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Zutao YU

Artificial Assemblies with Cooperative DNA Recognition

Doctoral Thesis accepted by
Kyoto University, Kyoto, Japan

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This book is lovingly and sincerely dedicated to
My love and spiritual partner
Dr. Shiqi Ye
Who is always supporting, helping, and standing by me.

Supervisor's Foreword

Synthetic DNA binder is a long-lasting attractive topic, but few progresses achieved. In this thesis by Dr. Zutao YU, the author makes an endeavor to understand the crucial limitations of synthetic DNA binding systems and construct novel cooperative DNA binding assemblies.

In Chap. 1, the author systematically reviews the engineering of synthetic PIPs-based DNA binding assembly. The author first separately describes the design of three structural domains (DBD, CID, and FD) and then summarizes the representative artificial assemblies from a combination of these domains. The author also points out the long-standing limitations of PIPs and provides thoughtful comments for future study.

Chapters 2 and 3 introduce two exemplary cooperative DNA binding systems PIP-HoGu and PIP-NaCo, respectively, in order to overcome the issues of off-target rate and un-flexible binding mode. The CID assures the synergistic binding of two PIPs at the nearby DNA sites and with a flexible gap distance between two PIPs. PIP-NaCo system utilizes left-handed PNA as CID that exhibits orthogonality to cellular nucleic acids and tunability of cooperation. In the in vitro and cell study, both PIP-HoGu and PIP-NaCo systems demonstrate advancement in sequence selectivity and flexible binding.

In Chap. 4, the author takes a brave step forward to construct an advanced genetic switch termed ePIP-HoGu, by installing epigenetic functionality into cooperative DNA binding system and demonstrates its effectiveness in an in vitro HAT assay.

This thesis clarifies the engineering of synthetic DNA binding systems and advanced genetic switches. Significantly, the sequential development of cooperative DNA binding system and epigenetic advanced DNA binding system will fast track the exploration and optimization of the biological study of synthetic DNA binding systems.

Kyoto, Japan
February 2020

Prof. Hiroshi Sugiyama

Part of this thesis has been published in the following journal articles:

1. **YU Z**, Ai M, … Pandian G.N*, Isaacs L*, Sugiyama H*. Advanced DNA binding system mimicking the cooperative function of transcription factor pairs precisely recruits the epigenetic modifiers to the DNA repeat binding sites. ***Chem. Comm***., **2020**, 56, 2296.
2. **YU Z**, Pandian G.N*, Hidaka T, Sugiyama H*. Pyrrole-imidazole polyamides as regulators of gene expression. ***Adv. Drug Deliv. Rev.***, **2019**, 147, 66.
3. **YU Z**, Hsieh W, Asamitsu S, Hashiya K, Bando T, Ly D.H*, Sugiyama H*. Orthogonal left-hand PNA empower DNA binders with cooperative DNA recognition capable of mimicking transcription factor pairs. ***Chem. Eur. J.***, **2018**, 24, 14183.
4. **YU Z**, Guo C, Wei Y, Hashiya K, Bando T, Sugiyama H*. Pip-HoGu: An Artificial Assembly with Cooperative DNA Recognition Capable of Mimicking Transcription Factor Pairs. ***J. Am. Chem. Soc.***, **2018**, 140, 2426.

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