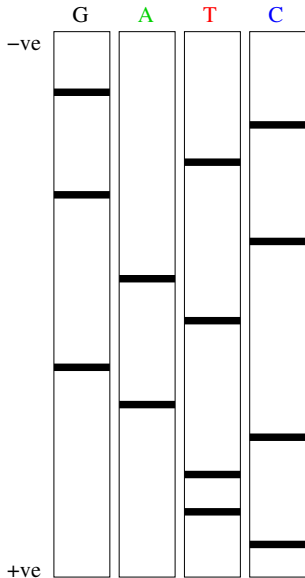


# Sanger sequencing

*Dideoxynucleotide*

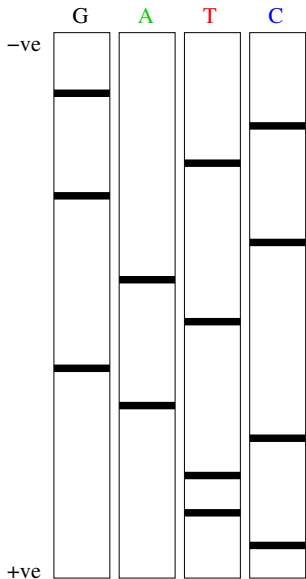


*Inferred DNA sequence*

p<sup>32</sup> CTTCA GTACGTCG  
p<sup>32</sup> CTTCA GTACGTC  
p<sup>32</sup> CTTCA GTACGT  
p<sup>32</sup> CTTCA GTACG  
p<sup>32</sup> CTTCA GTAC  
p<sup>32</sup> CTTCA GTA  
p<sup>32</sup> CTTCA GT  
p<sup>32</sup> CTTCA G  
p<sup>32</sup> CTTCA  
p<sup>32</sup> CTTC  
p<sup>32</sup> CTT  
p<sup>32</sup> CT  
p<sup>32</sup> C

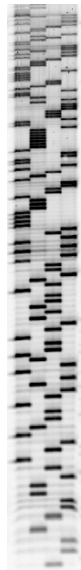
# Sanger sequencing

*Dideoxynucleotide*



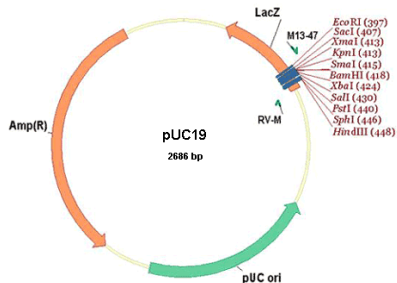
*Inferred DNA sequence*

p<sup>32</sup> CTTCAAGTACGTCTCG  
p<sup>32</sup> CTTCAAGTACGTC  
p<sup>32</sup> CTTCAAGTACGT  
p<sup>32</sup> CTTCAAGTACG  
p<sup>32</sup> CTTCAAGTAC  
p<sup>32</sup> CTTCAAGTA  
p<sup>32</sup> CTTCAAGT  
p<sup>32</sup> CTTCAAG  
p<sup>32</sup> CTTCA  
p<sup>32</sup> CTTTC  
p<sup>32</sup> CTTT  
p<sup>32</sup> CTT  
p<sup>32</sup> C



Autoradiogram courtesy of Dr. Rahat Zaheer

# A typical sequencing/cloning plasmid



pUC19 is a small, high-copy number *E. coli* plasmid cloning vector, with multiple cloning sites. The molecule is 2686 bp in length. pUC19 encodes the N-terminal fragment of  $\beta$ -galactosidase (*lacZa*), which allows for blue/white colony screening (i.e.,  $\alpha$ -complementation), as well as a pUC origin of replication.

The cloned sequence can be amplified by PCR with M13-47 and RV-M primers or sequenced with either primer.

