SUPPORTING INFORMATION FOR:

Rational design of a new fluorescent 'ON/OFF' xanthenic dye for phosphate detection in live cells

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SYNTHESIS SECTION: General Information

Deoxygenated solvents and reagents were used for all reactions involving Grignard or lithium reagents. THF was freshly distilled from Na. TLC was performed on aluminium-backed plates coated with silica gel 60 (230-240 mesh) with F254 indicator. The spots were visualized with UV light (254 nm) and/or staining with Ce/Mo reagent or phosphomolybdic acid solution and subsequent heating. Products were purified by flash chromatography on Merck silica gel 50. Yields refer to analytically pure samples.

^{1}H NMR spectra were recorded at 300 MHz. Chemical shifts are reported in ppm using residual solvent peak as reference ($CHCl_3$: δ 7.26, CH_3OH: δ 3.34). Data are reported as follows: chemical shift, multiplicity (s: singlet, d: doublet, t: triplet, m: multiplet, dd: doublet of doublets, dt: doublet of triplets, dq: doublet of quartets, td: triplet of doublets), coupling constant (*J* in Hz) and integration. ^{13}C-NMR spectra were recorded at 75.4 MHz using broadband proton decoupling and chemical shifts are reported in ppm using residual solvent peaks as reference ($CDCl_3$: δ 77.16, CD_3OD: δ 49.86). Carbon multiplicities were assigned by DEPT techniques. ^{1}H and ^{13}C NMR of compound **10** matched with previously described.[1] NMR Spectra were measured at room temperature.

High resolution mass spectra (HRMS) were recorded on a Micromass AutoSpec using EI at 70eV.

Synthetic Procedures.

Compound **1** (GG) was prepared in excellent yield (96 %) using a nucleophilic addition of the corresponding Grignard derivative of aryl bromide **9** to the TBDMS-protected 3,6-dihydroxy-xanthenone **10** and a subsequent dehydration with aqueous hydrochloric acid (Scheme S1).

Synthesis of compounds **8**[2] and **9**[3] was carried out according described protocols and their spectroscopic data matched with previously described.

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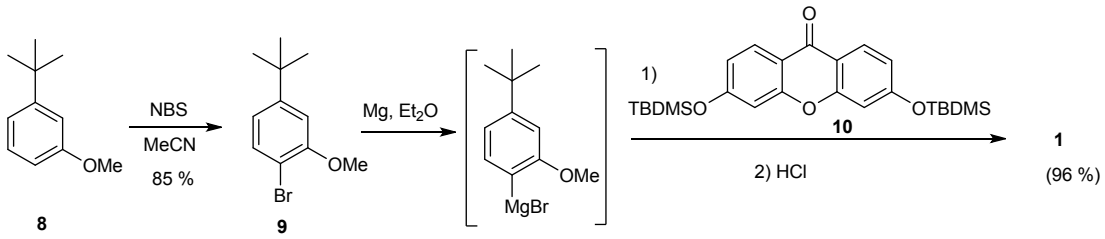
[1] R. L. M. Teeuwen, S. S. van Berkel, T. H. H. van Dulmen, S. Schoffelen, S. A. Meeuwissen, F. A. de Wolf, H. Zuilhof and J. C.M. van Hest, *Chem. Commun*, **2009**, 4022-4024

[2] S. Bhadra, I. W. Dzik, L. J. Goosen, *Angew. Chem. Int. Ed.* **2013**, *52*, 2959-2962.

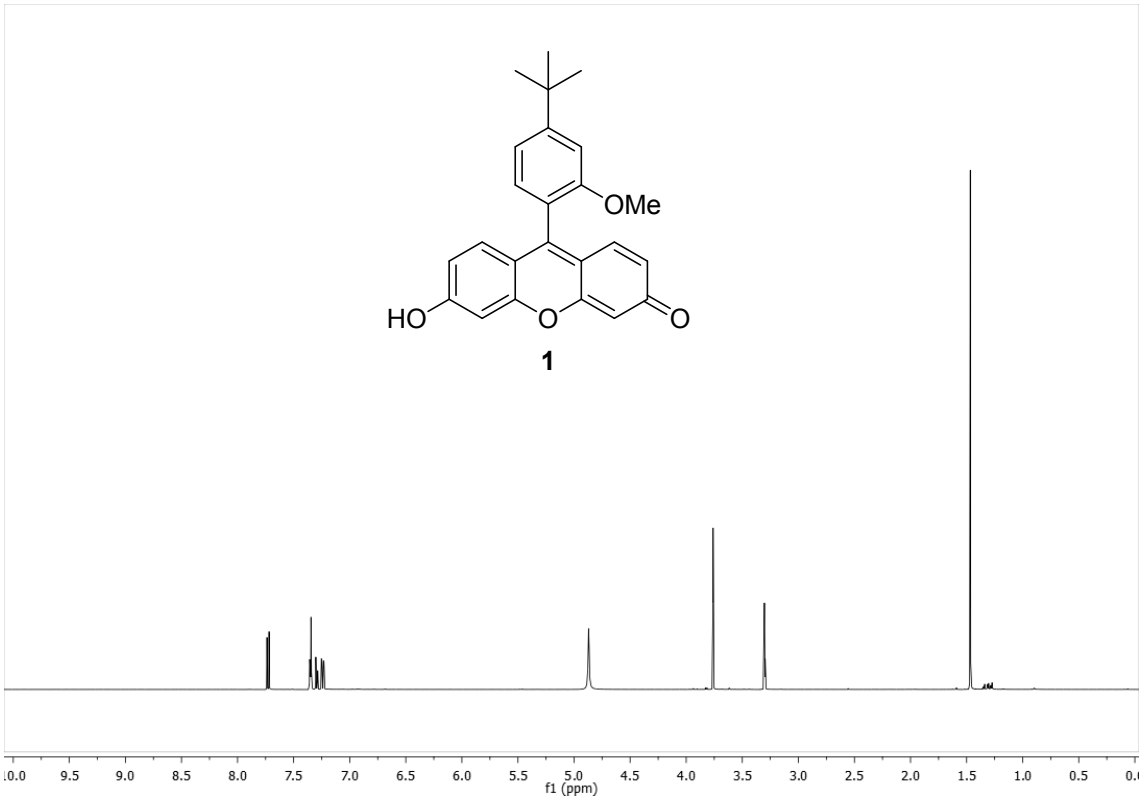
[3] H. Konishi, K. Aritomi, T. Okano, J. Kiji, *Bull. Chem. Soc. Jpn.* **1989**, *62*, 591-593.

