

DNA Sequencing lab, UCR-GI

We ask that you will bring us **12 μ l** of your sample DNA template and a primer sufficient for two reactions in a **0.2ml tube**. Please put **your initials** on the top of the sample tube and fill out the **login sheet** properly.

Required amount of primer and template

Template	Size of your template	In 12 μ l of water (a 0.2ml tube/ sample)	
		Template	Primer
PCR Products	>80*		2~10 pm
	80~150*	1~8ng	
	150~250bp	4~20 ng	
	300~500bp	15~25 ng	
	0.5~1kbp	25~50 ng	
	1kbp~2kbp	40~80 ng	
	>2 kbp	+5ng/100 bp	
	>8 kbp*		
Plasmids	~4 kbp	200~300 ng	2~10 pm
	4~8 kbp	250~350 ng	
	6~8 kbp	350~600 ng	
	8~10 kbp	500~700 ng	
Cosmid & BAC*		0.8~1.5 μ g	2~10 pm
Genomic DNA*		2~3 μ g	4~6 pm
ssDNA		50~100 ng	1.6 pm
Primer Walking*			

*See us before you bring us any samples or contact yoongi.choi@ucr.edu.

Most common contaminants are salts, bacterial proteins, cell wall carbohydrates and organic solvents. Following table contains tolerable concentration of contaminants but we are expecting your DNA and primer in water.

Contaminants	Maximum Tolerance level	Affect
RNA	1 µg	High background
PEG	0.3 %	Inhibition of polymerase
EtOH	1.2 %	Inhibition of polymerase
NaOAc	0.4 mM	Inhibition of polymerase
Phenol	0	Inhibition of polymerase
Chloroform	0	Inhibition of polymerase
CsCl	0.4 mM	Inhibition of polymerase
EDTA	0.2 mM	Weak Signal

Bacterial cells for Plasmid DNA preparation

Recommendation	Strain	Comparison (Success rate)
High	DH5α, HB101	99%
OK	JM109 > XL-1 Blue	90%
Not	MV1190 > JM101	60%