

## Supporting Information

for

# Halogen-containing thiazole orange analogues – new fluorogenic DNA stains

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**Characterisation data for the compounds: NMR and GS–MS spectra; validation of the theoretical computations; photostabilities at 532 nm**

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**Figures S1–S13:** characterisation data for the new compounds: copies of  $^1\text{H}$  and  $^{13}\text{C}$  DEPT135 NMR spectra;

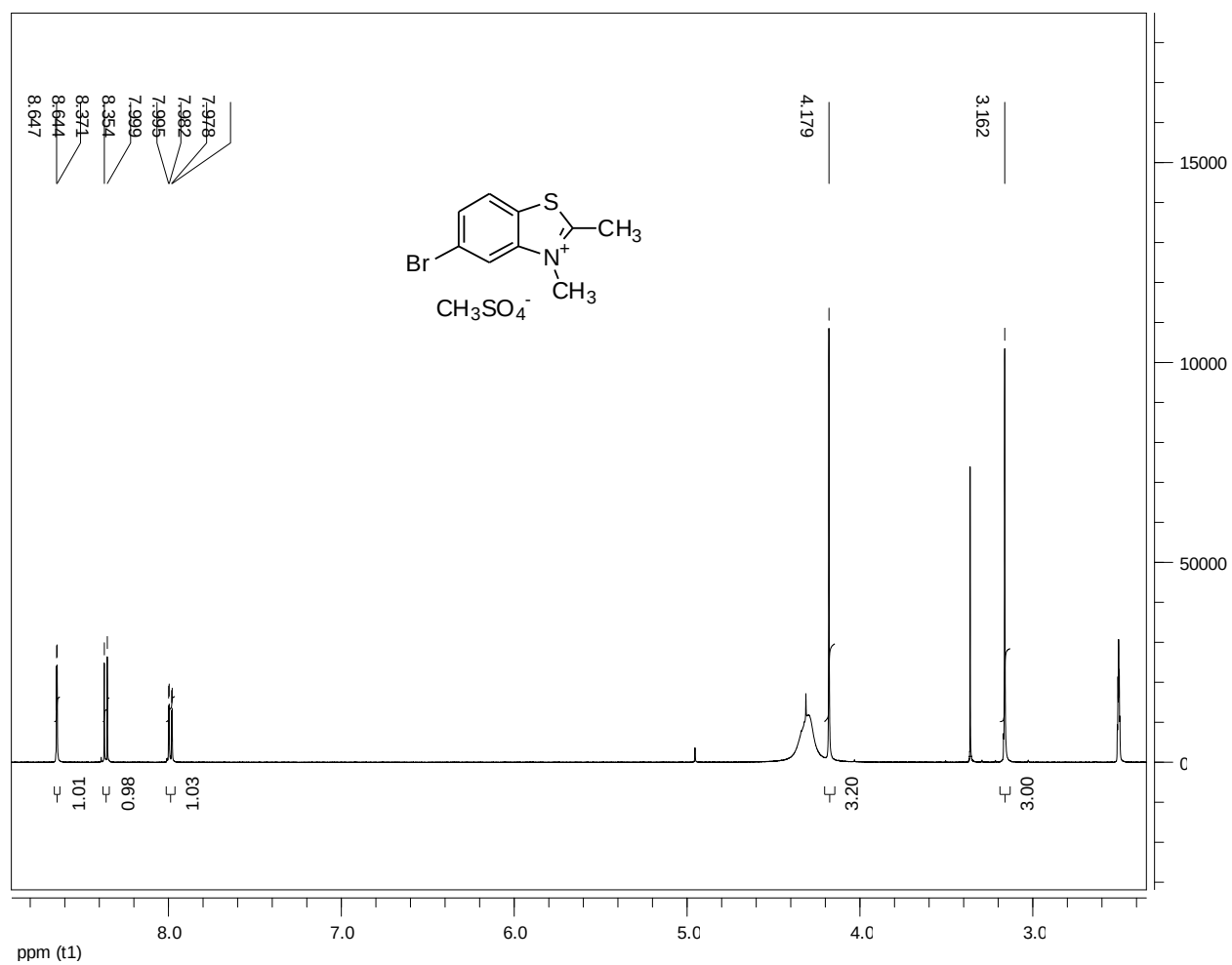
**Figures S14–S18:** copies of GC–MS spectra;

**Figure S19:** superimposed structures of 3-(4-((3-methyl-1,3-benzothiazol-2(3*H*)-ylidene)methyl)-quinolinium-1-yl)propanoate tetrahydrate (CSD entry OVUJUL / CCDC number 739300)[S1] and B3LYP/6-31+G\*\* optimized structure of 3-(4-((3-methyl-1,3-benzothiazol-2(3*H*)-ylidene)methyl)quinolinium-1-yl)propanoate;

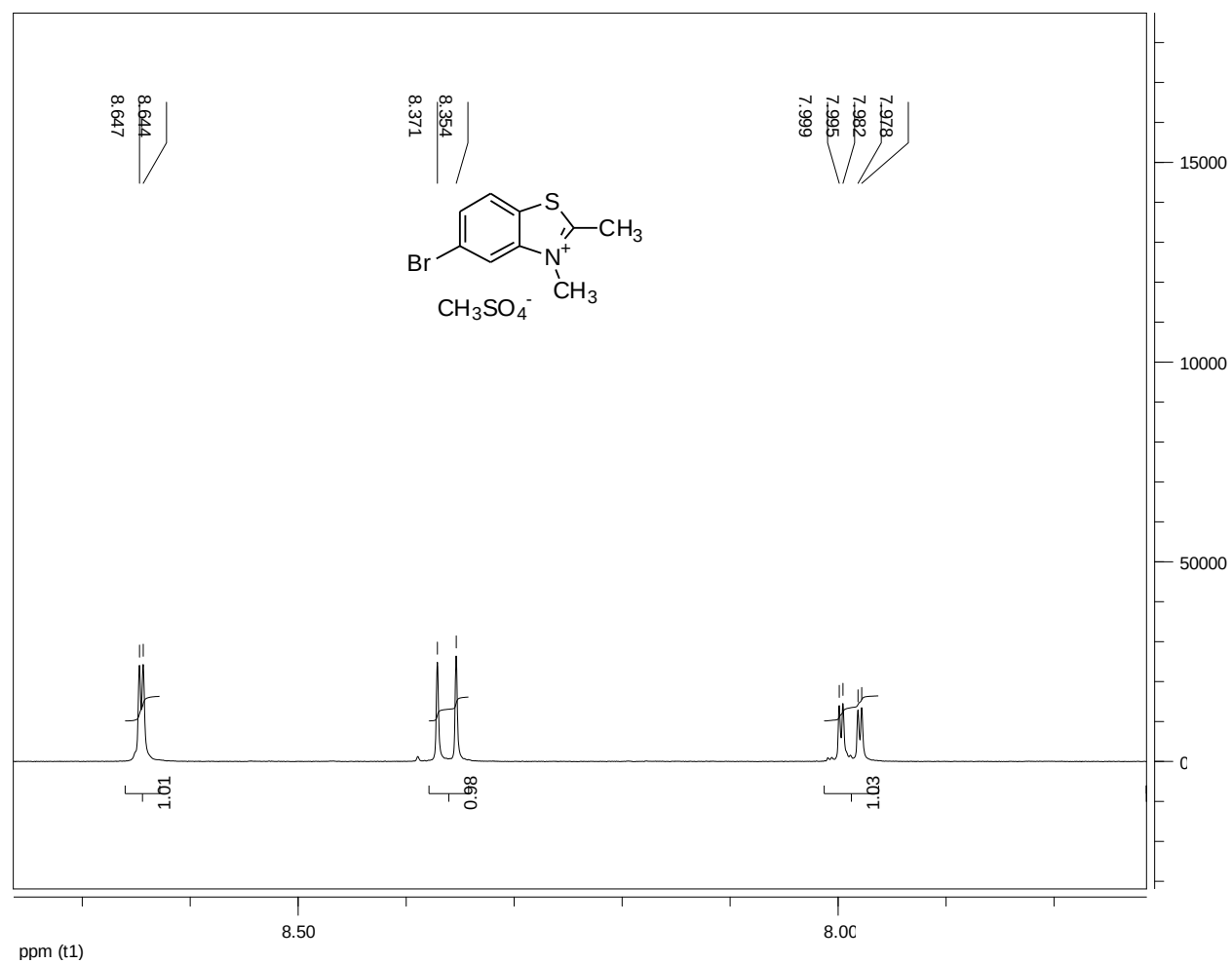
**Figure S20:** labels assigned to selected atoms in 3-(4-((3-methyl-1,3-benzothiazol-2(3*H*)-ylidene)methyl)quinolinium-1-yl)propanoate;

