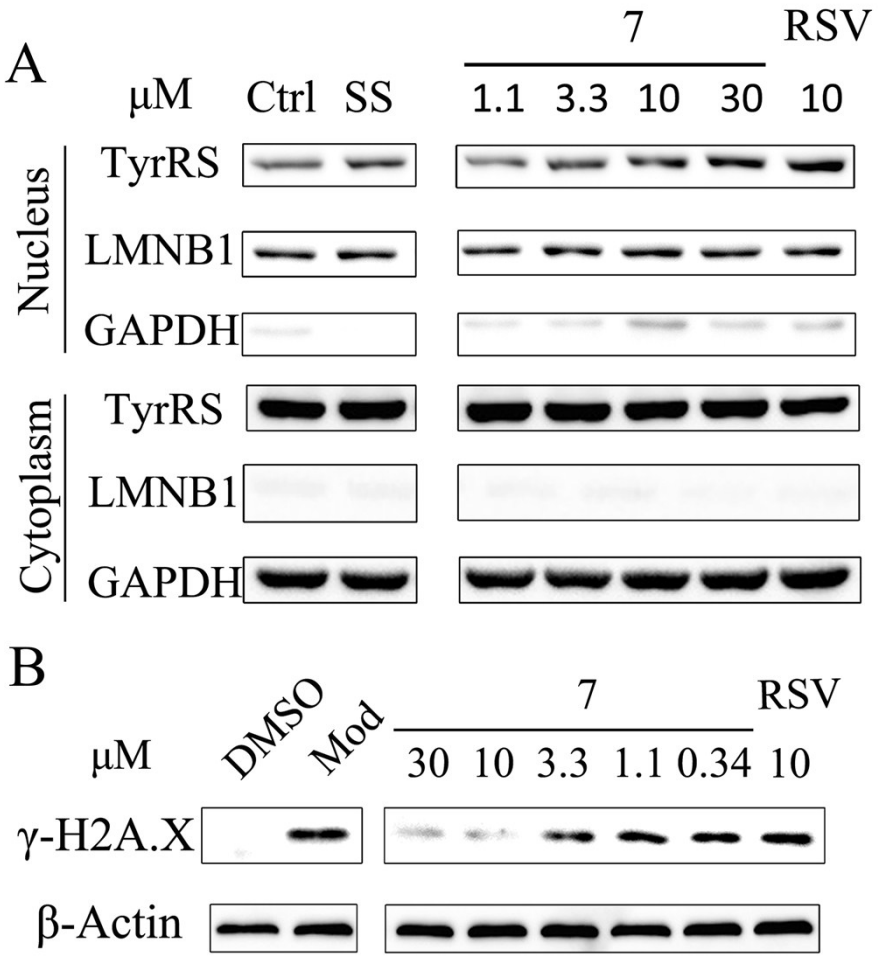


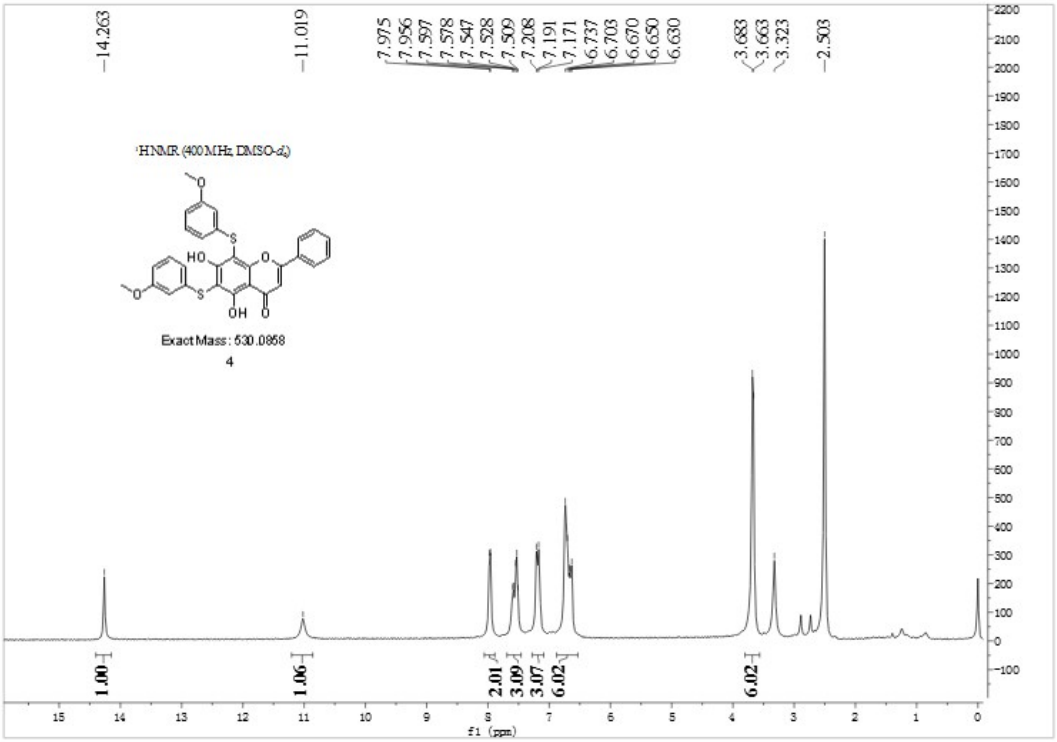
Fig. S7 (A) Compound **7** promote TyrRS nuclear localization in HeLa cells. Hela cells were treated with compound **7** (1.1, 3.3, 10, or 30 μM), RSV (1μM), or DMSO for 8 h. Cells cultured in serum-free media for 8 h were under serum starvation. Cytoplasmic fraction and nuclear fraction from cells were subjected to immunoblot assays. (B) Nuclear TyrRS prompted by compound **7** protect cells from DNA