SLiCE Reaction

1) Prepare 10X SLiCE Buffer in a 1.5 mL tube :

	500	μL	1 M Tris-HCl pH 7.5
+	50	μL	2 M $MgCl_2$
+	100	μL	100 mM ATP
+	10	μL	1 M DTT
+	_____		ddH_20 to
	1	mL	

Store at -20 C in 40-60 μl aliquots.

2) Add the following ingredients into a 0.2 mL tube in this order and **vortex**:

			linearized vector backbone (50-200 ng)
+			each additional assembly piece (1:1-1:10 molar ratio of vector:insert)
+	1	μL	10X SLiCE buffer
+	1	μL	PPY SLiCE extract
+	_____		ddH_20 to
	10	μL	

3) Incubate the SLiCE reaction mix as above at 37 C for 1 hour using a PCR machine or water bath, and then place on ice.

4) Transform 1 μL of the assembly reaction into 100 μL of competent *E. coli* and/or run a diagnostic agarose gel to check for successful assembly.

5) For electroporation, transform 1 μL into 20 μL electrocompetent cells. For large recombinant DNA, electroporation is required. In complex cloning, electroporation is recommended, as it is 10-100 times as efficient as chemical transformation.