**Table S1:** Experimental structural data (selected bond lengths and angles) from Ref. [S1] compared to the B3LYP/6-31+G\*\* calculated values for the fully optimized structure of 3-(4-((3-methyl-1,3-benzothiazol-2(3*H*)-ylidene)methyl)quinolinium-1-yl)propanoate without crystallization water molecules.

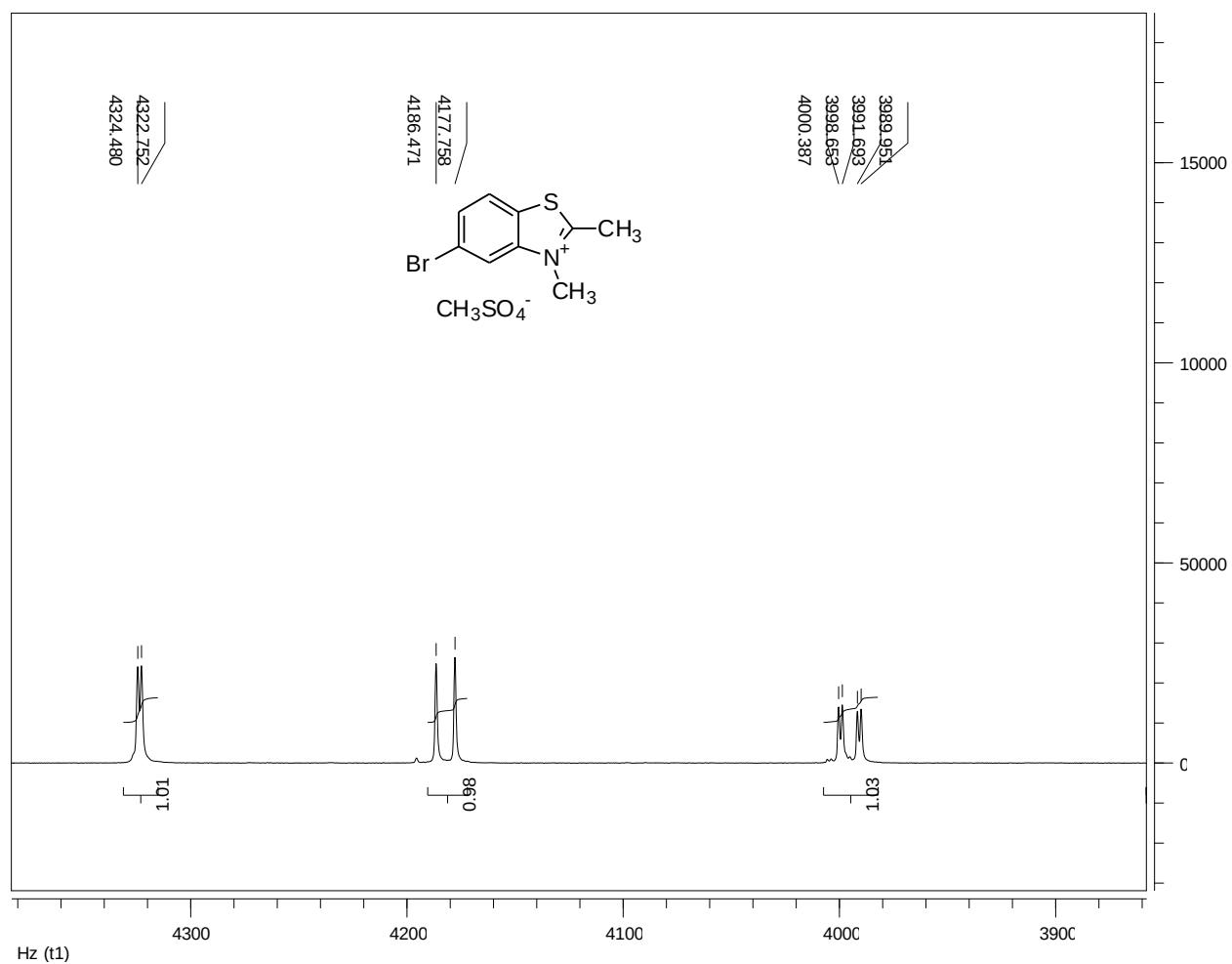
**Figures S21–S24:** Photostabilities of **TO**, **TO-7Cl**, **5a**, **5d** measured by a continuous wave frequency doubled DPSS (diode pumped solid state) - laser, operating at  $\lambda = 532$  nm. The maximal laser power is 80 mW. Via a ND (neutral density) - filter wheel (NDC-100S-4M, Thorlabs Inc.) the laser power can be tuned in broad range. The laser output via couple of cylindrical and spherical lenses is collimated to a nearly rectangular spot with dimensions of 20 mm x 40 mm, thus the whole entrance aperture of the 10 mm square cuvette is covered. The maximal excitation intensity was  $10 \text{ mWcm}^{-2}$ , with a possibility for controllable decrease.



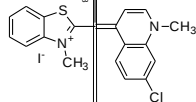
**Figure S1:** <sup>1</sup>H NMR spectrum of **2b**.



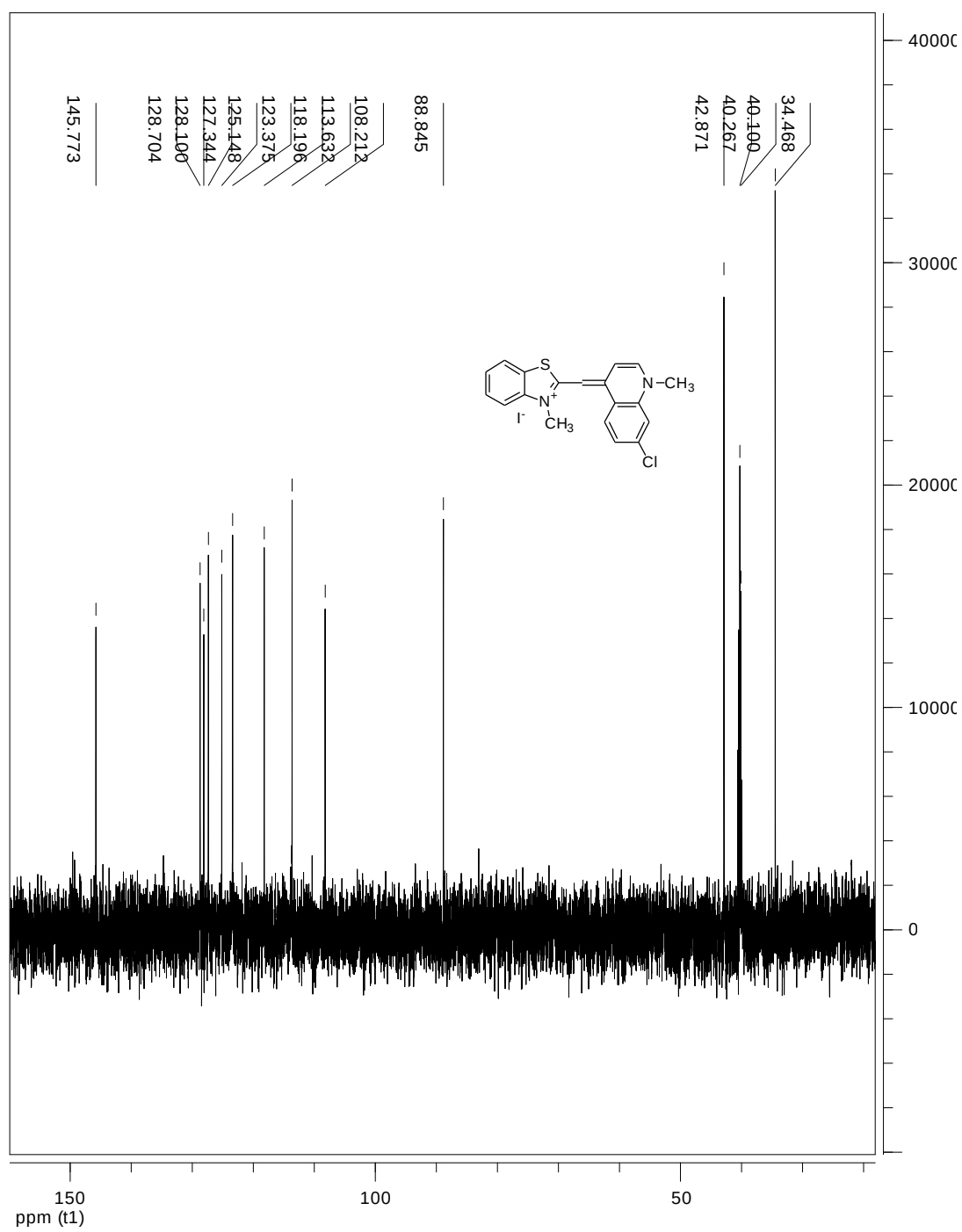
**Figure S2:** <sup>1</sup>H NMR spectrum of **2b** (7.5–9.0 ppm).