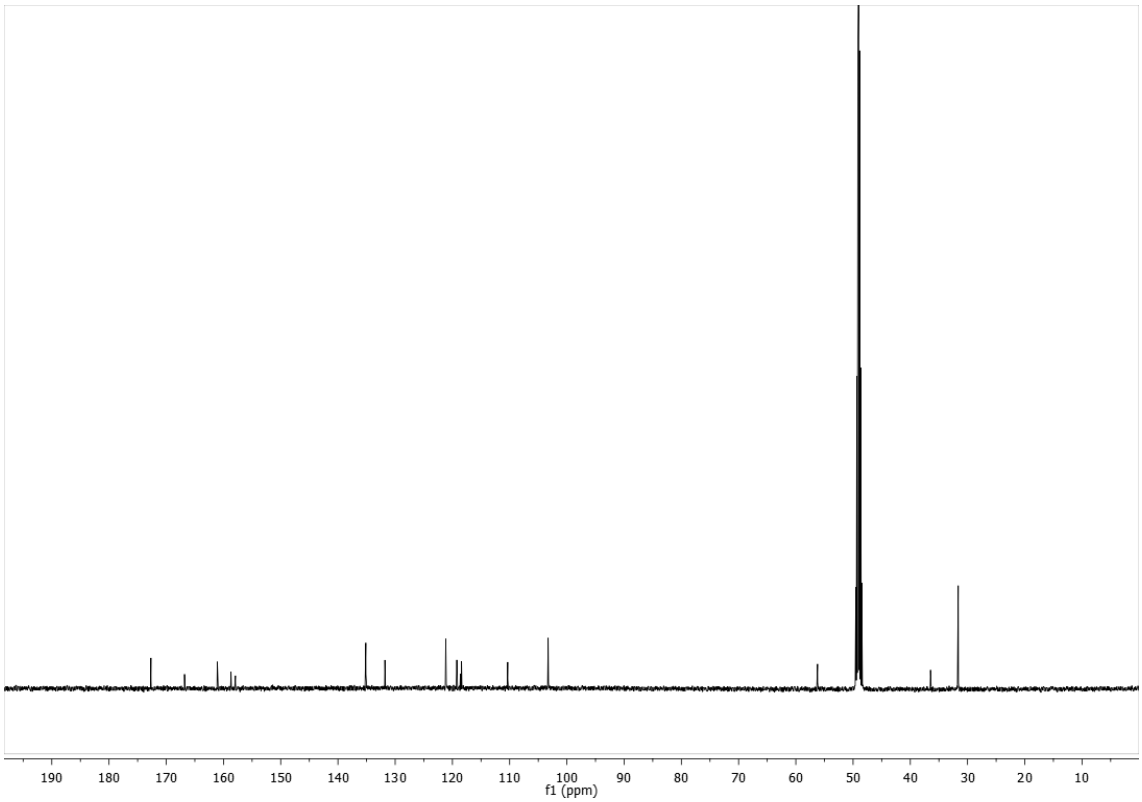


Scheme S1

To a solution of compound **10** (1 mmol) in THF (5 mL), the corresponding Grignard compound (5 mmol) was added, and the solution was stirred at room temperature for 2 h. Then, 2N HCl (10 mL) was added and the mixture was stirred for additional 10 min. The resulting solid is filtered and washed with water and diethyl ether, yielding compounds **1** (see Scheme 1). This compound was extensively purified by recrystallization in MeOH to give pure samples of the compound **1**, showing the following spectroscopic data: **^{1}H NMR** (300 MHz, CD_3OD) δ = 7.75 (s, 1H), 7.73 (s, 1H), 7.36 (dd, *J* = 4.8 Hz, 4H), 7.32-7.30 (m, 1H), 7.26 (t, *J* = 3.5 Hz, 1H), 7.24 (s, 1H), 3.77 (s, 3H), 1.48 (s, 9H); **^{13}C NMR** (75 MHz, CD_3OD): δ = 172.7 (C), 166.8 (C), 161.1 (C), 158.7 (C), 157.9 (C), 135.1 (CH), 131.8 (CH), 121.2 (CH), 119.2 (CH), 118.6 (C), 118.4 (C), 110.3 (CH_2), 103.3 (CH), 56.2 (CH_3), 36.5 (C), 31.6 (CH_3); **HRMS** (EI): *m*/*z* calcd. for $C_{24}H_{22}O_4$: [M^+] 374.1518; found: 374.1524.

Copies of ^{1}H-NMR and ^{13}C-NMR spectra of compound 1





THEORETICAL SECTION: Generalities

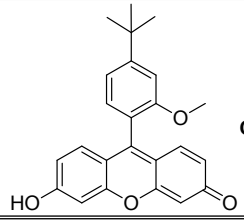
The calculations were carried out at the DFT-B3LYP level[4] with the Gaussian 09 program.[5] The geometries were fully optimized by the gradient technique with 6-31G* basis set for carbon, oxygen and hydrogen atoms.[6]

The nature of the optimized structures was assessed through a frequency calculation, and the changes of Gibbs free reaction energies (ΔG values) were obtained by taking into account zero-point energies, thermal motion, and entropy contribution at standard conditions (temperature of 298.15 K, pressure of 1 atm).

[4] (a) Becke, A. D. *Phys. Rev. A* **1988**, *38*, 3098-3100. (b) Lee, C.; Yang, W.; Parr, R. G. *Phys. Rev. B* **1988**, *37*, 785-789. (c) Becke, A. D. *J. Chem. Phys.* **1993**, *98*, 5648-5652.

[5] Gaussian 09, Revision B.01, Frisch, M. J.; Trucks, G. W.; Schlegel, H. B.; Scuseria, G. E.; Robb, M. A.; Cheeseman, J. R.; Scalmani, G.; Barone, V.; Mennucci, B.; Petersson, G. A.; Nakatsuji, H.; Caricato, M.; Li, X.; Hratchian, H. P.; Izmaylov, A. F.; Bloino, J.; Zheng, G.; Sonnenberg, J. L.; Hada, M.; Ehara, M.; Toyota, K.; Fukuda, R.; Hasegawa, J.; Ishida, M.; Nakajima, T.; Honda, Y.; Kitao, O.; Nakai, H.; Vreven, T.; Montgomery, Jr., J. A.; Peralta, J. E.; Ogliaro, F.; Bearpark, M.; Heyd, J. J.; Brothers, E.; Kudin, K. N.; Staroverov, V. N.; Kobayashi, R.; Normand, J.; Raghavachari, K.; Rendell, A.; Burant, J. C.; Iyengar, S. S.; Tomasi, J.; Cossi, M.; Rega, N.; Millam, N. J.; Klene, M.; Knox, J. E.; Cross, J. B.; Bakken, V.; Adamo, C.; Jaramillo, J.; Gomperts, R.; Stratmann, R. E.; Yazyev, O.; Austin, A. J.; Cammi, R.; Pomelli, C.; Ochterski, J. W.; Martin, R. L.; Morokuma, K.; Zakrzewski, V. G.; Voth, G. A.; Salvador, P.; Dannenberg, J. J.; Dapprich, S.; Daniels, A. D.; Farkas, Ö.; Foresman, J. B.; Ortiz, J. V.; Cioslowski, J.; Fox, D. J. Gaussian, Inc., Wallingford CT, **2010**.

[6] Hariharan, P. C.; Pople, J. A. *Mol. Phys.* **1974**, *27*, 209-214.



Compound 1

GEOMETRY for $C_{24}H_{22}0_4$

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C	8.944510000	3.846021000	1.617399000
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C	9.467834000	4.918778000	0.786577000
C	7.769589000	4.025823000	2.319045000
C	7.074871000	5.291371000	2.213861000
C	7.619207000	6.295662000	1.386667000
O	8.775053000	6.104949000	0.699637000
C	5.882339000	5.605086000	2.902646000
C	5.263315000	6.833103000	2.766092000
C	5.829412000	7.806587000	1.922433000
C	7.011354000	7.540840000	1.233094000
O	5.176842000	8.996176000	1.824309000
C	7.225431000	2.959368000	3.207790000
C	7.853724000	2.636234000	4.410657000
C	7.353459000	1.645675000	5.259950000
C	6.196062000	0.936364000	4.926477000
C	5.559230000	1.254866000	3.712734000
C	6.056928000	2.243894000	2.862867000
O	5.489803000	2.585057000	1.669676000
C	4.324900000	1.896045000	1.241788000
H	10.971978000	5.644614000	-0.528520000
H	4.348126000	7.069199000	3.298071000
H	7.475343000	8.276964000	0.582626000
H	8.753288000	3.177600000	4.690668000
H	4.664331000	0.715422000	3.426906000
H	3.490431000	2.039077000	1.940803000
H	4.515405000	0.822014000	1.119712000
H	4.063632000	2.328950000	0.274822000
H	11.437815000	1.571591000	0.932074000
H	5.449896000	4.858551000	3.559511000
H	5.662465000	9.581458000	1.221636000
O	12.467341000	3.462674000	-0.532980000
C	9.716494000	2.619584000	1.625933000
H	9.338546000	1.792522000	2.217558000
C	5.601983000	-0.164402000	5.825040000
H	7.882548000	1.440457000	6.182635000
C	4.168690000	0.235628000	6.249238000
H	3.508604000	0.367623000	5.385571000
H	4.175113000	1.175599000	6.812691000
H	3.729753000	-0.540424000	6.887907000
C	5.552888000	-1.499658000	5.044701000
H	6.556553000	-1.807760000	4.730815000
H	4.929351000	-1.428035000	4.147313000
H	5.135672000	-2.293292000	5.676155000
C	6.434810000	-0.387167000	7.101775000
H	6.482595000	0.515517000	7.721216000
H	7.459425000	-0.699745000	6.871207000
H	5.976406000	-1.177795000	7.706003000

