

Daisy Black Italian Hilo Fantasia Model



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The long-term objective of this project is to understand how proteins organize into complexes in cells. As complex assemblies control the activity of cells, their detailed structure and assembly must be known to construct a molecular map of how they function. Structural analyses of nucleic acids and proteins have been limited to short interactions of relatively small populations of molecules because of the tremendous complexity and heterogeneity of natural complex assemblies. Thus, to advance knowledge about the molecular structure and assembly of complexes, the goal of this project is to develop synthetic complex assemblies that model macromolecular assemblies relevant to regulatory, metabolic, signaling and structural cellular processes. The assembled complexes will be co-created through the in vitro evolution of a pool of diverse RNA and protein sequences. These initial sequences will be evolved through cycles of selection, mutagenesis, and recombination. Novel sequences will be screened for ability to rapidly and repeatedly form assemblies with characteristic molecular interactions that allow them to function in vivo and to be characterized structurally. Innovative biochemical, biophysical and computational analyses of the resulting libraries will enable the selective screening of sequences for novel properties, direct the computational and experimental design of functional assemblies, and enhance our understanding of how they function in vivo. The proposed Specific Aims are to (1) evolve and select recombinant DNA and protein sequences able to assemble in vitro to create regulated and static complex assemblies; (2) evaluate the resulting RNA and protein sequences for their ability to form well-defined dynamic and static complex assemblies in vitro; and (3) determine the detailed structure of a subset of RNA and protein sequences whose dynamic assembly enables new

functions. This project is innovative in its focus on RNA, protein and nucleic acid co-evolution and regulation, and in its use of rational computational design to evolve dynamic molecular assemblies. These approaches have not been applied to the problems of assembling dynamic molecular structures. The project is significant in its potential to provide critical molecular building blocks for complex cellular regulation and structure. Just five days after the

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