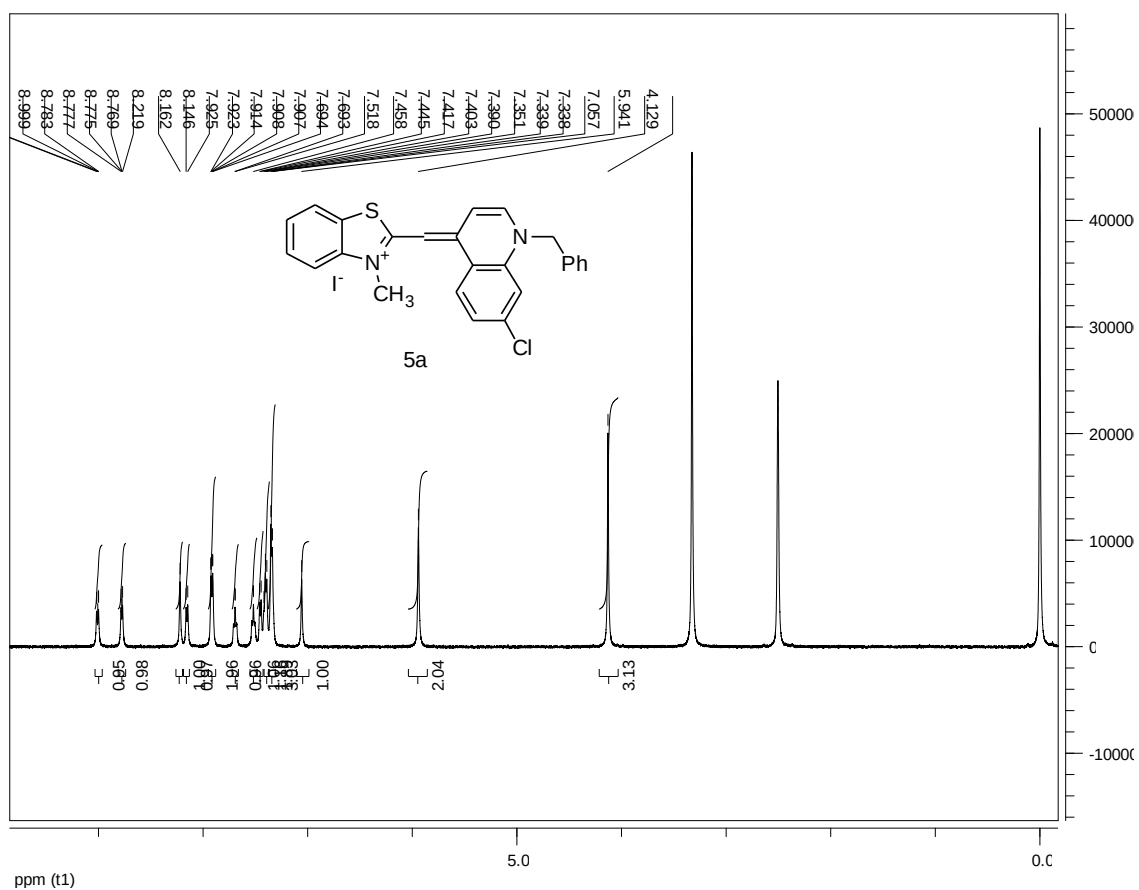
**Figure S3:** <sup>1</sup>H NMR spectrum of **2b** (in Hz) (7.5–9.0 ppm).



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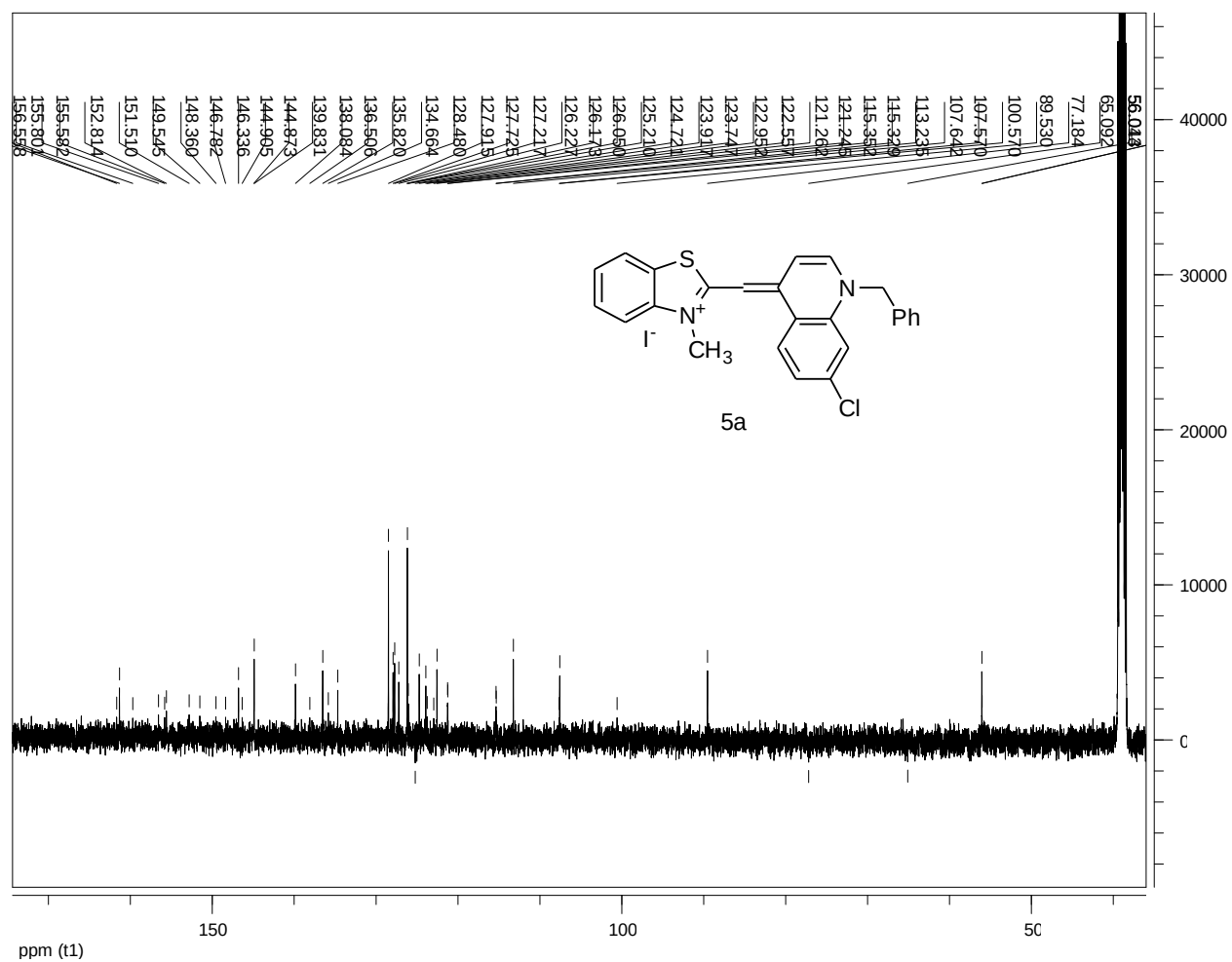