THERMAL INFORMATION

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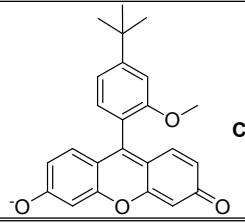

Compound 1_anion

GEOMETRY for $C_{24}H_{21}0_4$

C	10.758034000	2.430673000	0.811878000
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C	8.903333000	3.832505000	1.579287000
C	10.554589000	4.737253000	-0.013644000
C	9.424366000	4.882199000	0.746752000
C	7.735120000	4.070124000	2.325952000
C	7.091514000	5.318452000	2.260801000
C	7.638260000	6.343034000	1.414965000
O	8.780267000	6.095036000	0.691083000
C	5.907615000	5.669022000	2.987829000
C	5.327587000	6.898259000	2.879267000
C	5.869590000	7.951815000	2.014098000
C	7.073323000	7.584828000	1.289863000
O	5.328533000	9.074373000	1.919709000
C	7.192144000	2.996447000	3.215657000
C	7.755888000	2.746231000	4.464521000
C	7.265978000	1.750144000	5.317053000
C	6.179397000	0.959935000	4.936908000
C	5.603923000	1.202065000	3.675773000
C	6.095325000	2.196438000	2.826652000
O	5.577895000	2.463258000	1.591828000
C	4.458756000	1.725100000	1.146122000
H	10.905375000	5.563779000	-0.623601000
H	4.429582000	7.147968000	3.439701000
H	7.528025000	8.322772000	0.636500000
H	8.602425000	3.351356000	4.777188000
H	4.761843000	0.600505000	3.353890000
H	3.592098000	1.858262000	1.808998000
H	4.683595000	0.652122000	1.062574000
H	4.217607000	2.119539000	0.157063000
H	11.297709000	1.486439000	0.811025000
H	5.475033000	4.919069000	3.645782000
O	12.341525000	3.332827000	-0.716237000
C	9.631734000	2.597759000	1.562338000
H	9.258890000	1.779436000	2.173208000
C	5.596400000	-0.151151000	5.830473000
H	7.747921000	1.606876000	6.277507000
C	4.116027000	0.162934000	6.151859000
H	3.508657000	0.234513000	5.243432000
H	4.026906000	1.115426000	6.686548000
H	3.684000000	-0.624954000	6.782537000
C	5.681745000	-1.509749000	5.095112000
H	6.722501000	-1.762471000	4.862595000
H	5.124738000	-1.498504000	4.152386000
H	5.266246000	-2.311354000	5.719602000
C	6.354593000	-0.285026000	7.164977000
H	6.306542000	0.638031000	7.753465000
H	7.410101000	-0.535097000	7.009404000
H	5.908465000	-1.086206000	7.766403000

THERMAL INFORMATION

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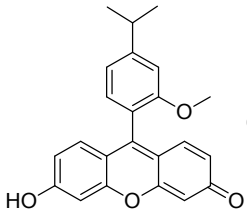
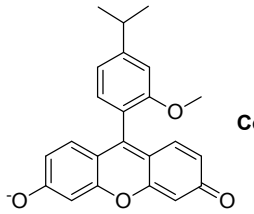


Compound 2



Compound 2_anion

GEOMETRY for $C_{23}H_{20}0_4$

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C	8.927936000	3.823381000	1.701984000
C	10.643849000	4.761172000	0.185549000
C	9.478889000	4.882666000	0.871850000
C	7.739406000	4.020412000	2.375094000
C	7.058849000	5.290899000	2.242924000
C	7.631613000	6.283004000	1.420090000
O	8.800241000	6.075014000	0.760154000
C	5.853521000	5.620899000	2.901065000
C	5.249775000	6.853685000	2.740887000
C	5.845070000	7.815261000	1.903629000
C	7.039800000	7.532814000	1.243474000
O	5.207004000	9.010542000	1.782128000
C	7.165274000	2.966640000	3.260891000
C	7.755784000	2.663599000	4.489674000
C	7.224798000	1.684309000	5.331482000
C	6.081342000	0.973535000	4.960578000
C	5.480653000	1.266762000	3.725811000
C	6.008958000	2.247343000	2.882519000
O	5.484749000	2.575896000	1.666883000
C	4.332006000	1.885106000	1.209215000
H	11.016639000	5.580211000	-0.419606000
H	4.324778000	7.102673000	3.249514000
H	7.525596000	8.259426000	0.598242000
H	8.645059000	3.210873000	4.790145000
H	4.596813000	0.716843000	3.423356000
H	3.477003000	2.036213000	1.881076000
H	4.523927000	0.809654000	1.103091000
H	4.101813000	2.309555000	0.230708000
H	11.413985000	1.519223000	1.094126000
H	5.398956000	4.883321000	3.553165000
H	5.712825000	9.587026000	1.187640000
O	12.489246000	3.383361000	-0.372269000
C	9.688323000	2.590282000	1.740580000
H	9.290823000	1.773657000	2.333998000
C	5.502532000	-0.099561000	5.873687000
H	7.707047000	1.474719000	6.282564000
C	4.074047000	0.245510000	6.335889000
H	6.137815000	-0.132030000	6.768815000
H	3.375610000	0.274330000	5.490855000
H	4.041682000	1.222504000	6.830130000
H	3.707091000	-0.507508000	7.043088000
C	5.552043000	-1.495145000	5.223075000
H	6.572218000	-1.757299000	4.922864000
H	4.917831000	-1.542607000	4.329708000
H	5.195145000	-2.258852000	5.923965000

THERMAL INFORMATION

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Sum of electronic and thermal Enthalpies = -1188.969424

