Identifying Peptide Ligands Through mRNA Display

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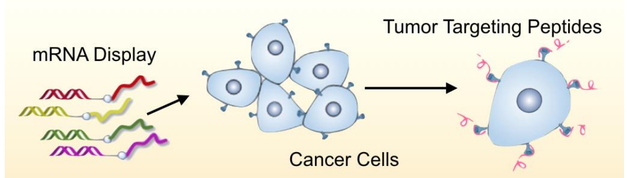
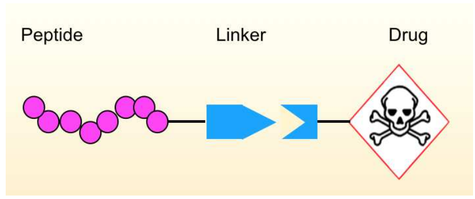
Introduction and Objective

Professor Roberts invented mRNA display, a method to identify the peptide ligands that bind with specific target proteins. Chloe and I worked to characterize the effect of salt concentration and bead matrix on nonspecific binding.

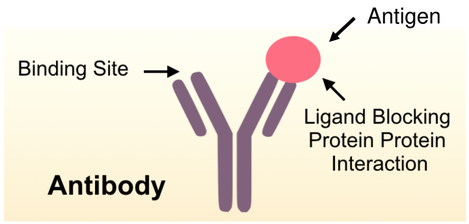
Applications & Impact of Professor's Research

Locate Tumor Cells and Treat Cancer





Break Protein Protein Interaction

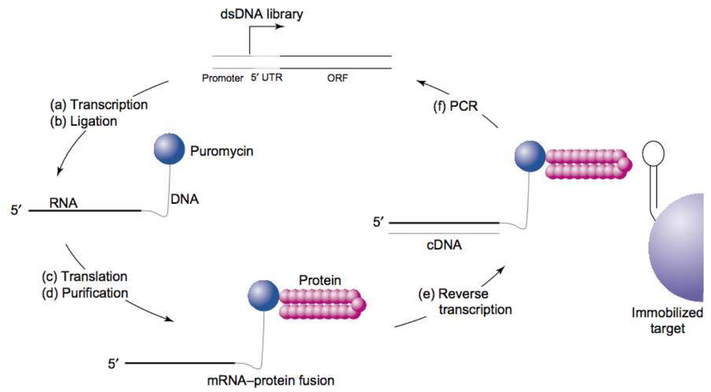


Acknowledgements

Thank you to everyone who has helped me throughout my SHINE experience. A special thanks goes out to Professor Roberts, Dr. Noridomi, Dr. Mills, Dr. Herrold, Chloe Kim, Marymount Highschool, and my parents.

Methods

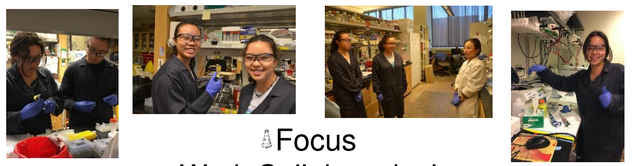
PCR: DNA library is amplified/multiplied.
Transcription: DNA is converted to mRNA.
Ligation: Puromycin/DNA strand is ligated to mRNA strand.
Urea Gel Purification: Ligated mRNA is separated from unligated mRNA.
Translation: mRNA is converted to a sequence of amino acids (peptide).
* It remains attached to the mRNA sequence.



Typical mRNA Display Process Diagram Courtesy of Terry Takahashi, Ryan Austin, and Richard Roberts

dT Purification: The ligated mRNA/peptide is purified.
Reverse Transcription: The synthesis of DNA from an RNA template.
* The DNA sequence allows us to identify the sequence of binder peptides.
Selection: Selection of binder peptides by introducing to a target protein.
PCR and Next Round of Selection: Repeat round of selections until the library gets enriched.

Advice for Future SHINE Students



- Focus
- Work Collaboratively
- Take Advantage of All Learning Opportunities
- Listen
- Be Prepared
- Have Fun!

Connections to STEM Coursework

mRNA display revolves around the process of creating proteins. This directly relates to the coursework I studied in Biology. Additionally, I applied many chemical principles and information to accurately perform tasks. My work in the lab involved making buffers and altering the concentration of samples. Contrasting to knowledge I gained in the classroom, the lab stresses the bigger picture as well as the purpose of every step and experiment. I hope to continue looking for the bigger picture when I return to the classroom.

Skills Learned

PCR

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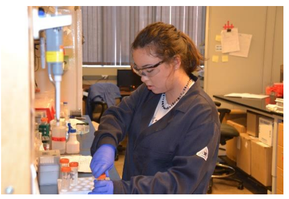
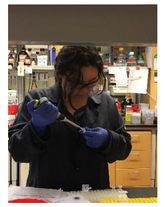
Lab Safety



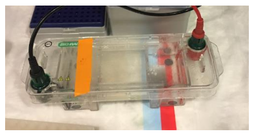
Nanodrop

Centrifuge

Micro Pipetting



Agarose Gel Electrophoresis



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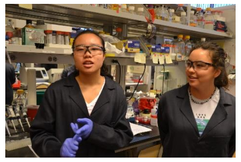
Communicating Research



Valuable Student Techniques

- Practice preparing for labs
- Updating lab notebook
- Importance of understanding the purpose and bigger picture of each step