## Beyond Optical Imaging: Development of Acoustogenic Probes for Deep-Tissue Analyte Detection

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Fluorescence imaging is a privileged optical method for studying biological systems due to its non-invasiveness and high spatial-temporal resolution. In particular, reaction-based small-molecule fluorescent probes have become indispensible tools in modern biology because of their rapid response, high sensitivity and excellent selectivity towards various analytes such as metal ions, reactive oxygen and nitrogen species, as well as countless enzymatic activities. Despite their utility, imaging is typically performed at the cellular level owning to poor resolution at imaging depths greater than 1 mm. Imaging beyond this limit is challenging for optical methods owing to light scattering. Photoacoustic tomography (PAT), on the other hand, can image biological events beyond 7 cm because reconstructed images can be obtained through the detection of ultrasound, rather than light, which scatters 3 orders of magnitude less, making it an attractive method for in vivo deep-tissue imaging. Nonetheless, reaction-based small-molecule photoacoustic probes that are able to detect a target analyte with fast kinetics, high sensitivity and excellent selectivity have not been reported. To this end, we have developed and evaluated two acoustogenic small-molecule probes to address this unmet need. APC-1 and APC-2 (Acoustogenic Probe for Copper-1 and -2, Figure 1a) are based on the Aza-BODIPY scaffold and feature a picolinic ester reactive group that can be chemoselectively removed via Cu(II)-mediated hydrolysis to elicit a robust photoacoustic signal enhancement (Figure 1b). We envision that these first-generation acoustogenic probes will serve as powerful tools for the in vivo molecular imaging of Cu(II) as well as other biologically relevant analytes.

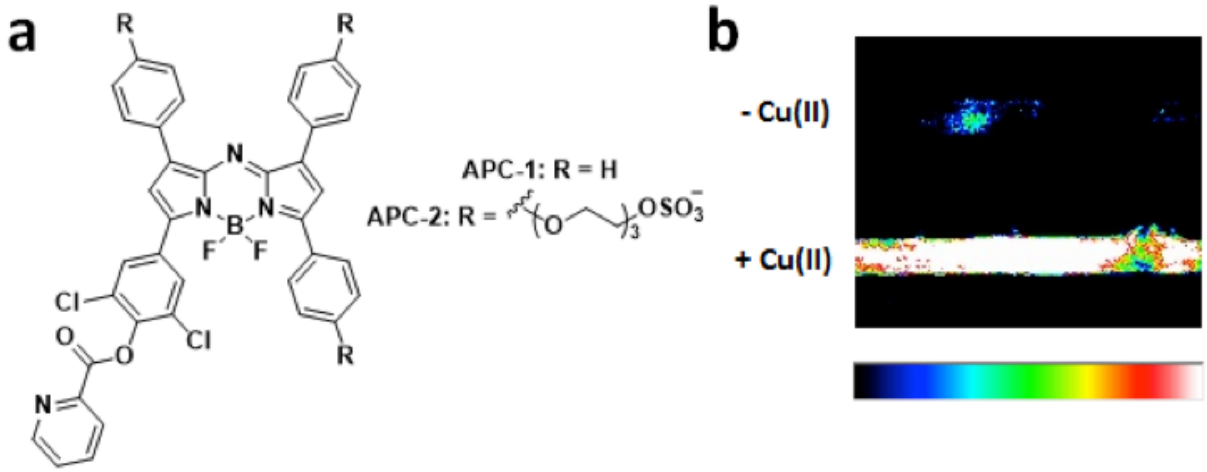


**Figure 1:** a) Chemical structures of APC-1 and APC-2; b) Photoacoustic images acquired from samples of APC-1 treated with a vehicle control or 50 μM Cu(II).