Sum of electronic and thermal Free Energies = -1189.048941

GEOMETRY for $C_{23}H_{19}0_4$

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C	11.333784000	3.441239000	0.129779000
C	8.888507000	3.813552000	1.671457000
C	10.590019000	4.688251000	0.115117000
C	9.440279000	4.849919000	0.842154000
C	7.701429000	4.067569000	2.381999000
C	7.069154000	5.319561000	2.285266000
C	7.647473000	6.331082000	1.444593000
O	8.807330000	6.066856000	0.755631000
C	5.867333000	5.686139000	2.974098000
C	5.300180000	6.918494000	2.836602000
C	5.874929000	7.959034000	1.976702000
C	7.095993000	7.575679000	1.291018000
O	5.345822000	9.084753000	1.855959000
C	7.126453000	3.007852000	3.268770000
C	7.649380000	2.780235000	4.541316000
C	7.126789000	1.797498000	5.387729000
C	6.054390000	1.006484000	4.975387000
C	5.517999000	1.220658000	3.695293000
C	6.043107000	2.203320000	2.850511000
O	5.572374000	2.453747000	1.594002000
C	4.472032000	1.706484000	1.117243000
H	10.963897000	5.505338000	-0.493841000
H	4.388331000	7.180448000	3.368331000
H	7.574700000	8.303469000	0.643451000
H	8.484813000	3.391442000	4.871265000
H	4.685932000	0.610601000	3.360269000
H	3.580820000	1.847889000	1.744793000
H	4.701770000	0.632972000	1.058758000
H	4.268029000	2.085381000	0.113856000
H	11.286710000	1.442437000	0.998061000
H	5.410113000	4.945860000	3.626301000
O	12.385359000	3.263171000	-0.521391000
C	9.608243000	2.573750000	1.689396000
H	9.212735000	1.765234000	2.299146000
C	5.481068000	-0.066683000	5.892579000
H	7.559592000	1.650035000	6.374667000
C	4.012282000	0.212935000	6.262884000
H	6.064731000	-0.035377000	6.823014000
H	3.366829000	0.187824000	5.376665000
H	3.903471000	1.200558000	6.723979000
H	3.638228000	-0.539339000	6.968965000
C	5.640728000	-1.479415000	5.299510000
H	6.690231000	-1.694035000	5.070779000
H	5.069617000	-1.587482000	4.369591000
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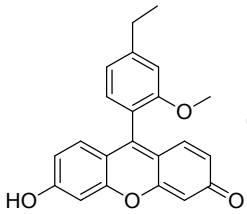
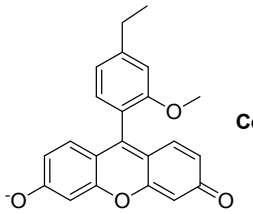


Compound 3



Compound 3_anion

GEOMETRY for $C_{22}H_{18}0_4$

C	10.885960000	2.474650000	1.088194000
C	11.463971000	3.578772000	0.295617000
C	8.932966000	3.815186000	1.709253000
C	10.670520000	4.802536000	0.249953000
C	9.489059000	4.896948000	0.911794000
C	7.727889000	3.985702000	2.359592000
C	7.035278000	5.250721000	2.236879000
C	7.613494000	6.264673000	1.445220000
O	8.798170000	6.082614000	0.806745000
C	5.812664000	5.555007000	2.875487000
C	5.197965000	6.783708000	2.725135000
C	5.799222000	7.767308000	1.918364000
C	7.010822000	7.510649000	1.278762000
O	5.150030000	8.957369000	1.804637000
C	7.144456000	2.908307000	3.210558000
C	7.703094000	2.593413000	4.452209000
C	7.163031000	1.592785000	5.261524000
C	6.041216000	0.873944000	4.841371000
C	5.474459000	1.175641000	3.594044000
C	6.012239000	2.179199000	2.783172000
O	5.522602000	2.518119000	1.556365000
C	4.387567000	1.825225000	1.058881000
H	11.046541000	5.637245000	-0.331289000
H	4.259569000	7.012607000	3.218425000
H	7.501410000	8.254480000	0.657178000
H	8.576395000	3.147098000	4.785990000
H	4.611877000	0.608786000	3.259619000
H	3.516346000	1.956459000	1.713763000
H	4.592053000	0.753717000	0.937971000
H	4.176372000	2.265217000	0.082982000
H	11.460198000	1.553094000	1.114375000
H	5.352981000	4.800584000	3.504463000
H	5.661592000	9.549710000	1.231024000
O	12.545371000	3.458293000	-0.290914000
C	9.707181000	2.590649000	1.741413000
H	9.305815000	1.758173000	2.309577000
C	5.417340000	-0.188189000	5.722407000
H	7.620554000	1.370889000	6.222068000
C	4.257462000	0.351216000	6.579137000
H	6.185630000	-0.611453000	6.380726000
H	5.050984000	-1.014108000	5.099283000
H	3.456821000	0.755274000	5.949276000
H	4.598706000	1.155987000	7.239694000
H	3.831126000	-0.443200000	7.202077000

GEOMETRY for $C_{22}H_{17}0_4$

C	10.717948000	2.399213000	0.913088000
C	11.292319000	3.465996000	0.085742000
C	8.862915000	3.812388000	1.656210000
C	10.553489000	4.715856000	0.105269000
C	9.411309000	4.865728000	0.846571000
C	7.684712000	4.054269000	2.384159000
C	7.057251000	5.310854000	2.324831000
C	7.632013000	6.340134000	1.503203000
O	8.782932000	6.087043000	0.795306000
C	5.860723000	5.663805000	3.030060000
C	5.297539000	6.901451000	2.927389000
C	5.870824000	7.961496000	2.090705000
C	7.084300000	7.590223000	1.384939000
O	5.346028000	9.092261000	2.002950000
C	7.108030000	2.974619000	3.246507000
C	7.546134000	2.796432000	4.559068000
C	7.022158000	1.794245000	5.380728000
C	6.032564000	0.936105000	4.899571000
C	5.583795000	1.098354000	3.580288000
C	6.109990000	2.102264000	2.760408000
O	5.721401000	2.309256000	1.468534000
C	4.720156000	1.477566000	0.918478000
H	10.925536000	5.544703000	-0.488706000
H	4.389509000	7.152965000	3.470544000
H	7.559897000	8.331445000	0.750414000
H	8.318696000	3.459899000	4.937854000
H	4.820479000	0.426752000	3.200114000
H	3.772839000	1.560613000	1.469790000
H	5.031100000	0.423395000	0.898518000
H	4.571940000	1.827793000	-0.104932000
H	11.249911000	1.450633000	0.915686000
H	5.403997000	4.908191000	3.664853000
O	12.336721000	3.298104000	-0.579602000
C	9.579776000	2.571323000	1.644161000
H	9.188188000	1.753795000	2.244557000
C	5.417731000	-0.127390000	5.786421000
H	7.389449000	1.682002000	6.398492000
C	4.132251000	0.345836000	6.488613000
H	6.148122000	-0.436770000	6.545024000
H	5.191740000	-1.021465000	5.189861000
H	3.370604000	0.637510000	5.756516000
H	4.332093000	1.218230000	7.120844000
H	3.711889000	-0.446122000	7.120976000

THERMAL INFORMATION

Sum of electronic and thermal Energies = -1149.686813

Sum of electronic and thermal Enthalpies = -1149.685869

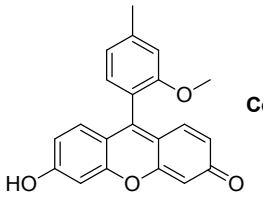
Sum of electronic and thermal Free Energies = -1149.761610

THERMAL INFORMATION

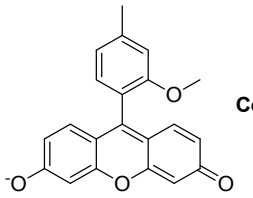
Sum of electronic and thermal Energies = -1149.167299