**Figure S5:** DEPT135 spectrum of **TO-7Cl**.

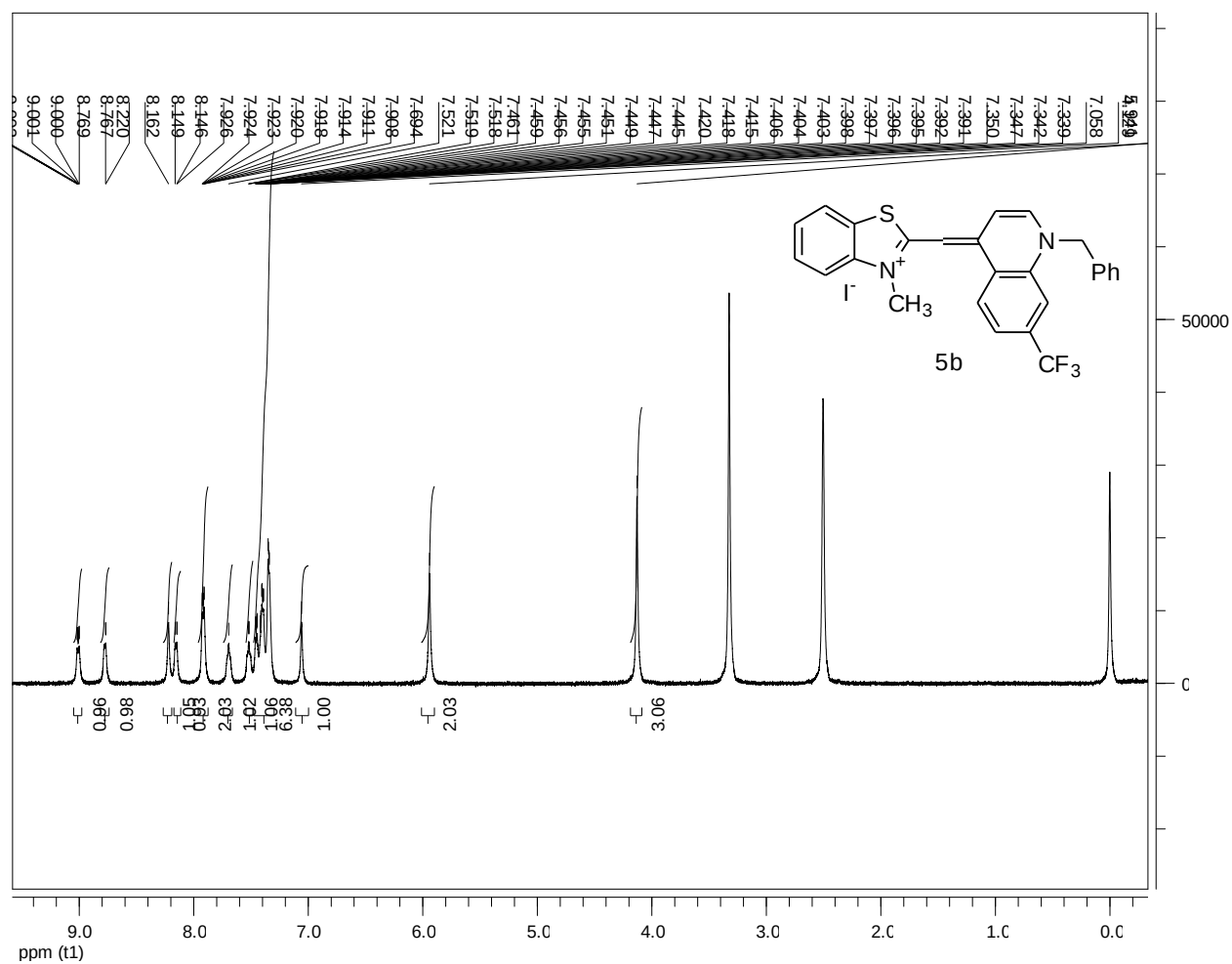


**Figure S6:** <sup>1</sup>H NMR spectrum of **5a**.

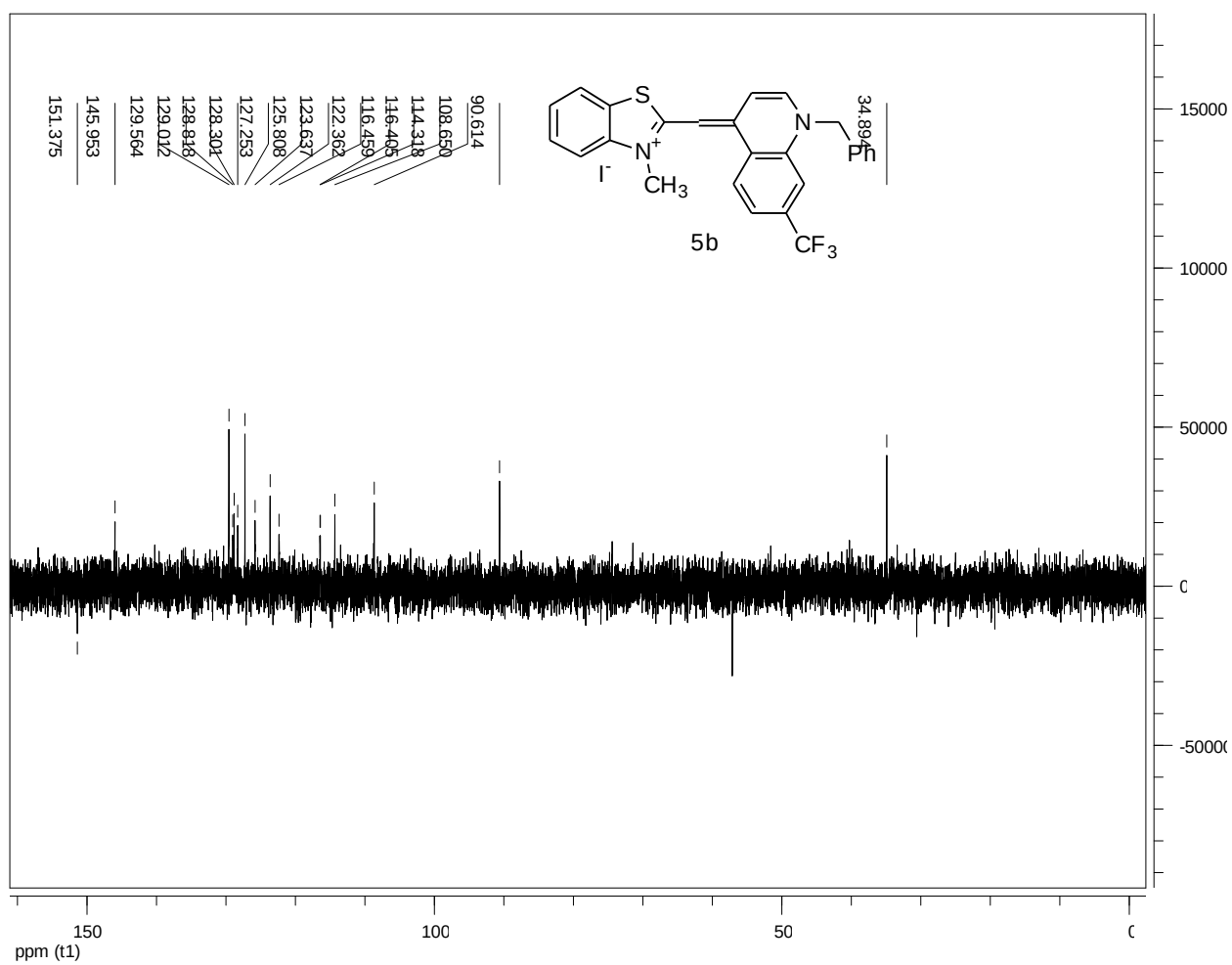