damage in HeLa cells. Hela cells were pretreated with compound **7** (30, 10, 3.3, 1.1, or 0.34 μM), RSV (10 μM), or DMSO for 24 h. To induced DNA damage, cells were treated with cisplatin (30 μM) or DMSO at the present of previous compounds or DMSO for 24 h. Then whole cell lysates were subjected to immunoblot assays. Quantification of immunoblots was showed below.

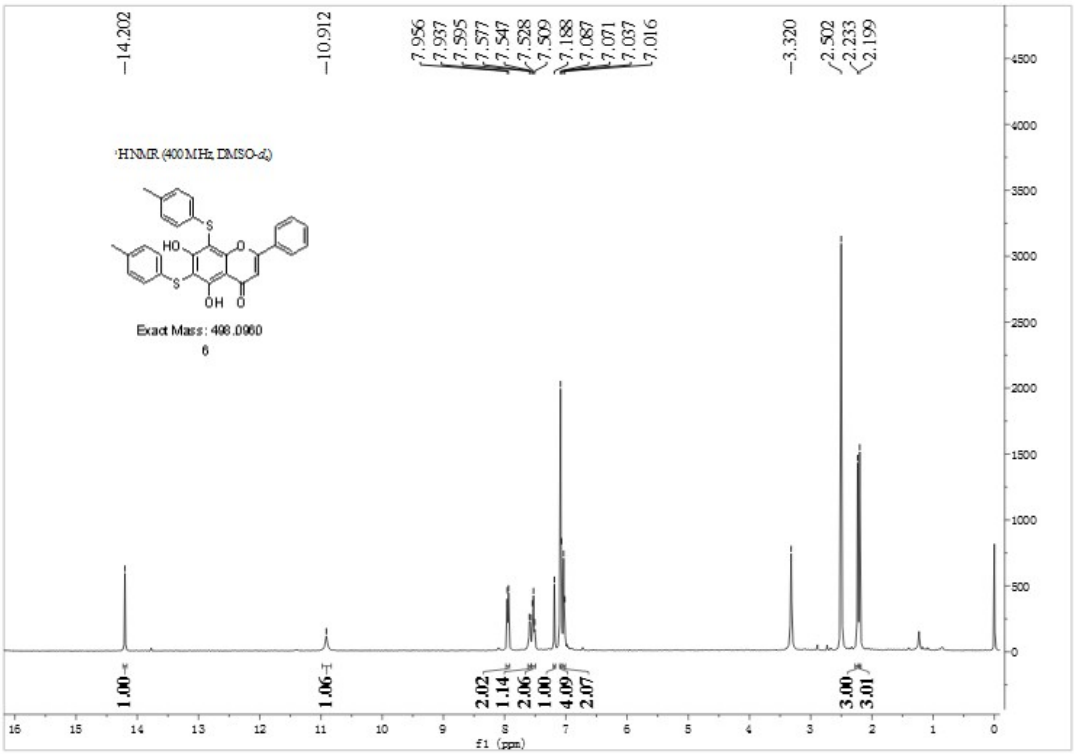
Table S1. The GoldScore and Enzyme Activity against TyrRS of 30 candidate compounds.