Sum of electronic and thermal Enthalpies = - 1149.166355

Sum of electronic and thermal Free Energies = -1149.242468



Compound 4



Compound 4_anion

GEOMETRY for $C_{21}H_{16}0_4$

C	10.862218000	2.443174000	1.163471000
C	11.465437000	3.530306000	0.366458000
C	8.915707000	3.814139000	1.737284000
C	10.687658000	4.762944000	0.293955000
C	9.497281000	4.879344000	0.936085000
C	7.701991000	4.006078000	2.365394000
C	7.026333000	5.277315000	2.216288000
C	7.629534000	6.275258000	1.422851000
O	8.822553000	6.072124000	0.806485000
C	5.796786000	5.602437000	2.830652000
C	5.198904000	6.836172000	2.656289000
C	5.825128000	7.803640000	1.848913000
C	7.044195000	7.526106000	1.232601000
O	5.191602000	8.999615000	1.711490000
C	7.093701000	2.945528000	3.219760000
C	7.634494000	2.633928000	4.468971000
C	7.072726000	1.647003000	5.281925000
C	5.946849000	0.939850000	4.856997000
C	5.396118000	1.238705000	3.600967000
C	5.955186000	2.227284000	2.787745000
O	5.480842000	2.563782000	1.554354000
C	4.346962000	1.875914000	1.047243000
H	11.082751000	5.586155000	-0.291053000
H	4.255203000	7.081482000	3.131203000
H	7.553729000	8.257380000	0.611391000
H	8.510564000	3.179528000	4.808541000
H	4.527523000	0.683121000	3.263691000
H	3.467751000	2.019504000	1.688736000
H	4.544546000	0.801804000	0.938335000
H	4.152694000	2.309390000	0.064942000
H	11.425013000	1.515365000	1.209803000
H	5.318460000	4.860290000	3.460196000
H	5.719634000	9.580377000	1.140970000
O	12.554008000	3.389761000	-0.202065000
C	9.674972000	2.581129000	1.796774000
H	9.255720000	1.760344000	2.369092000
C	5.315327000	-0.124913000	5.723239000
H	7.516269000	1.429632000	6.249634000
H	4.324546000	0.186552000	6.078314000
H	5.930083000	-0.339461000	6.602499000
H	5.179001000	-1.062088000	5.170579000

GEOMETRY for $C_{21}H_{16}0_4$

C	10.788179000	2.399349000	1.088098000
C	11.397453000	3.457788000	0.275650000
C	8.885727000	3.808334000	1.712220000
C	10.647077000	4.700172000	0.234069000
C	9.466165000	4.851647000	0.911728000
C	7.667743000	4.052153000	2.372425000
C	7.033221000	5.301331000	2.254467000
C	7.640424000	6.319629000	1.443067000
O	8.829804000	6.064968000	0.802543000
C	5.801649000	5.658343000	2.894014000
C	5.233443000	6.887979000	2.737525000
C	5.837310000	7.935139000	1.906171000
C	7.088400000	7.561671000	1.271083000
O	5.306809000	9.058195000	1.767350000
C	7.060124000	2.984254000	3.226989000
C	7.533954000	2.744982000	4.516273000
C	6.981283000	1.753317000	5.333533000
C	5.926969000	0.967666000	4.870568000
C	5.438596000	1.192449000	3.573825000
C	5.994172000	2.182870000	2.758931000
O	5.572555000	2.444032000	1.487446000
C	4.492355000	1.699424000	0.962732000
H	11.041574000	5.522175000	-0.354948000
H	4.298795000	7.142750000	3.231822000
H	7.589606000	8.294809000	0.647010000
H	8.355398000	3.353726000	4.883711000
H	4.620046000	0.580473000	3.208182000
H	3.577961000	1.833991000	1.557521000
H	4.726053000	0.626771000	0.904065000
H	4.325851000	2.086751000	-0.044358000
H	11.325354000	1.454727000	1.132172000
H	5.322377000	4.913037000	3.524275000
O	12.477300000	3.289098000	-0.330273000
C	9.611622000	2.572679000	1.755482000
H	9.195079000	1.758949000	2.343821000
C	5.309555000	-0.109406000	5.733835000
H	7.376064000	1.595895000	6.334478000
H	4.248739000	0.091726000	5.933698000
H	5.820333000	-0.185196000	6.699345000
H	5.362881000	-1.093648000	5.250452000

THERMAL INFORMATION:

Sum of electronic and thermal Energies = -1110.403499

Sum of electronic and thermal Enthalpies = -1110.402554

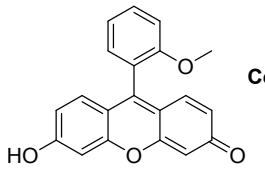
Sum of electronic and thermal Free Energies = -1110.475838

THERMAL INFORMATION:

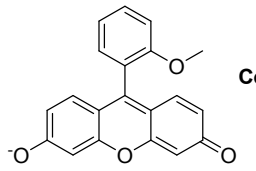
Sum of electronic and thermal Energies = - 1109.883882

Sum of electronic and thermal Enthalpies = - 1109.882938

Sum of electronic and thermal Free Energies = -1109.956538



Compound 5



Compound 5_anion

GEOMETRY for $C_{20}H_{14}0_4$

C	10.499829000	2.515629000	0.626124000
C	10.911900000	3.606967000	-0.160806000
C	9.472903000	2.679822000	1.536244000
C	8.815162000	3.918900000	1.701448000
C	10.294739000	4.849118000	-0.022176000
C	9.261503000	4.992215000	0.902574000
O	11.926377000	3.388374000	-1.040074000
C	7.739345000	4.153601000	2.640456000
C	7.190814000	5.416411000	2.731106000
C	7.693449000	6.490018000	1.888415000
O	8.711296000	6.230113000	0.999091000
C	6.099164000	5.766584000	3.617679000
C	5.597702000	7.021694000	3.670466000
C	6.119890000	8.118947000	2.830973000
C	7.206658000	7.756589000	1.926973000
O	5.653750000	9.261864000	2.895063000
C	7.271761000	3.039533000	3.516561000
C	7.587876000	3.026751000	4.878730000
C	7.167993000	1.988246000	5.712951000
C	6.416605000	0.946123000	5.176582000
C	6.082116000	0.934373000	3.820071000
C	6.505957000	1.976168000	2.987394000
H	11.002278000	1.562279000	0.503836000
H	9.161923000	1.840189000	2.148037000
H	10.597962000	5.710423000	-0.610818000
H	12.122188000	4.210310000	-1.517246000
H	5.688444000	4.981421000	4.243682000
H	4.777711000	7.276886000	4.335410000
H	7.615278000	8.525427000	1.280519000
H	8.176989000	3.845457000	5.282504000
H	5.490315000	0.117861000	3.422709000
O	6.223634000	2.058017000	1.656480000
C	5.447195000	1.029291000	1.059644000
H	6.080186000	0.130779000	5.811266000
H	4.449160000	0.966204000	1.511758000
H	5.943339000	0.053165000	1.137265000
H	5.351218000	1.303515000	0.007988000
H	7.427392000	1.997738000	6.767118000

THERMAL INFORMATION:

Sum of electronic and thermal Energies = -1071.114512

Sum of electronic and thermal Enthalpies = -1071.113568

Sum of electronic and thermal Free Energies = -1071.181450

GEOMETRY for $C_{20}H_{13}0_4$

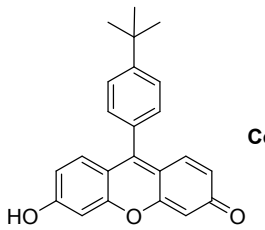
C	10.775302000	2.613941000	0.795929000
C	11.228393000	3.727712000	-0.044657000
C	9.706214000	2.717623000	1.636015000
C	8.947907000	3.927792000	1.757315000
C	10.449561000	4.946138000	0.087706000
C	9.378245000	5.027042000	0.937535000
O	12.209995000	3.622245000	-0.810744000
C	7.835831000	4.099578000	2.599837000
C	7.159694000	5.331079000	2.644088000
C	7.616421000	6.408106000	1.809451000
O	8.703582000	6.223594000	0.988678000
C	6.021515000	5.612503000	3.468227000
C	5.405854000	6.829207000	3.462166000
C	5.859274000	7.937758000	2.614874000
C	7.014096000	7.638239000	1.787042000
O	5.287334000	9.048832000	2.616302000
C	7.382842000	2.969465000	3.471889000
C	7.928473000	2.788535000	4.744226000
C	7.521839000	1.740049000	5.575838000
C	6.549001000	0.852958000	5.125993000
C	5.983659000	1.006884000	3.856663000
C	6.396949000	2.059362000	3.031055000
H	11.340272000	1.688063000	0.716452000
H	9.405162000	1.867003000	2.242697000
H	10.732800000	5.807311000	-0.509246000
H	5.653630000	4.818735000	4.114063000
H	4.543436000	7.026269000	4.094752000
H	7.399843000	8.417802000	1.137653000
H	8.687244000	3.489387000	5.081226000
H	5.226977000	0.306697000	3.520377000
O	5.901875000	2.284538000	1.779400000
C	4.906537000	1.415980000	1.277642000
H	6.219380000	0.031282000	5.757680000
H	4.000586000	1.431726000	1.899913000
H	5.269451000	0.381254000	1.200081000
H	4.663461000	1.787008000	0.280061000
H	7.963365000	1.623303000	6.561822000