**Figure S7:** DEPT135 spectrum of **5a**.



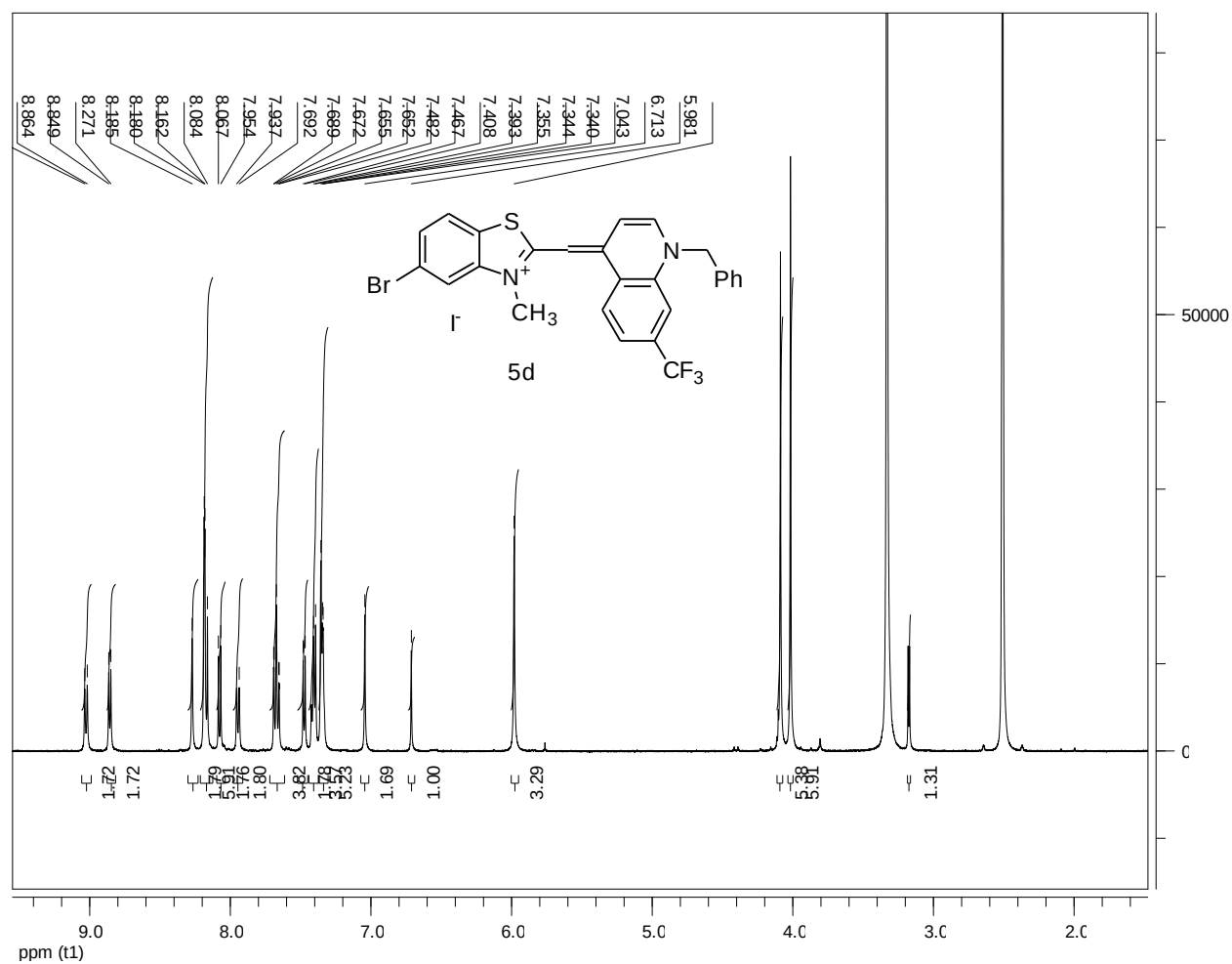
**Figure S8:**  $^1\text{H}$  NMR spectrum of **5b**.



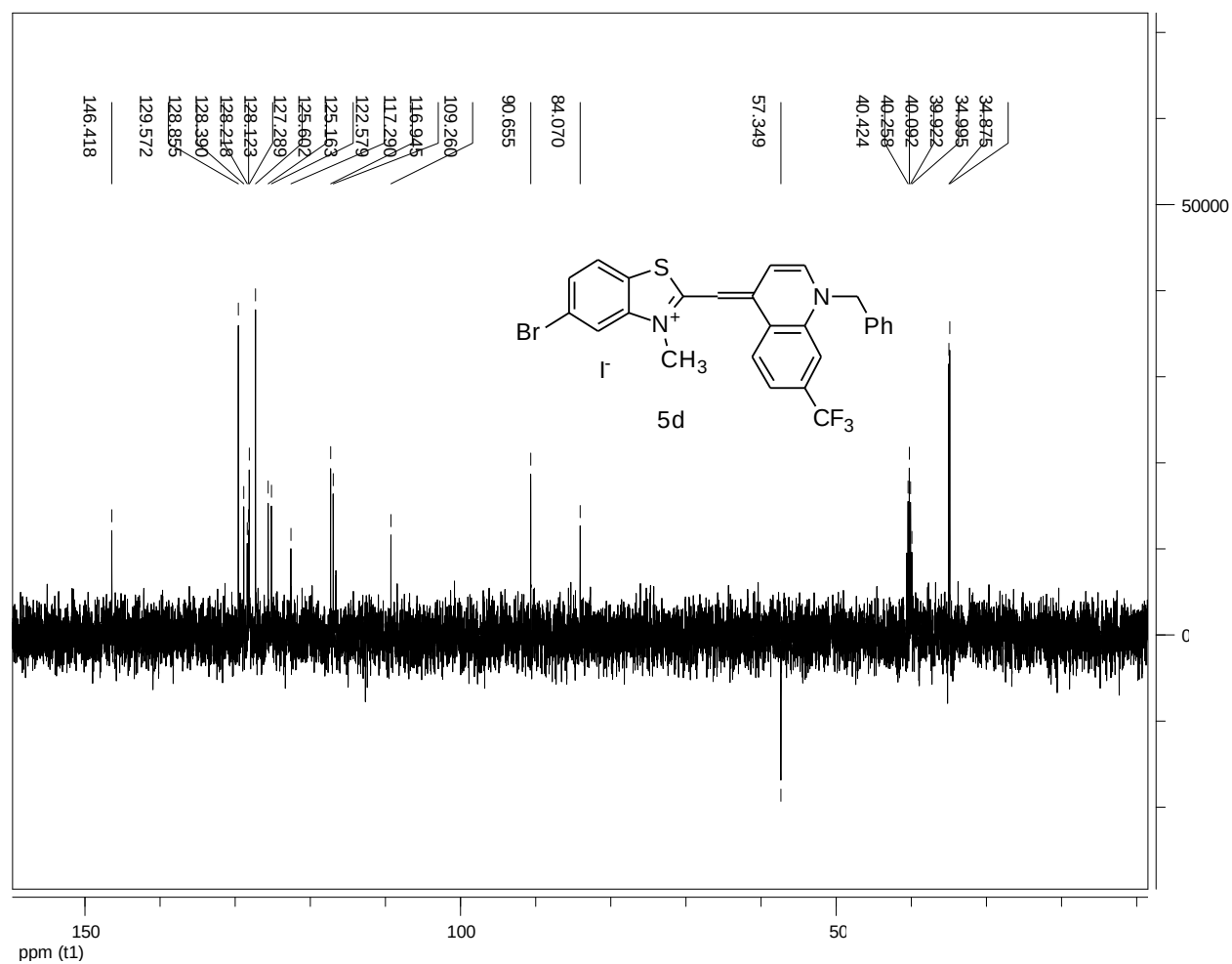
**Figure S9:** DEPT135 spectrum of **5b**.