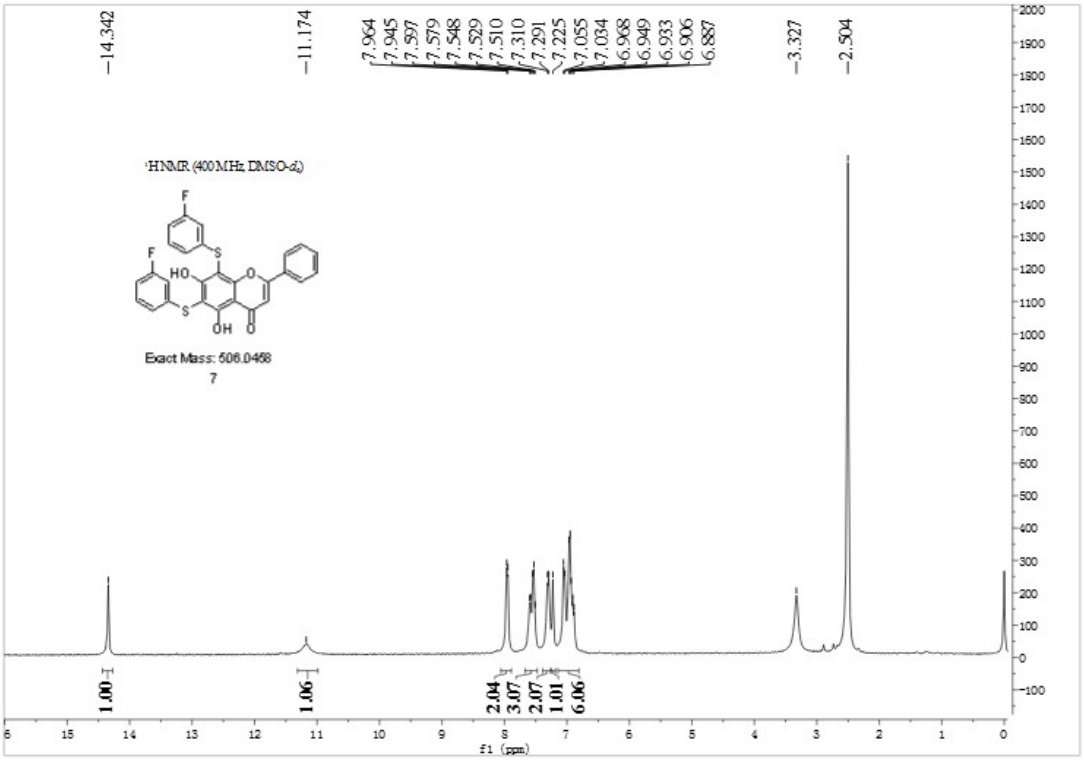
NO	IDNUMBER	SIMILE	GoldScore	IDScore	% Enzyme Activity (relative to DMSO controls)	
					Data 1	Data 2
1	AN-329/10722044	CCC(=N/N=C(CC)\C1=CC=C(O)C=C1)/C2=CC=C(O)C=C2	47.8686	5.57313	74.40	68.27
2	AA-516/31407002	CC(C)(C)OC(=O)NC(CC1=CC=C(O)C=C1)C(O)=O	45.7594	5.92388	92.75	89.53
3	AN-329/40366545	CC(=N/NC(=O)C1=CC(=CC(=C1)O)O)/C2=CC=C(O)C=C2	52.4547	6.38852	70.49	68.96
4	T5797242	CCC1=CC=C(C=CC(=O)C2=CC=C(O)C=C2)C=C1	46.2377	5.86204	67.28	65.79
5	T0510-4721	OC1=CC=C(C=C1)C=C(C#N)C(=O)NCC=C	43.9134	5.44258	93.86	93.28
6	T6351265	C[S](=O)(=O)C1=CC(=CC=C1)O	34.6108	4.51629	92.32	89.81