THERMAL INFORMATION:

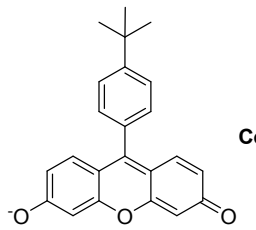
Sum of electronic and thermal Energies = -1070.595598

Sum of electronic and thermal Enthalpies = -1070.594653

Sum of electronic and thermal Free Energies = -1070.663920



Compound 6



Compound 6_anion

GEOMETRY for $C_{23}H_{20}O_3$

C	11.157590000	2.899979000	1.134672000
C	11.524697000	3.988588000	0.322747000
C	9.971946000	2.945285000	1.843793000
C	9.110052000	4.064736000	1.786156000
C	10.695012000	5.104659000	0.224203000
C	9.503470000	5.129523000	0.946620000
O	12.701792000	3.890310000	-0.349945000
C	7.853593000	4.172047000	2.500592000
C	7.080807000	5.304797000	2.331339000
C	7.528003000	6.365359000	1.442485000
O	8.733411000	6.236251000	0.789838000
C	5.825469000	5.543537000	3.015755000
C	5.101696000	6.667436000	2.808419000
C	5.537520000	7.730381000	1.880711000
C	6.817053000	7.499533000	1.217864000
O	4.860340000	8.747429000	1.695651000
C	7.423056000	3.059704000	3.393514000
C	8.141268000	2.749842000	4.559815000
C	7.733896000	1.711568000	5.391557000
C	6.603659000	0.927406000	5.099345000
C	5.898106000	1.241094000	3.930167000
C	6.295183000	2.286530000	3.093976000
H	11.814033000	2.038100000	1.184059000
H	9.688939000	2.099988000	2.460760000
H	10.946797000	5.954147000	-0.404430000
H	12.842691000	4.694043000	-0.875188000
H	5.479859000	4.787670000	3.712746000
H	4.163654000	6.840644000	3.327760000
H	7.191291000	8.261567000	0.543194000
H	9.016676000	3.337690000	4.822706000
H	5.020140000	0.669105000	3.651777000
C	6.192956000	-0.214939000	6.045441000
H	8.311593000	1.513749000	6.290169000
C	5.902147000	0.362247000	7.451450000
C	4.931921000	-0.953301000	5.558751000
C	7.346849000	-1.241492000	6.140489000
H	5.725632000	2.498645000	2.193331000
H	5.083810000	1.090254000	7.415419000
H	6.777513000	0.864053000	7.876847000
H	5.612106000	-0.441077000	8.139316000
H	5.082221000	-1.415276000	4.576320000
H	4.065792000	-0.284875000	5.495011000
H	4.679121000	-1.753180000	6.263395000
H	8.266741000	-0.787393000	6.523246000
H	7.569607000	-1.673278000	5.158067000
H	7.073293000	-2.060085000	6.817089000

GEOMETRY for $C_{23}H_{19}O_3$

C	11.178817000	2.915287000	1.159679000
C	11.620773000	4.025120000	0.308947000
C	10.004021000	2.943650000	1.851476000
C	9.115056000	4.068778000	1.802458000
C	10.700316000	5.146438000	0.248448000
C	9.524383000	5.152461000	0.950635000
O	12.701913000	3.991702000	-0.315999000
C	7.888849000	4.164150000	2.490033000
C	7.076673000	5.305449000	2.337099000
C	7.515465000	6.363393000	1.467713000
O	8.716948000	6.255441000	0.809730000
C	5.830685000	5.529256000	3.012471000
C	5.092076000	6.659042000	2.818747000
C	5.514863000	7.733029000	1.913970000
C	6.789166000	7.505260000	1.256457000
O	4.827777000	8.761492000	1.739080000
C	7.448967000	3.052297000	3.384830000
C	8.142030000	2.753733000	4.568152000
C	7.727295000	1.718942000	5.402221000
C	6.605715000	0.928294000	5.098399000
C	5.919812000	1.229720000	3.914487000
C	6.330243000	2.269361000	3.076810000
H	11.838209000	2.051919000	1.209383000
H	9.715779000	2.091337000	2.460806000
H	10.959156000	5.993990000	-0.378267000
H	5.479098000	4.765970000	3.701280000
H	4.150502000	6.809999000	3.341552000
H	7.168958000	8.270317000	0.586708000
H	9.013208000	3.345960000	4.833703000
H	5.047781000	0.652899000	3.624658000
C	6.182716000	-0.207295000	6.048381000
H	8.293689000	1.529201000	6.310902000
C	5.866728000	0.377868000	7.445376000
C	4.931345000	-0.954444000	5.549920000
C	7.334730000	-1.232344000	6.173714000
H	5.775265000	2.476146000	2.166043000
H	5.048018000	1.104246000	7.387977000
H	6.733866000	0.888498000	7.876904000
H	5.568220000	-0.419441000	8.138483000
H	5.098760000	-1.420838000	4.572423000
H	4.066006000	-0.287300000	5.465482000
H	4.669020000	-1.750613000	6.256851000
H	8.248429000	-0.769433000	6.560363000
H	7.572266000	-1.672841000	5.198617000
H	7.055406000	-2.045100000	6.856899000

THERMAL INFORMATION:

Sum of electronic and thermal Energies = -1113.765131

Sum of electronic and thermal Enthalpies = -1113.764187

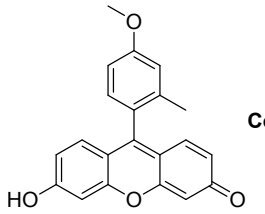
Sum of electronic and thermal Free Energies = -1113.838428

THERMAL INFORMATION:

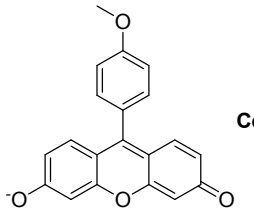
Sum of electronic and thermal Energies = -1113.248877

