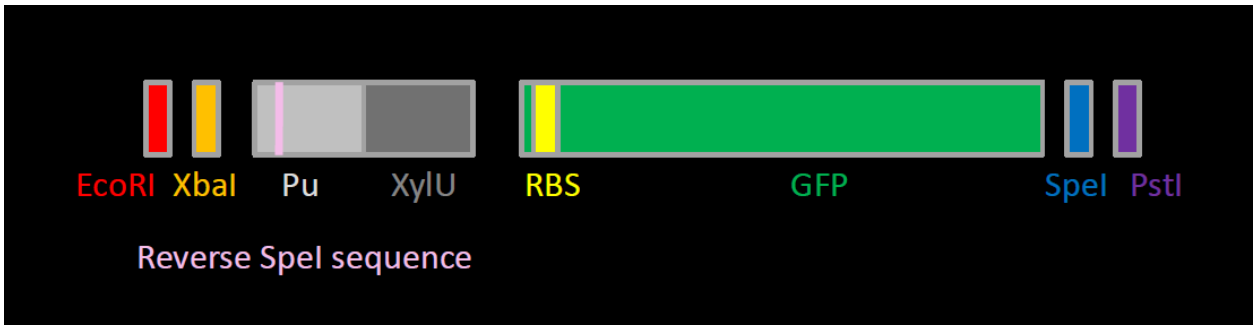
Tuesday 9/22/09

Weekly Team Meeting: Project Modifications

At the team meeting this week we decided to take the project in a new direction to finish in time for the jamboree

- Revising Pu promoter
    - When designing the primers for the Pu promoter, the beginning of the XylU gene was also amplified. Since we don't know what the consequences of having start codon with no stop codon, we will redesign the Pu promoter to only include the 200 bp promoter portion plus any downstream sequence that will make primer design easier.
        - The promote region is 200 bp because there are upstream sequences where the regulator proteins need to bind
    - The previous Pu construct is listed below:



- Create a working part in E. coli
    - Our previous goal was to get this part functioning in *P. putida* KT2440. However, since it has high antibiotic resistances to the antibiotic resistant markers we chosen to use, it has made genetic manipulation, especially transformations difficult. To have a goal that can be finished in time for the jamboree, we will work on getting this construct functioning in *E. coli*. This will require the addition of the Pr XylR regulating promoter and gene on the same plasmid as the Pu promoter. The Pr XylR will be amplified from the Tol plasmid with colony PCR
    - If we decided to continue this project after the jamboree we will work on getting this construction functioning in *P. putida* KT2440