BABS UNSW iGEM Lab Protocol

Procedure	Name	Overlap PCR				
	Description	For when you want to stick two DNA bits together and don't want to use restriction enzymes				
Doccument	Name	Mackenzie Labine-Romain	Date	29/08/15	Version	1
Requirements	Time	2-3 hrs				
	PPE	Gloves, Labcoat				
	Equipment	Taq polymerase Taq buffer dNTP mix Two DNA samples with at least 20bp overhangs				
	Materials	Thermal cycler				
Step 1	Add 5ul 10x Taq buffer, 1ul dNTPs, 0.25ul Tag polymerase, and equimolar amounts of two DNA samples (as much as possible or ~200ng each) to a 200ul PCR tube					
Step 2	Make up to 50ul with MilliQ water.					
Step 3	Run reaction in a thermal cycler, varying annealing temperature based on overlap Tm.					
Step 4	Run on a gel to check for desired product, then PCR purify (Qiagen kit) and nanodrop.					
Notes	It is probably best to make a small sacrifice, say a piece of Parafilm or a small eppendorf tube, to the gods of PCR prior to protocol for best results.					
Version History						