Sum of electronic and thermal Enthalpies = -1113.247932

Sum of electronic and thermal Free Energies = -1113.321743



Compound 7



Compound 7_anion

GEOMETRY for $C_{21}H_{16}0_4$

C	10.748270000	2.547849000	0.795195000
C	11.127635000	3.637596000	-0.010283000
C	9.673463000	2.676381000	1.655024000
C	8.933802000	3.876994000	1.747380000
C	10.428683000	4.842541000	0.056817000
C	9.347433000	4.950340000	0.929454000
O	12.191833000	3.454993000	-0.836314000
C	7.801520000	4.068766000	2.628592000
C	7.172242000	5.296244000	2.649494000
C	7.639448000	6.371975000	1.789197000
O	8.711973000	6.150168000	0.954061000
C	6.029779000	5.603125000	3.486536000
C	5.442782000	6.821605000	3.471413000
C	5.919516000	7.918901000	2.604640000
C	7.066150000	7.601605000	1.758314000
O	5.372653000	9.026970000	2.603197000
C	7.355732000	2.944700000	3.507738000
C	7.774134000	2.913454000	4.848473000
C	7.398156000	1.885500000	5.701620000
C	6.580728000	0.854059000	5.221391000
C	6.151997000	0.875994000	3.889451000
C	6.530985000	1.910835000	3.022724000
O	6.258009000	-0.116110000	6.122345000
C	5.427255000	-1.186174000	5.697772000
C	6.034803000	1.906227000	1.594027000
H	11.313650000	1.624797000	0.727946000
H	9.384532000	1.840260000	2.283106000
H	10.704350000	5.700766000	-0.549597000
H	12.359549000	4.271211000	-1.333653000
H	5.657818000	4.815751000	4.134204000
H	4.586699000	7.046990000	4.100653000
H	7.448145000	8.374322000	1.100336000
H	8.406911000	3.713409000	5.223207000
H	7.721704000	1.861416000	6.737118000
H	5.510189000	0.090637000	3.505688000
H	5.890742000	-1.758285000	4.883353000
H	4.441970000	-0.827716000	5.372169000
H	5.305768000	-1.832558000	6.568660000
H	5.363133000	1.061749000	1.413852000
H	6.863180000	1.834397000	0.879166000
H	5.489678000	2.827314000	1.357535000

GEOMETRY for $C_{21}H_{15}0_4$

C	10.830775000	2.575389000	0.844838000
C	11.259871000	3.676894000	-0.024359000
C	9.762782000	2.679582000	1.686339000
C	8.980260000	3.877684000	1.779461000
C	10.459592000	4.884289000	0.082501000
C	9.389683000	4.965613000	0.933456000
O	12.239028000	3.570802000	-0.792749000
C	7.864330000	4.044450000	2.619807000
C	7.164191000	5.264665000	2.631908000
C	7.600739000	6.331895000	1.773140000
O	8.692911000	6.150520000	0.958283000
C	6.015855000	5.541241000	3.445072000
C	5.373385000	6.743491000	3.406415000
C	5.807005000	7.841351000	2.534916000
C	6.972356000	7.547575000	1.719243000
O	5.210750000	8.938639000	2.506244000
C	7.418345000	2.918211000	3.502348000
C	7.937955000	2.801765000	4.800512000
C	7.552647000	1.773475000	5.653065000
C	6.623061000	0.826897000	5.211153000
C	6.094516000	0.926585000	3.921195000
C	6.482728000	1.964167000	3.059992000
O	6.294076000	-0.157034000	6.112017000
C	5.359652000	-1.138818000	5.709545000
C	5.892486000	2.044304000	1.670702000
H	11.413856000	1.659288000	0.787313000
H	9.481016000	1.839876000	2.317080000
H	10.725940000	5.736134000	-0.535180000
H	5.663614000	4.755341000	4.108860000
H	4.504170000	6.936784000	4.030646000
H	7.343386000	8.319460000	1.052425000
H	8.660225000	3.538153000	5.141951000
H	7.956870000	1.686518000	6.656992000
H	5.372277000	0.200752000	3.562469000
H	5.716224000	-1.710637000	4.841366000
H	4.383060000	-0.697718000	5.465443000
H	5.245050000	-1.814111000	6.560943000
H	5.186439000	1.226208000	1.492484000
H	6.676129000	1.995278000	0.906198000
H	5.364766000	2.993044000	1.520964000

THERMAL INFORMATION:

Sum of electronic and thermal Energies = -1110.403560

Sum of electronic and thermal Enthalpies = -1110.402616

Sum of electronic and thermal Free Energies = -1110.475068

THERMAL INFORMATION:

Sum of electronic and thermal Energies = -1109.887383

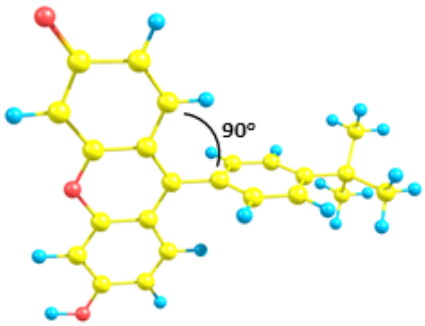
Sum of electronic and thermal Enthalpies = -1109.886439

Sum of electronic and thermal Free Energies = -1109.958874

COMPOUND 6 WITH THE XANTHONE AND C9 ARYL GROUP FORCED TO BE AT 90°

GEOMETRY for $C_{23}H_{20}O_3$

C	-1.656274000	-3.608819000	0.028002000
C	-3.059805000	-3.518776000	-0.004435000
C	-0.897046000	-2.453734000	0.031030000
C	-1.490053000	-1.170638000	-0.005874000
C	-3.685064000	-2.272833000	-0.023972000
C	-2.900972000	-1.120681000	-0.019320000
O	-3.755442000	-4.686595000	-0.007965000
C	-0.751478000	0.076399000	-0.000407000
C	-1.449343000	1.268854000	-0.000377000
C	-2.903573000	1.261600000	0.000075000
O	-3.573010000	0.058634000	-0.018693000
C	-0.825628000	2.577055000	-0.030154000
C	-1.555119000	3.716236000	-0.020502000
C	-3.031339000	3.711259000	0.013948000
C	-3.655020000	2.391835000	0.012310000
O	-3.683168000	4.761033000	0.031291000
C	0.738082000	0.043767000	0.010815000
C	1.469560000	0.021024000	-1.187736000
C	2.860500000	-0.007180000	-1.171984000
C	3.588136000	-0.025081000	0.031328000
C	2.848349000	-0.008464000	1.221127000
C	1.452481000	0.028011000	1.214561000
H	-1.194487000	-4.589717000	0.055988000
H	0.183938000	-2.528244000	0.065531000
H	-4.766754000	-2.172867000	-0.036565000
H	-4.706921000	-4.496702000	-0.026637000
H	0.257700000	2.618084000	-0.066124000
H	-1.080426000	4.692775000	-0.046517000
H	-4.737939000	2.335184000	0.014940000
H	0.942731000	0.040546000	-2.138045000
H	3.354809000	-0.023983000	2.179644000
C	5.126466000	-0.061788000	0.001007000
H	3.386983000	-0.011794000	-2.122334000
C	5.655307000	1.186784000	-0.744627000
C	5.739682000	-0.073469000	1.413835000
C	5.594131000	-1.337891000	-0.738699000
H	0.911890000	0.036312000	2.157023000
H	5.342954000	2.107558000	-0.239081000
H	5.291334000	1.231919000	-1.776385000
H	6.751328000	1.173005000	-0.778701000
H	5.428689000	-0.954188000	1.987245000
H	5.467968000	0.821588000	1.984870000
H	6.832494000	-0.097672000	1.340248000
H	5.225639000	-1.370569000	-1.769259000
H	5.239654000	-2.239983000	-0.227045000
H	6.689510000	-1.376643000	-0.775554000



THERMAL INFORMATION:

Sum of electronic and thermal Energies = -1113.765579

Sum of electronic and thermal Enthalpies = - -1113.764634

Sum of electronic and thermal Free Energies = -1113.836503

Photophysics.

