Bioengineering Day Poster Addendum (ABET questions)

1. List two to four **Desired Needs** of your project that led to your final design objectives.
   Answer in two to four bullet points or concepts within a sentence or two.
   - Stable, low dose, protein expression vectors do not exist currently.
   - Stable, low dose, persistent RNA transcripts also do not currently exist.

2. List the major **Constraints** on your design/project
   a) Safety/Regulatory Affairs
      a. mRNAs, and especially virally derived mRNAs, are highly regulated as a biologic capable of reprogramming cells.
   b) Risks
      a. Any saRNA based system is highly immunogenic and therefore cytotoxic. Without extensive engineering, it will potentially act as a vaccine against the expressed protein which could be extremely dangerous.
   c) Global Impact
      a. RNA needs to be stored very cold to be stable, and our circularizable elements make that doubly important, so low income areas would likely not get access to any RACER tech.
   d) Manufacturability
      a. RNA manufacture is already extremely well optimized.
   e) Quality Control/Marketability
      a. dsRNA chromatography and construct testing are also well optimized and are not a limiting factor. The viral elements may be a difficult sell, but viral vectors are already used for gene therapies so that issue is likely limited.

3. List the major **Engineering Standards** on your design/project

a) Standards for saRNA and mRNA manufacture and delivery are already established by existing or past clinical trials. Persistent mRNAs, however, have not yet been fully developed and will carry their own new set of risks that will need to be mitigated by standards developed from our RACER construct.

3. Explain **Ethical, Environmental, or Societal concerns** for practical applications of your project.

There are limited concerns regarding our project. RNA is cheap to produce, degrades rapidly outside of freezers, and poses little threat to society. The only risk is what GOI is transferred. There is already a rapid increase in the recreational use of untested "peptides" for performance enhancement and health. The possibility of acquiring a long-lasting platform that generates proteins endogenously could easily be misused, though the limited stability of RNA should mitigate this compared to purified proteins.

5. Describe **Active Teamwork** and **Leadership** in your design group

a) **collaboration** and inclusion of diverse opinions?

This happened frequently. All opinions from lab members and team members were actively considered.

b) **delegation** of leadership on subprojects?

Each team member often worked nearly independently on various sub-projects. Delegation was decided on democratically.

c) establishing and reaching **goals and deadlines**?

This was less concrete. The project was more exploratory so deadlines were uncertain at best.

d) received or given **constructive feedback**?

Constructive feedback was frequently received from the project mentors.

6. What were the most significant motivating factors that led you to
   a) acquire **new knowledge**

Almost entirely curiosity, but resume building didn't hurt

b) be **self-initiating**

It is all but necessary when working solo on a sub-project

c) **persist** against challenges and setbacks.

Definitely the fear of not getting a good grade or producing good work.

7. What are your most **innovative and/or entrepreneurial ideas** for this project

The long term expression of proteins endogenously would have huge applications in the performance enhancement market. The platform could also likely express gene or base editors and the RNAs needed to program them. This could ensure highly successful in-vivo gene editing.