

Expanding the detectable pH range of DNA nanostructure-based fluorescent pH sensors

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Cellular processes are mediated through intertwined complex chemical and physical interactions of biological molecules. Therefore, analytical devices that simultaneously monitor the change of the concentration and the location of biologically important molecules, and the change of cellular environment indicated by various physical parameters will give a comprehensive understanding of molecular mechanisms underlying of various cellular processes.

Compared to the conventional sensors comprised of a single sensing molecule, assembling several kinds of fluorescent sensors on a DNA scaffold has advantages in monitoring different targets at the same time within the same nanospace by combining individual functions of fluorescent sensors. A single DNA scaffold allows the assembly of multiple fluorophores, with different responses towards pH changes, to be localized in a defined space, enabling ratiometric detection and the expansion of the detectable pH range.

We have developed a ratiometric pH sensor by constructing a DNA nanostructure labeled by two types of fluorophores based on a DNA origami method¹. The combination of a pH sensitive fluorophore fluorescein (CF) with pK_a of 6.5² and tetramethylrhodamine (CR), a pH insensitive fluorophore under the physiological conditions, assembled on a DNA origami scaffold had resulted in a reliable ratiometric pH detection in test tube. The sensor was successfully applied for real-time sensing of cellular pH changes throughout its internalization by its fluorescence signal variation under microscopic conditions.³ In this study, Oregon Green 488 (OG) with pK_a of 4.8² was coassembled with CF and CR on a DNA origami scaffold. The pH monitoring was carried out in a test tube by the ratiometric fluorescent pH sensor to successfully realize an expanded detectable pH range.

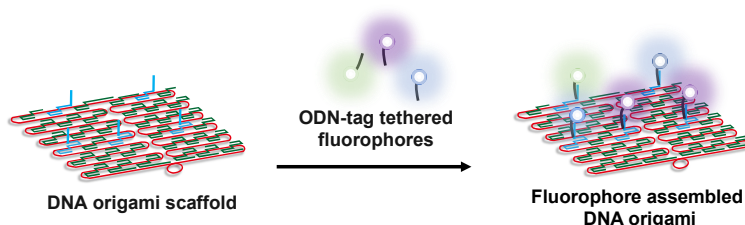


Fig. Illustration of fluorophores assembled DNA origami scaffold for intracellular pH monitoring

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