TEAM: SEEK & DESTROY
Tech Spec: Strengths/Weaknesses/Opportunities
Grade for Presentation: 90

Each project idea must have
+ A description of your system's design in terms of **devices**
+ A description of your system's design in terms of **parts**
+ A **timing diagram** to show anticipated system operation
+ A plan for **testing and debugging** your first generation system
+ A description of the **impact** you envision for your system
+ A description of any concerns raised and **open issues** within your team
+ A **"GO/NO GO"** decision
plus simulation/model in Tinkercell!

You've done such a great job of narrowing the focus of your project to a "manageable" but still ambitious effort. You have also found a key contributor to the repair of the defect, namely the "minidystrophin" gene. I also like that you've thought carefully about how much expression you'll need (or is tolerated) from the newly inserted RNA. There are some adjustments you'll have to make based on some limitations of biology, but I think they are refinements that you'll come to appreciate and will make your system more attractive.

1. Strengths
   - *AAV and minidystrophin seem like two great tools for the targeting and the repair of the defect*
   - *Need only a small amount of gene expression in defective cells since protein or RNA is so long-lived*
   - *Good to think about delivering RNA "payload" only at muscle cells since this will keep off-target effects of virus to a minimum*
   - *Nice, step-wise testing plan for system from petri dish to mice to humans.*

2. Weaknesses
   - *E. coli is not a cell that will produce AAV so need to reconsider the chassis*
   - *mRNA from AAV may not survive in muscle cell as long as natural mRNA since viral mRNA may not be 5'capped or 3' polyAdenylated (2 RNA processing events that make normal RNA in eukaryotic cells more stable)*
   - *Not clear how much of natural genome from AAV will be needed in addition to minidystrophin gene in order to make AAV infect muscle cells and to make AAV replicate in "messenger" cell*

3. Opportunities

I really like Tom's idea of separating out the virus producer from the virus itself, especially if the virus you've chosen targets muscle cells on its own. You do have a real challenge in figuring out what kind of cell you can use to get the virus produced and a second challenge surrounding the targeting and delivery of the RNA into the muscle cell (if the virus doesn't do that naturally). If finding a good chassis to produce AAV becomes a huge obstacle, then maybe consider modifying a yogurt-bacteria with a "phage" that could find muscle cells as well as deliver RNA to them. One other key thing to note: you will have to verify that the RNA from the virus can be directly expressed in cells or if it has to be made into DNA first. The good news is that you can apply this project in cells that don't have any dystrophin (Duchenne's patients) and so you don't have to worry about the interactions with the defective copy in the cell (a question that was asked by a few people during your talk).

Next steps

1. Define a receptor or targeting mechanism that could bring virus to muscle cells
2. Look into which genes in AAV are absolutely needed for infectivity into muscle cells and which are necessary for replication in host cell. You'll need to make sure you still have enough packaging room in the virus after that.
3. Determine if RNA from virus is really expressed or if it's reverse-transcribed into DNA and then expressed when AAV normally infects cells
4. Find a cell that can produce AAV and then decide where to "place" it to be the virus sender in the body

Candidate consultants:

Immunology expert
Muscular Dystrophy worker (Lou Kunkel?)
Virologist (Don Coen?)

Other notes:

- Nice to use Tinkercell to set up system—more of a "model" though than a "simulation" as it's labeled
- GREAT job with Q/A
- Loved the hand-drawn pictures you included in the ppt