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Installation of orthogonality to the interface that assembles two modular domains in the *Tetrahymena* group I ribozyme

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Two modular elements (P5abc and ΔP5) in the *Tetrahymena* group I ribozyme can be separated physically to generate a two-piece ribozyme derivative consisting of a separately prepared P5abc (P5 RNA) and the rest of the intron (ΔP5 RNA) [1]. Molecular recognition in the interface assembling P5 RNA and ΔP5 RNA is strong and specific, and the catalytic ability of the two-piece ribozyme is comparable to that of the parent unimolecular ribozyme [2].

In this study, we designed alternative tertiary interactions participating in the assembly of P5 and ΔP5. Using alternative interactions, orthogonality was successfully introduced into the P5/ΔP5 assembly interface. The artificial P5/ΔP5 assembly interface would expand the utility of the bimolecular complex as a structural platform for RNA nanotechnology and RNA synthetic biology [3].

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