7	T6247548	CCNC(C)C1=CC(=CC=C1)O	40.7449	4.51629	90.11	88.90
8	T6203137	CNC(=O)C1=CC(=CC=C1)O	37.5035	5.62536	92.54	92.24
9	T5764459	NC(=O)C1=CC=C(O)C=C1	36.5454	4.27773	93.84	90.20
10	T6445890	CNCC1=CC(=CC=C1)O	33.1064	3.8742	104.54	99.24
11	T6542389	CC(N)CC1=CC(=CC=C1)O	37.5174	4.54192	87.07	86.58
12	AE-646/31213037	CCOC(=O)C1=CC=C(O)C=C1	37.5828	5.7979	82.19	79.15
13	AI-372/20970054	OC1=CC=C(C(=C1)O)C(=O)CC2=CC=CC=C2	40.7495	5.01859	75.85	75.78
14	AK-087/41343686	COC1=CC=C(/C=C/C(=O)C2=CC=C(O)C=C2)C=C1	49.9454	5.5113	61.50	55.48
15	AG-205/12075185	BrC1=CC=C(C=C1)C(=O)CSC2=NC=NC3=C2N=C[NH]3	41.385	6.29161	87.30	85.54
16	AE-641/11434265	CC(C)OC(=O)C1=C(C)NC2=C(N=C[N]2C1C3=CC=CC(=C3)[N+]([O-])=O)C(N)=O	48.6221	5.78024	91.13	90.84
17	AP-263/43370	CC1=CC=C(C=C1)C(=O)COC(=O)C2=C(O)C=C(O)C=C2	48.7816	5.5957	95.37	93.67

	871					
18	AG-205/37107180	COC1=CC=C(COC(=O)C2=C(C)NC(=O)NC2C3=CC(=C(O)C=C3)OC)C=C1	58.6627	6.23483	90.41	86.69
19	AK-087/40191206	COC(=O)C1=CC=C(O)C=C1	35.2655	4.7891	82.03	81.82
20	T6558601	CC(N)CC1=CC=C(O)C=C1	34.877	3.03893	78.42	77.50
21	AG-219/36431054	O=C(ON=C/1C)C1=C/C2=CC=C(OCC3=CC=CC=C3)C(OCC)=C2	50.4637	6.57768	101.45	91.33
22	AH-357/03514064	OC(C1=O)C(C2=CC=C(O)C(O)=C2)OC3=C1C(O)=CC(O)=C3	54.7992	6.47299	52.77	52.67
