

The Effect of Holliday Junction Sequence and Isomeric Form on the Self-assembly of Rationally Designed DNA Crystals

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The majority of protein crystal structures to date have been determined via X-ray crystallography; however, producing high resolution crystals has proven to be a major obstacle for structural analysis of hundreds of proteins which rely upon arbitrary crystallization screens with severely limited predictability. Consistent with the fundamental goal of DNA nanotechnology, we have sought to rationally design 3D DNA crystals a priori, with a fully addressable interconnected network of helical arrays that can immobilize guest molecules at specified positions, to allow their structure to be solved without having to find suitable crystallization conditions. DNA nanotechnology has emerged as a prominent field that uses DNA as a molecular scaffold for programmable self-assembly of 3D crystalline arrays. The central building block of these systems is an immobile Holliday junction (HJ), a branched nucleic acid motif inspired by the structure formed during homologous recombination. DNA-directed crystal design introduces single-stranded overhangs ("sticky ends") into these branched four arm junctions, allowing them to self-assemble sequence specifically via canonical Watson-Crick base pairing. Our laboratory has recently determined that junction sequence of the DNA crystals affects their crystallization capability, structure, and symmetry. In addition, we have probed the effect of the alternative isomer (IsoII) of 36 unique HJ sequences, and thus far, we show that IsoII can rescue junction sequences previously deemed "fatal" for crystallization and affect crystal symmetry. Lastly, we demonstrate the ability to scaffold small molecules within the minor grooves of the DNA lattice to aid in the determination of their structures. Taken together, this work will inform the improved design and crystallization of self-assembled DNA crystals to host larger biomolecules within the molecular framework for de novo structure solution.

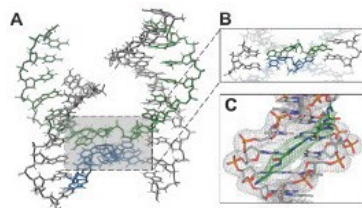


Figure 1. Holliday junction sequence determines crystallization and programmably scaffolds materials

References

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