CALIFORNIA STATE SCIENCE FAIR
2015 PROJECT SUMMARY

Name(s)

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Project Number

35577

Project Title

Towards Rational RNA Therapeutics: 3D RNA Engineering in a Massive Open Laboratory

Abstract

Objectives/Goals

Rationally designed therapeutics based on RNA molecules have taken years to develop due to poor understanding of how RNAs fold to specific secondary and tertiary structures. This study aims to uncover the missing design rules using a game-based crowdsourcing approach (EteRNA3D), the #massive open laboratory#. Goal1 of this research was to create an interface that enables humans to tackle canonical RNA 3D design with the prospect of reaching atomic accuracy. Goal2 was to test if this interface elicits solutions from human players that can explore a larger space of realistic 3D RNA designs than a computational algorithm REDESIGN. Goal3 sought to both rigorously analyze human designs in comparison to computer designs via in vitro synthesis and structure mapping and to formulate new hypotheses for RNA 3D design.

Methods/Materials

Towards Goal1,a gamified web interface using HTML5 was created. The interface was tested on two paradigmatic puzzles that arise frequently in RNA therapeutics. Motifs were aligned using output from 3DNA. For each of the designs created with EteRNA3D, the sequence of special 3D noncanonical motifs was taken from the crystallographic database but the sequence of canonical helices remained optimizable. RNA sequences were synthesized in-vitro using PCR and then transcribed using RNA polymerase. To analyze the RNA for stability, the molecule was exposed to 1M7, the SHAPE procedure modifier.

Results

A total of 68 unique solutions were gathered from EteRNA#s top players in the initial phases of testing. Whereas the computer solutions for the tetraloop receptor puzzle ranged from 8 to 24 motifs, the player solutions were far more varied,with a range of 9 to 35 motifs. For the FMN puzzle,the player designs ranged from 20 to 36 motifs, and the computer designs only had a range from 16 to 23 motifs. The SHAPE reactivity analysis showed that the construct with the lowest deviation was designed by a human and that the designs that were created by players are more stable than those of the computer designed solutions. The study also revealed three 3D design rules that are the first of their kind in the field of RNA design.

Conclusions/Discussion

This study has demonstrated for the first time that a game-based crowdsourcing approach can generate successful RNA designs that outperform computer modeling techniques and fold into target shapes in three dimensions, allowing for generalizable 3D design rules.

Summary Statement

By integrating RNA motifs into a 3D game, and using crowdsourcing based on the community of EteRNA players, our work paves the way for discovering design principles that can be applied to many problems in RNA therapeutics.

Help Received

Worked under the mentorship of Dr.Rhiju Das at Stanford University school of Medicine