6/9/2015

Aims for today:

1. PCR cleanup for lacZ
2. Digest lacZ and pDawn
3. Digest cleanup
4. Ligation of pDawn and lacZ
5. Lux box transformation
6. Design one plasmid system with Lux box/lacZ and design two plasmid system, one with Lux box and the other with the lacZ reporter

Accomplishments:

1. PCR cleanup for lacZ
2. Digest of lacZ and pDawn
3. Digest cleanup of lacZ and pDawn
4. Ligation
5. Lux box transformation
6. Began working on plasmid design
7. Inoculated pDawn

Aims for tomorrow:

1. Potentially design primers for cloning Lux box into pDawn/lacZ
2. Miniprep pDawn
3. Design SapI primers
4. Design compatibility system
5. Pick colonies for BioBrick, inoculate, glycerol stock
6. Colony PCR pDawn + lacZ, glycerol stock, miniprep, sequencing
7. Induce lux box w/ arabinose

Questions:

In the “empty” region of pDawn (the only space we think a biobrick can be cloned) there’s only one restriction enzyme that cuts once in that region and nowhere else, SapI.

1. Are we right in assuming that empty space is the only place we can clone in a biobrick?
2. Would it be okay if we used only one restriction enzyme for the ligation instead of two?