**Figure S12:**  $^1\text{H}$  NMR spectrum of **5d**.



**Figure S13:** DEPT135 spectrum of **5d**.

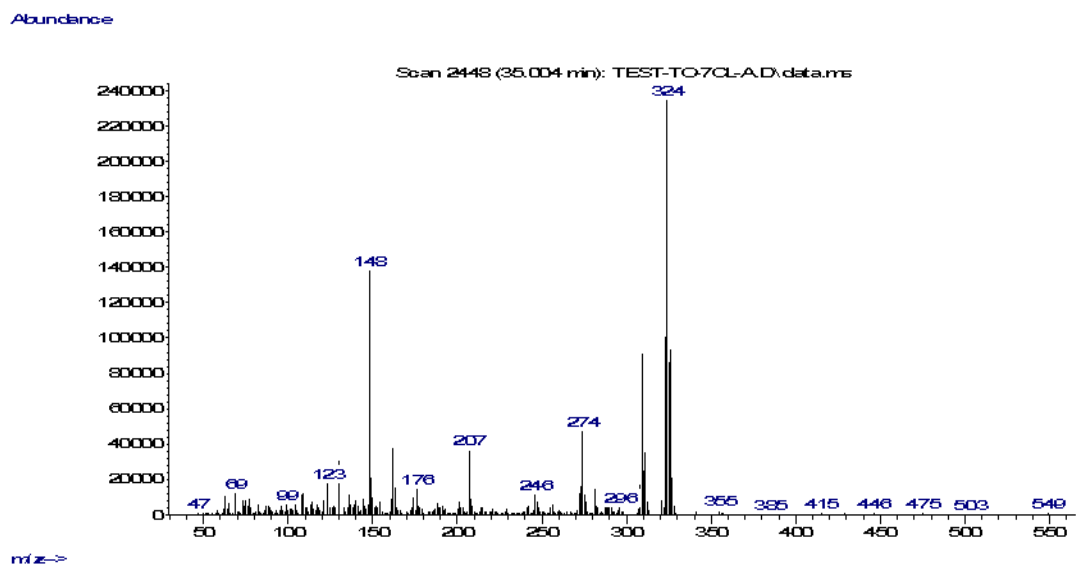


Figure S14: GC-MS spectrum of TO-7Cl.

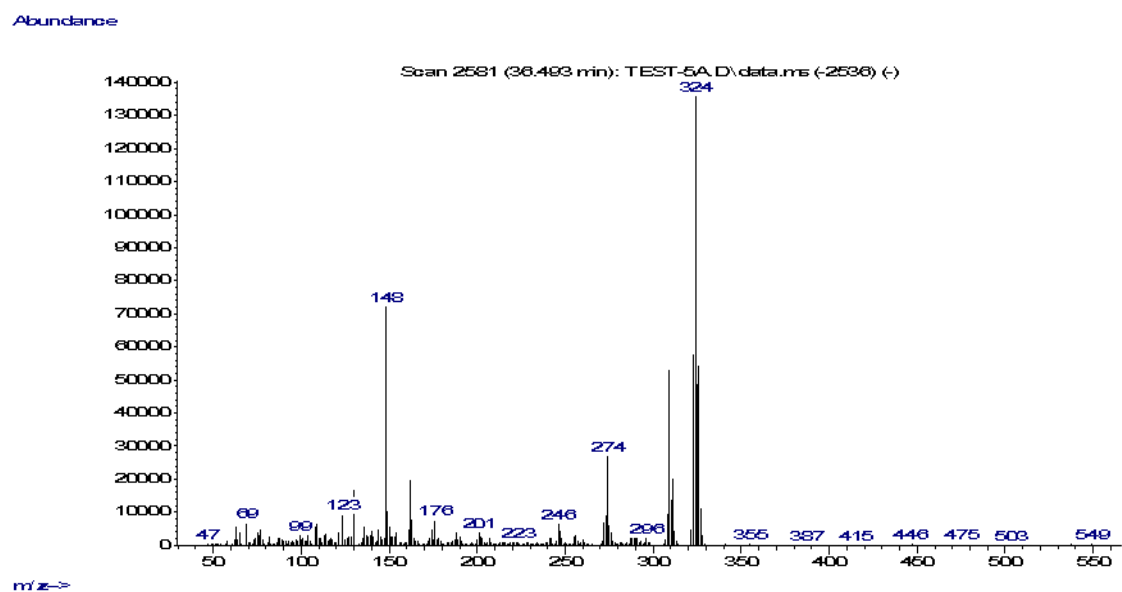


Figure S15: GC-MS spectrum of 5a.



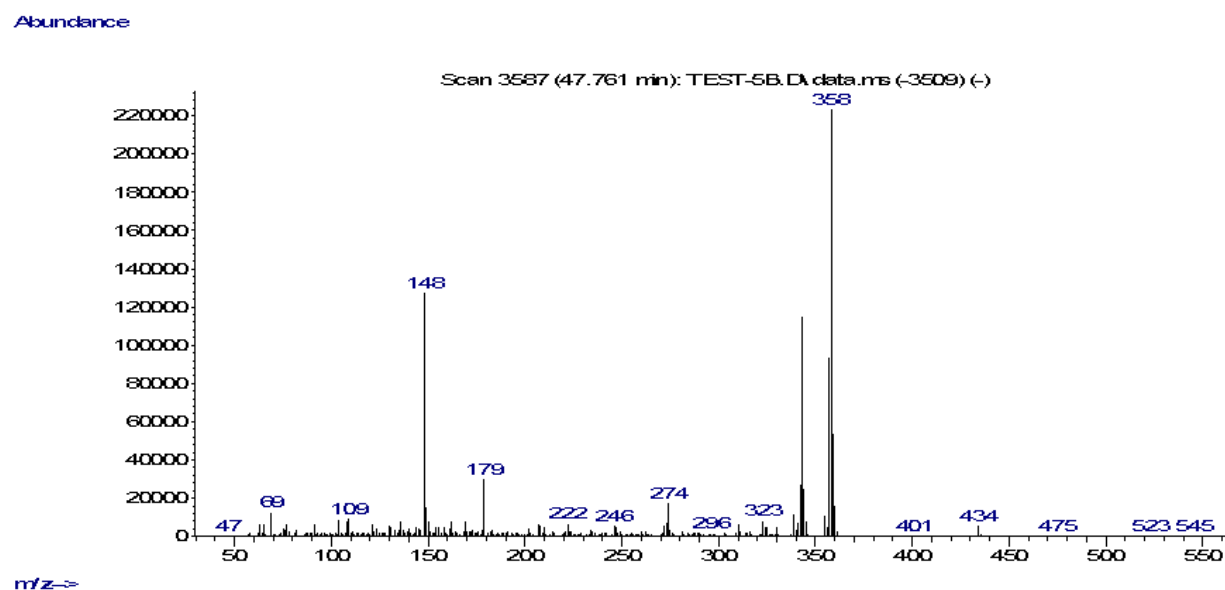


Figure S16: GC-MS spectrum of **5b**.

