Genetic alphabet expansion by an unnatural base pair system toward diagnostic and therapeutic applications using xeno-nucleic acids

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Standard nucleic acids comprise four different nucleotide components bearing each of A, G, C or T(U)base as a genetic alphabet. Nucleic acids can be replicated themselves through the complementary base pairings of A–T(U) and G–C, and act as functional molecules, such as catalysts and ligands. However, their functionality is restricted by the limited number of the components, as compared with the 20 standard amino acid components of proteins. If we could expand the genetic alphabet by artificially creating a new unnatural base pair, these extra base components in xeno-nucleic acids might augment their functions.

Recently, several groups reported different types of unnatural base pairs that function as a third base pair in replication and transcription. Among them, we developed a hydrophobic unnatural base pair between 7-(2-thienyl)imidazo[4,5-*b*]pyridine (Ds) and a diol-modified 2-nitro-4-propynylpyrrole (Px) (Kimoto, M. et al., 2009). Chemically synthesized DNA fragments containing the Ds–Px pair are amplified ~10^{28}-fold by PCR corresponding to 100 cycles, and more than 97% of the Ds−Px pairs survived at the initial positions in the amplified DNA (Yamashige, R. et al., 2012).

We applied the Ds–Px pair system to generating Ds-containing DNA aptamers that specifically bind to targets. Then, we found that a few hydrophobic Ds bases efficiently increased the affinity of the DNA aptamers to target proteins, achieving significantly higher affinities than those of the conventional DNA aptamers (Kimoto, M. et al., 2013). Furthermore, the aptamers that we obtained can be stabilized against nucleases by modifying using a mini-hairpin technology that we previously developed (Hirao, I. et al., 1994). Here, I will talk about the unnatural-base DNA aptamer generation for their application to diagnostics and therapeutics.

References:

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