23	AE-765/20006021	OC1CC2=C(OC1C3=CC(=C(O)C=C3)O)C=C(O)C=C2O	51.5019	6.01997	87.11	87.01
24	AE-641/30085010	OC(=O)C(CC1=CC=CC=C1)NC(=O)OCC(Br)(Br)Br	41.6623	5.55134	87.20	86.31
25	AN-979/41971920	OC1=CC=CC(=C1)C(=O)/C=C/C2=CC=C(F)C=C2	42.8585	6.06173	89.12	88.77
26	AN-829/41530199	COC1=C(OC2=CC(=CC(=C2C1=O)O)O)C3=CC=CC=C3	46.3302	5.32877	92.12	91.67
27	AQ-911/42464272	OC1=C(OC2=CC=CC=C2C1=O)C3=CC=CC(=C3Cl)Cl	45.4803	6.62675	93.55	89.85
28	SKLB2002	O=C1C2=C(O)C(SC3=CC(Br)=CC=C3)=C(O)C(SC4=CC(Br)=CC=C4)=C2OC(C5=CC=CC=C5)=C1	68.2953	6.38619	31.76	30.05
29	AN-829/42007372	COC1=CC(=CC=C1O)/C=C/C(=O)C2=C(O)C=CC3=C2OC(=O)C=C3C	51.1084	6.37188	93.66	93.32