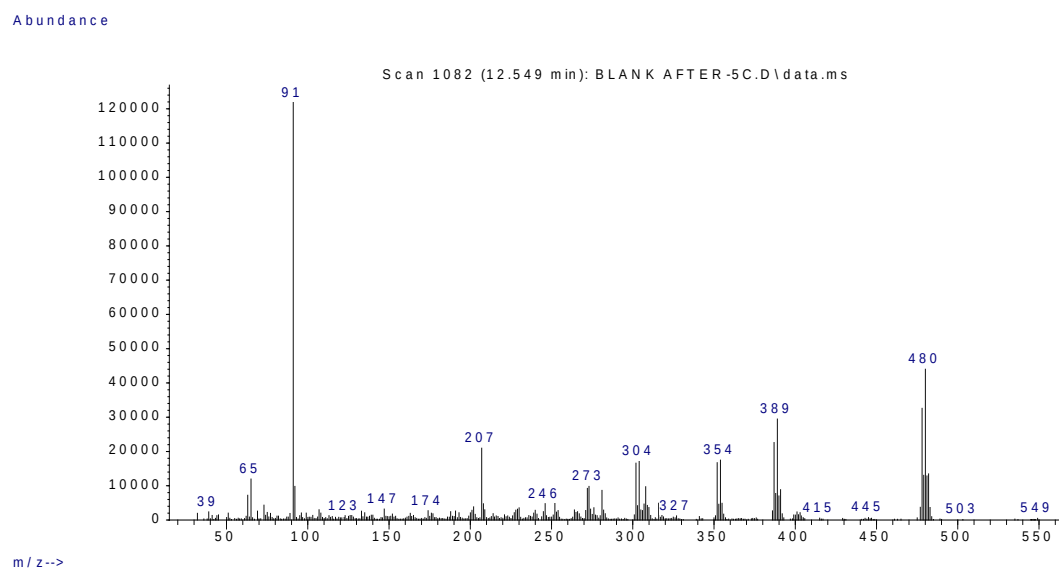
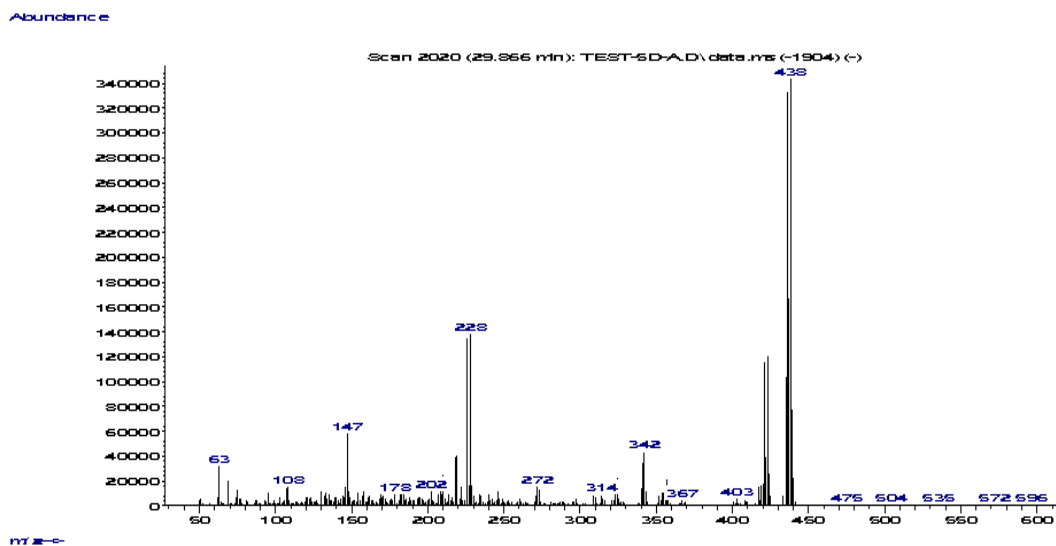
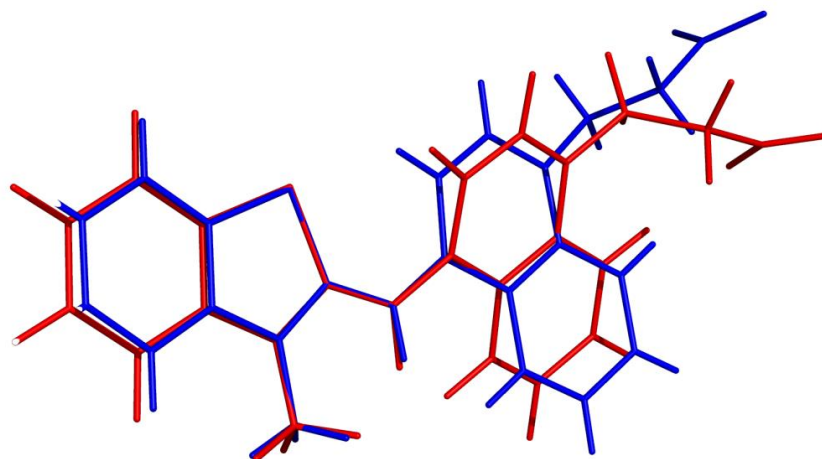


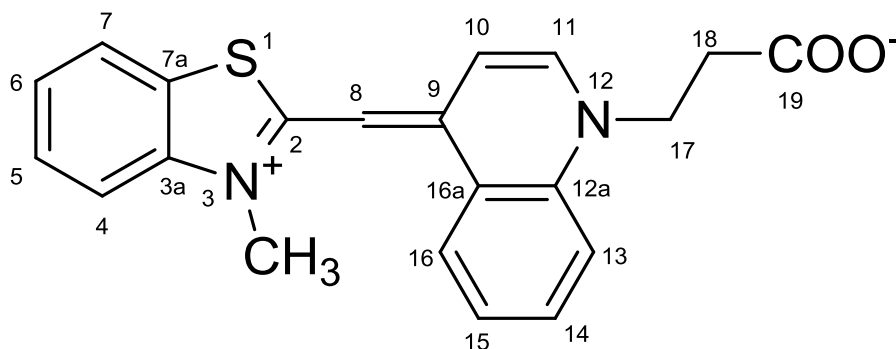
Figure S17: GC-MS spectrum of **5c**.



**Figure S18:** GC-MS spectrum of **5d**.



**Figure S19:** Overlay of 3-(4-((3-methyl-1,3-benzothiazol-2(3H)-ylidene)methyl)quinolinium-1-yl)propanoate tetrahydrate (CSD ENTRY OVUJUL / CCDC number 739300) (blue) and B3LYP/6-31+G\*\* optimized structure of 3-(4-((3-methyl-1,3-benzothiazol-2(3H)-ylidene)methyl)quinolinium-1-yl)propanoate (red).



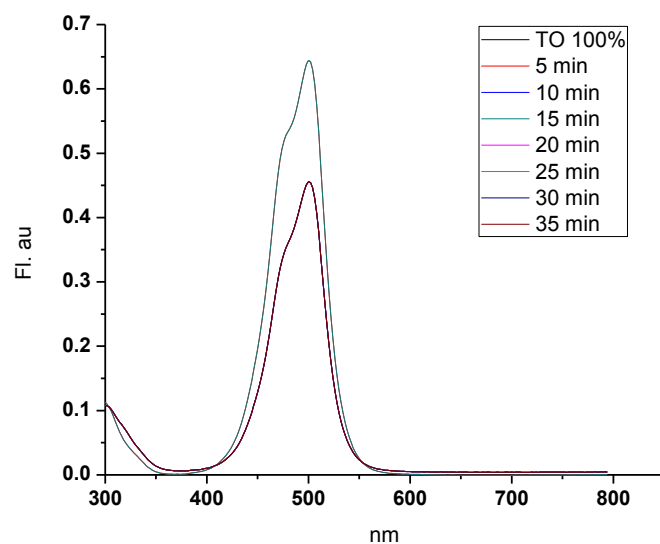
**Figure S20:** Labels assigned to selected atoms in the structure from ref. [S1].

