

## **PROGRAMMABLE DNA-GUIDED ARTIFICIAL RESTRICTION ENZYMES: DISCOVERY, ENGINEERING, AND APPLICATIONS**

Huimin Zhao, University of Illinois at Urbana-Champaign  
zhao5@illinois.edu

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Restriction enzymes are essential tools for recombinant DNA technology that have revolutionized modern biological research. However, they have limited sequence specificity and availability. To address these limitations, we recently developed a *Pyrococcus furiosus* Argonaute (*PfAgo*) based platform for generating artificial restriction enzymes (AREs) capable of recognizing and cleaving DNA sequences at virtually any arbitrary site and generating defined sticky ends of varying length. We demonstrated the utility of AREs in DNA profiling and PCR-based gene cloning. In addition, we are developing a new versatile and flexible strategy based on AREs for assembly of large DNA molecules. This strategy can be readily implemented in our biofoundry for automated DNA assembly of pathways, plasmids, and genetic circuits, which will accelerate the development of new synthetic biology applications.