Absorption spectroscopy

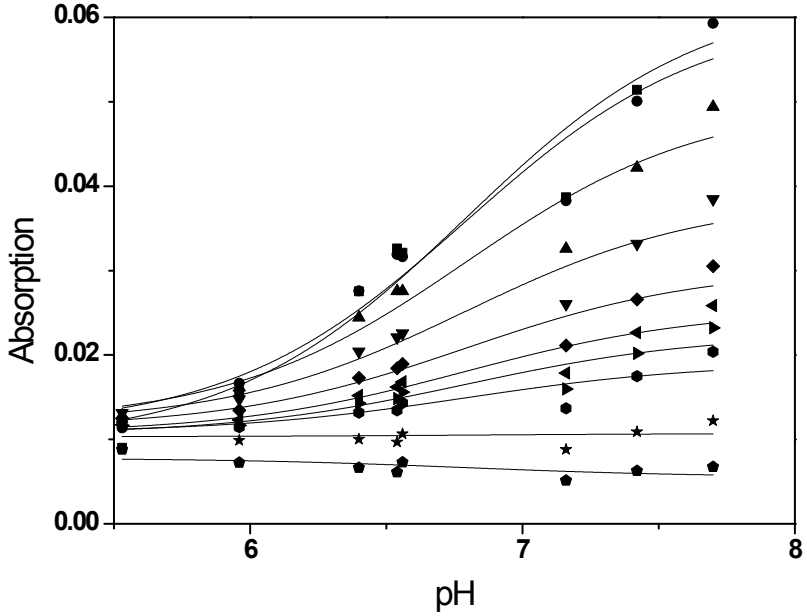


Figure S1. Global fitting of the *A* vs. pH curves of **GG** solutions in phosphate buffer 0.005 M. The wavelengths shown are 500 (■), 495 (●), 490 (▲), 485 (▼), 480(♦), 470 (◄), 465 (►), 460 (⬟), 455 (★) and 450 nm (⬣).

The absorbance *A* of the aqueous solutions of **GG** depends on pH according to Beer's law and the simple acid-base equilibrium theory. To analyze the experimental absorbance vs pH, we implemented a global nonlinear least-squares curve fitting method. Assuming that the neutral/anion system follows Beer's law, at any wavelength λ_{abs} and pH, the absorbance *A* is given by the expression:

$$A(pH, \lambda_{abs}) = C^{GG}\left(\sum_i \alpha_i(pH, pK_a^{app})\,\varepsilon_i(\lambda_{abs})\right)d \qquad (1)$$

where C^{GG} is the total concentration of **GG**, d is the optical path length, $\varepsilon_i(\lambda_{abs})$ is the molar absorption coefficient of the ith prototropic form of **GG** depending on wavelength, and $\alpha_i(\mathrm{pH},\ \mathrm{p}K_a^{app})$ is the fraction of **GG** in the ith prototropic form. The values of α_i depend on both pH and the apparent ground-state acidity constant ($\mathrm{p}K_a^{app}$ value) according to the simple acid-base equilibrium equations.

The nonlinear global fitting of the entire A vs pH vs λ_{abs} surface to eq 1 and the corresponding acid-base equilibrium equations allows the determination of the molar absorption coefficients $\varepsilon_i(\lambda_{abs})$ and $\mathrm{p}K_a^{app}$. In this global fitting, the apparent $\mathrm{p}K_a^{app}$ was a linked parameter over the whole surface, while $\varepsilon_i(\lambda_{abs})$ were locally adjustable parameters at each wavelength for each species. (Figure 5)

Emission Spectroscopy

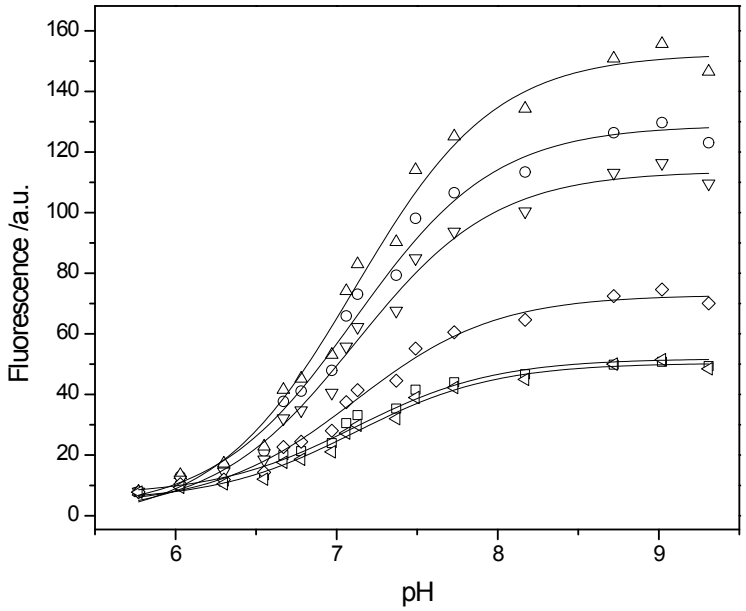


Figure S2. Plot of the experimental steady-state fluorescence(λ_{ex} =485 nm and λ_{em} 500 (■), 510 (○), 520 (Δ), 530 (♦) 540 (❋) and 550nm (❖)) versus pH of 7×10^{-6} M **GG** aqueous solutions at pH values between 5.5 and 9 in phosphate buffer 0.005 M. Equation 2 was fitted to these experimental values. The lines represent the best fitting curves.

Assuming that there is no excited-state proton exchange between neutral and anion in the absence of phosphate buffer, the value of the acidity constant of **GG** was obtained by means the following equation:

$$I = C^{dye} K_a \left[\phi_N \alpha_N \varepsilon_N + \phi_A \alpha_A \varepsilon_A \right] \qquad (2)$$

in which ϕ_N and ϕ_A represent the quantum yield of the neutral and anion forms, respectively. The experimental results of Figure 6 were fitted to equation 2 by non-linear least-squares estimation and very good fit parameters were obtained (r^2 = 0.998), with a pK_a value of 7.26±0.019. ϕ_1 and ϕ_2 were floating values during the fitting.

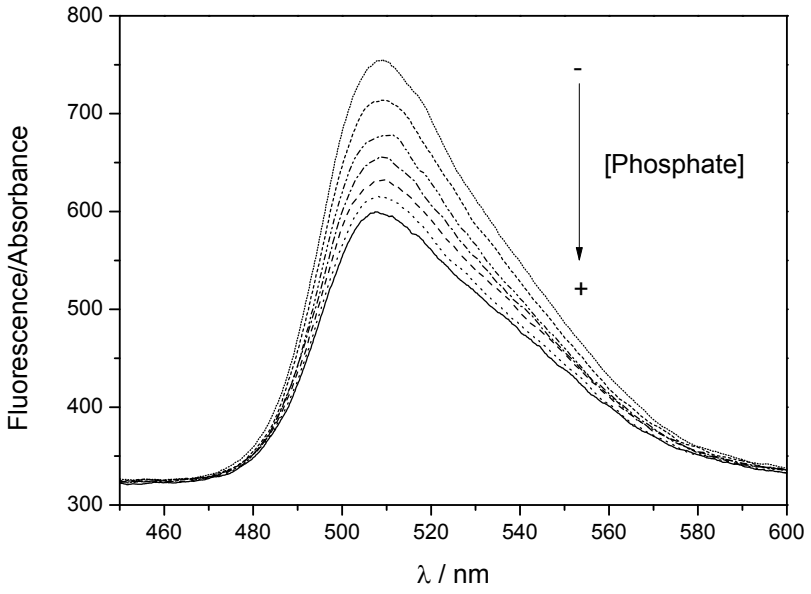


Figure S3. Steady-state emission spectra of **GG** at pH 7.00 and different [Phosphate] (0.05 to 0.40 M).

Quantum yield

Quantum yield values from steady-state fluorescence measurements were calculated for the anion forms using fluorescein in 0.1 M NaOH as a reference (ϕ_{fluo} = 0.95). The quantum yields of the neutral forms were obtained from the fit of equation 1 to the experimental data once obtained the values of $\phi_{A.}$ The recovered quantum yields are 0.97 and 0.10 to anion and neutral form respectively.