**Table S1:** Experimental structural data (selected bond lengths and angles) from ref. [S1] compared to the B3LYP/6-31+G\*\* calculated values for the fully optimized structure of 3-(4-((3-methyl-1,3-benzothiazol-2(3H)-ylidene)methyl)quinolinium-1-yl)propanoate without crystallization water molecules.

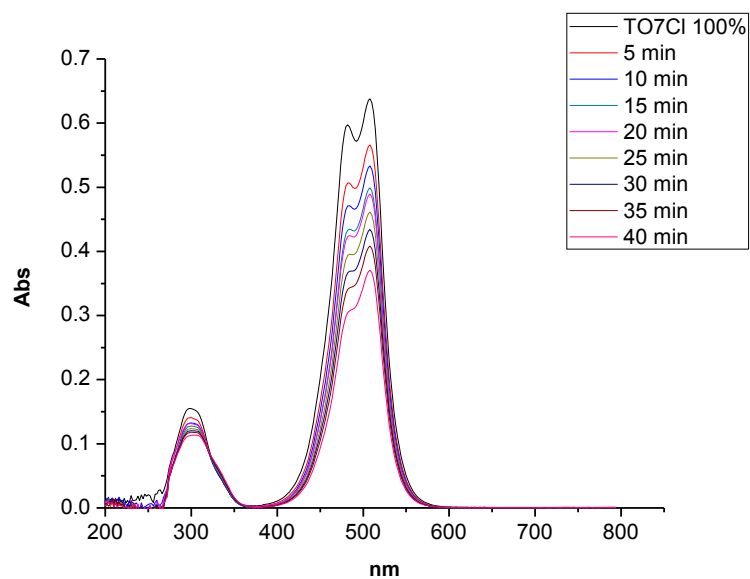
|                           | Calculated | Experimental (ref. [S1]) | Exp. – Calc. |
|---------------------------|------------|--------------------------|--------------|
| <i>Bond distances (Å)</i> |            |                          |              |
| S1-C2                     | 1.77       | 1.74                     | -0.03        |
| C2-N3                     | 1.38       | 1.37                     | -0.01        |
| N3-C3a                    | 1.40       | 1.39                     | -0.01        |
| C3a-C4                    | 1.40       | 1.40                     | 0.00         |
| C4-C5                     | 1.40       | 1.39                     | -0.01        |
| C5-C6                     | 1.40       | 1.39                     | -0.01        |
| C6-C7                     | 1.40       | 1.38                     | -0.02        |
| C7-C7a                    | 1.39       | 1.39                     | 0.00         |
| C7a-S1                    | 1.76       | 1.73                     | -0.03        |
| C2-C8                     | 1.38       | 1.39                     | 0.01         |
| C8-C9                     | 1.43       | 1.40                     | -0.03        |
| C9-C10                    | 1.41       | 1.40                     | 0.00         |
| C10-C11                   | 1.38       | 1.38                     | -0.01        |
| C11-N12                   | 1.34       | 1.34                     | 0.00         |
| N12-C12a                  | 1.39       | 1.39                     | 0.00         |
| C12a-C13                  | 1.41       | 1.40                     | -0.02        |
| C13-C14                   | 1.38       | 1.37                     | -0.01        |
| C14-C15                   | 1.41       | 1.41                     | 0.00         |
| C15-C16                   | 1.38       | 1.35                     | -0.03        |
| C16-C16a                  | 1.42       | 1.42                     | 0.00         |

|                            |      |      |        |
|----------------------------|------|------|--------|
| C16a-C9                    | 1.45 | 1.46 | 0.01   |
| N12-C17                    | 1.49 | 1.50 | 0.00   |
| C17-C18                    | 1.54 | 1.50 | -0.04  |
| C18-C19                    | 1.58 | 1.54 | -0.05  |
| <i>Dihedral angles (°)</i> |      |      |        |
| S1-C2-C8-C9                | 7.9  | 0.6  | -7.30  |
| C2-C8-C9-C10               | 19.2 | 0.7  | -18.50 |

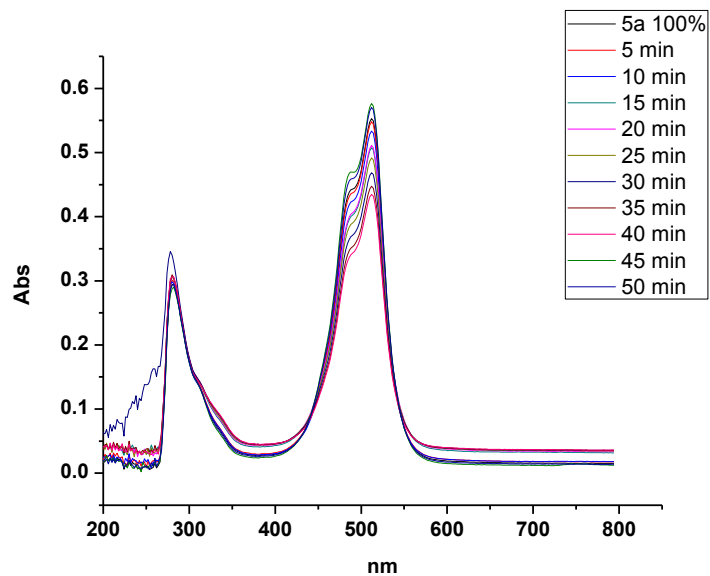
[S1]. Fei, X., Gu, Y., Lan, Y. et al., J. Chem. Crystallogr. (2011) 41: 1232.  
<https://doi.org/10.1007/s10870-011-0080-0>, Synthesis and Crystal Structure of Thiazole Orange Derivative



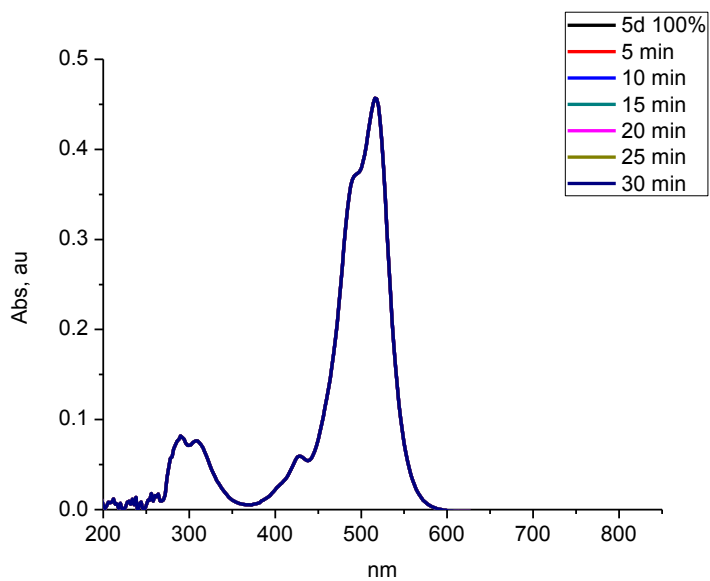
**Figure S21:** Photostability of **TO** measured by a continuous wave frequency doubled DPSS laser, operating at  $\lambda = 532$  nm.



**Figure S22:** Photostability of **TO-7CI** measured by a continuous wave frequency doubled DPSS laser, operating at  $\lambda = 532$  nm.



**Figure S23:** Photostability of **5a** measured by a continuous wave frequency doubled DPSS laser, operating at  $\lambda = 532$  nm.



**Figure S24:** Photostability of **5d** measured by a continuous wave frequency doubled DPSS laser, operating at  $\lambda = 532$  nm.