Development of ALDH1A1-Responsive Turn-on Fluorescent Probe for High-Contrast Cancer Stem Cell Imaging

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Cancer stem cells (CSCs) have been proposed to be responsible for cancer proliferation, metastasis, and therapeutic resistance.[1] The development of new fluorescent probe that can visualize and detect CSCs is important for evaluating cancer malignancy. Although aldehyde dehydrogenase-1A1 (ALDH1A1) is known as a reliable biomarker of CSC,[2] only a few ALDH1A1-responsive probes have been reported so far.[3] Recently, we have developed ALDH1A1-responsive turn-on fluorescent probe **CyA4** based on a near infrared (NIR) cyanine dye bearing a formyl group (Figure 1a). Upon treatment with ALDH1A1, the calboxylate-assisted ring-opening of 1,3-thiazinane in **CyC4-C** smoothly took place, and then open-ring form **CyC4-O** emitted fluorescence. In this case, however, the contrast between CSCs and normal cancer cells (NCCs) was low because the open-ring form **CyA4-O** also emitted inherent background-fluorescence.

Here, we report a new probe **CyA4-1** bearing a methyl group in the 1,3-thiazinane ring to improve CSC/NCC contrast (Figure 1b). The introduction of the sterically demanding methyl group prohibited the ring-opening of 1,3-thiazinane. Thus **CyA4-1** hardly emit fluorescence before ALDH1A1 treatment. The ratio of fluorescence intensity before/after treating ALDH1A1 with **CyA4-1** was 10.1, whereas that of **CyA4** was 3.0 (Figure 1c).

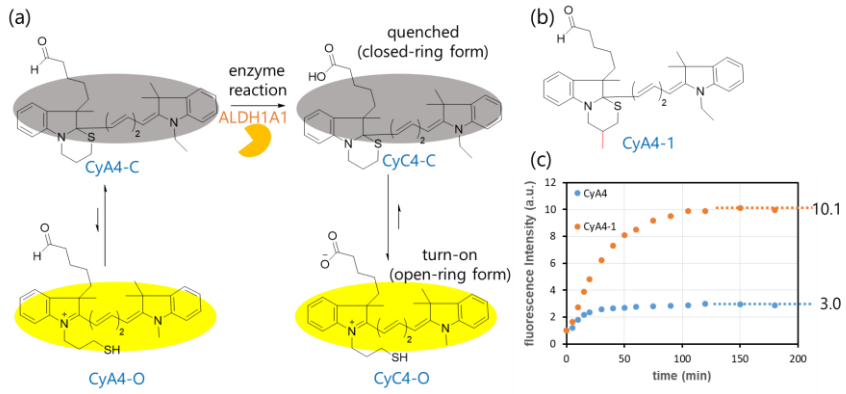

Figure 1. (a) ALDH1A1-responsive **CyA4** and its transformation after treatment of ALDH1A1. (b) Structure of **CyA4-1**. (c) Time-dependent fluorescence intensity change at 620 nm upon treatment with ALDH1A1 50 nM.

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