30	AJ-292/11529006	OC1=CC=C(C=C1)\C=C\C2=NC3=C(C=CC=C3)C=C2	44.7537	4.85796	103.91	96.77
31	Resveratrol	OC1=CC=C(C=C1)/C=C/C2=CC(O)=CC(O)=C2	44.6173	5.95076	61.98	63.12

Chemical spectrum section



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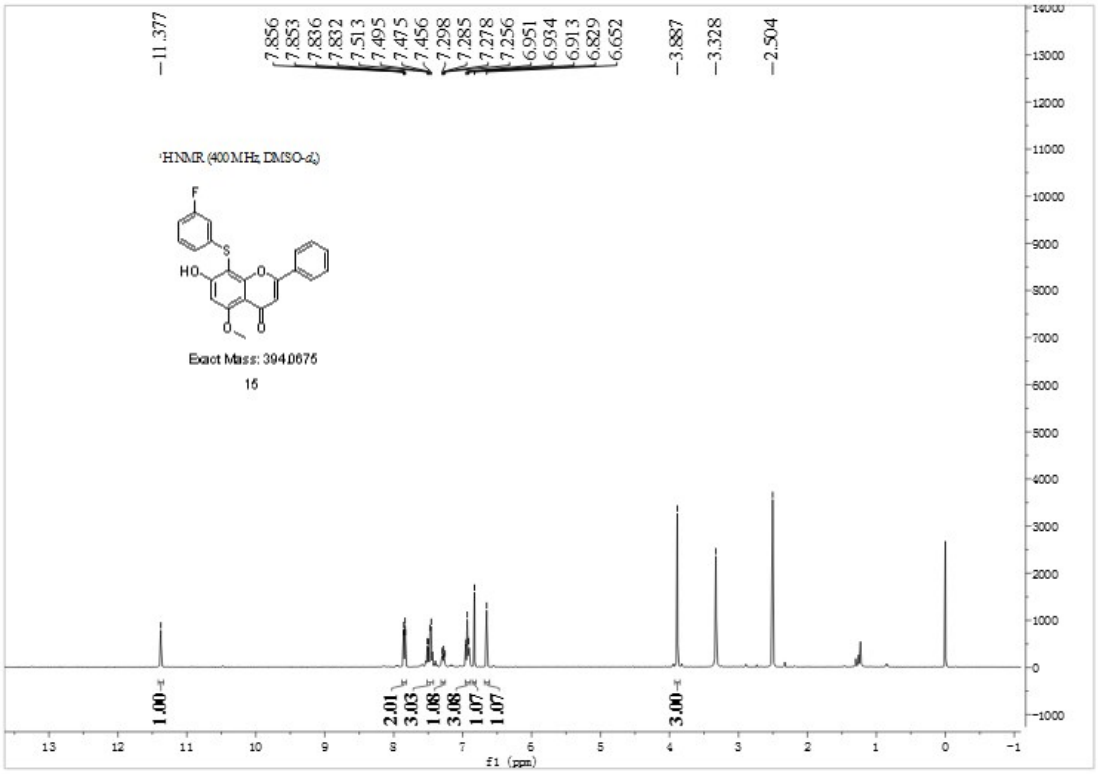
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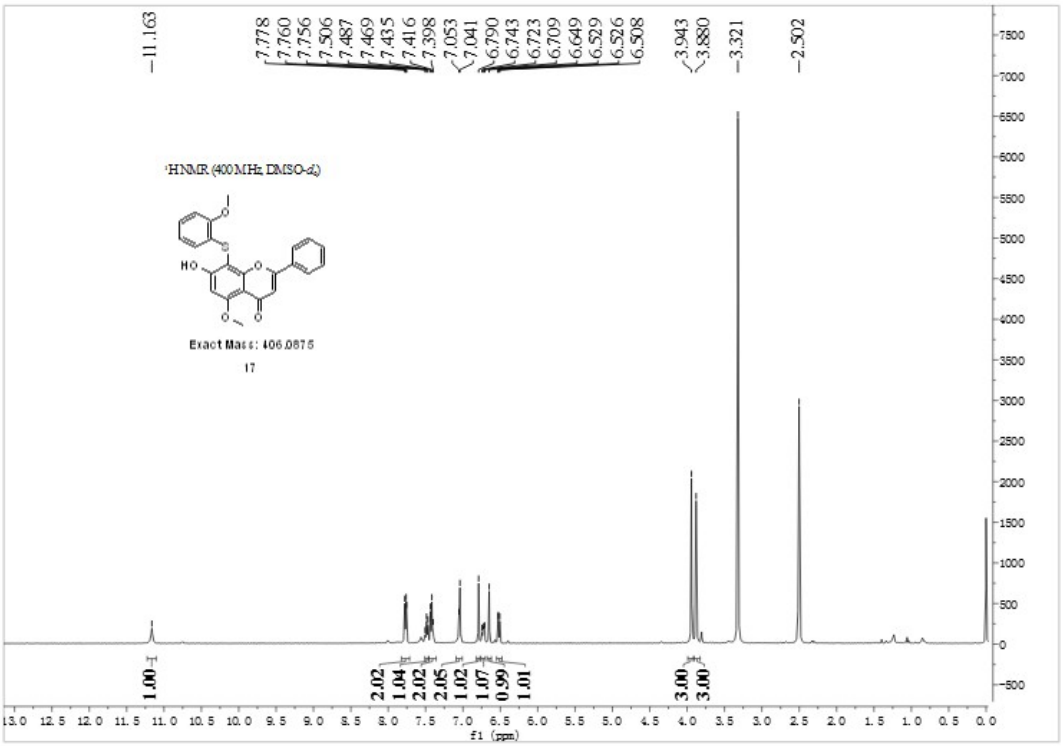
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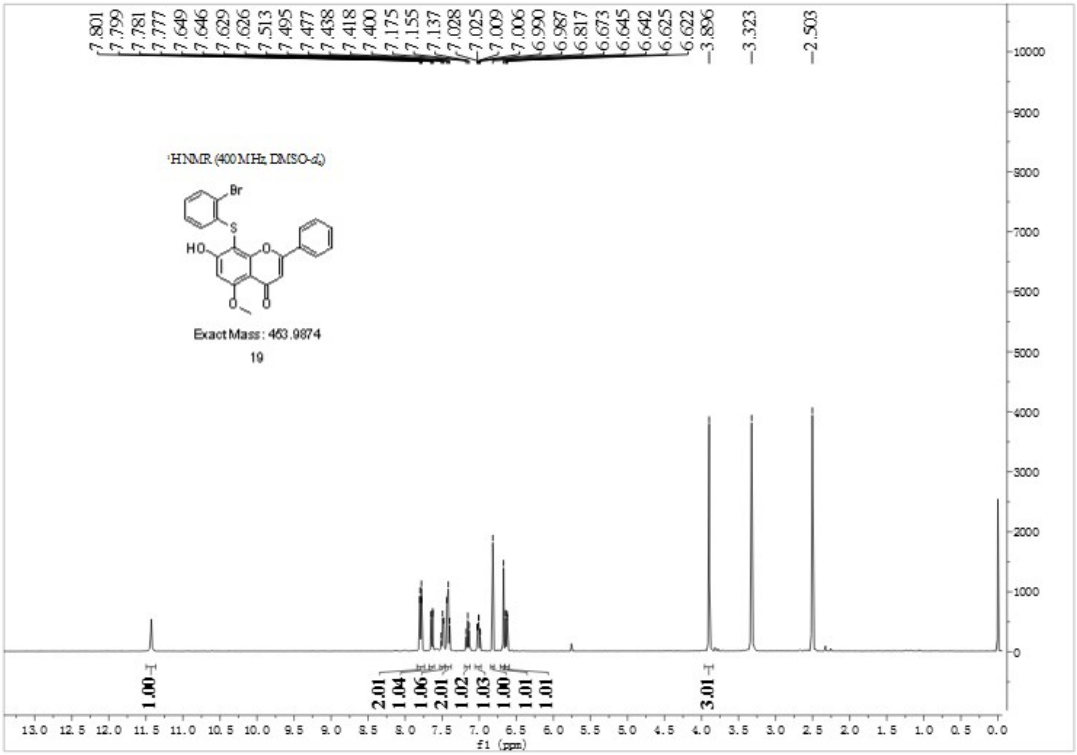
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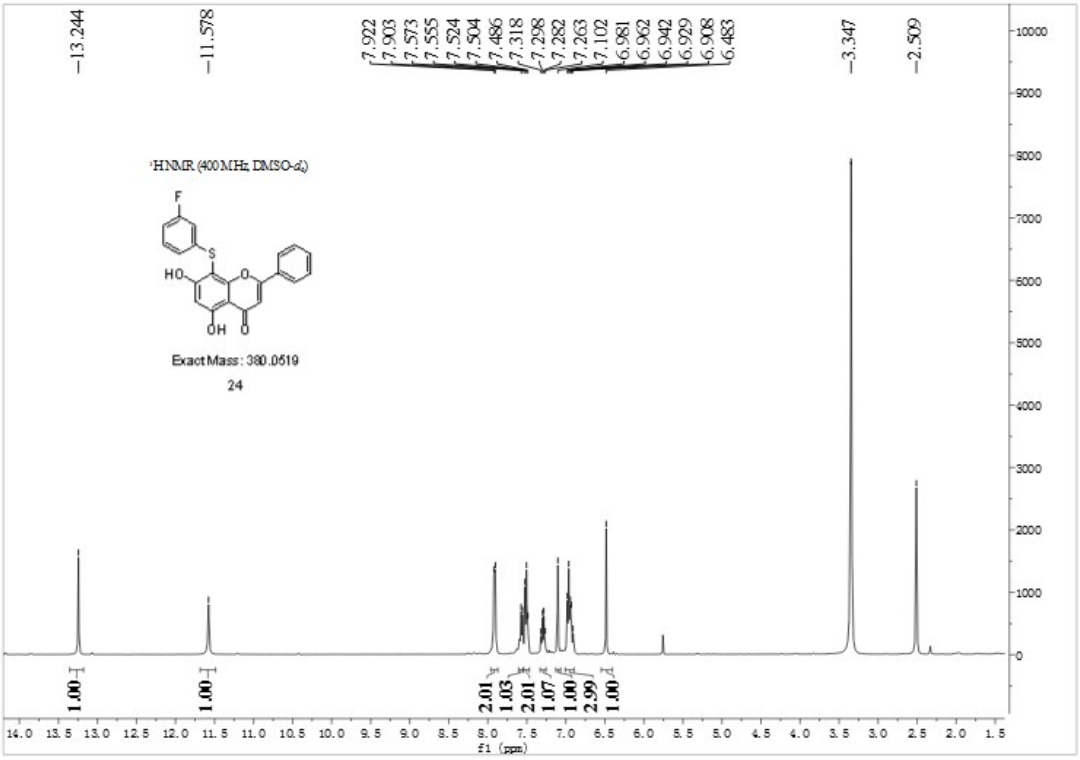
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