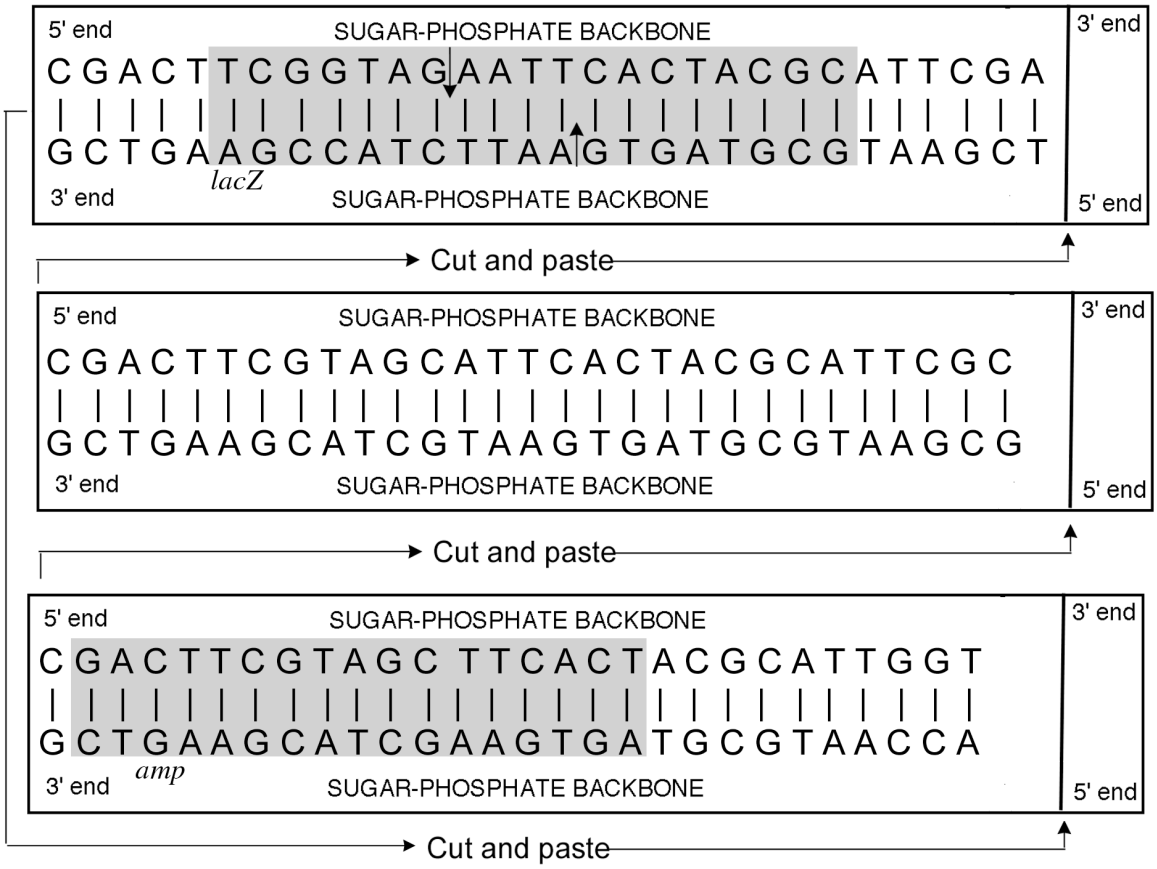
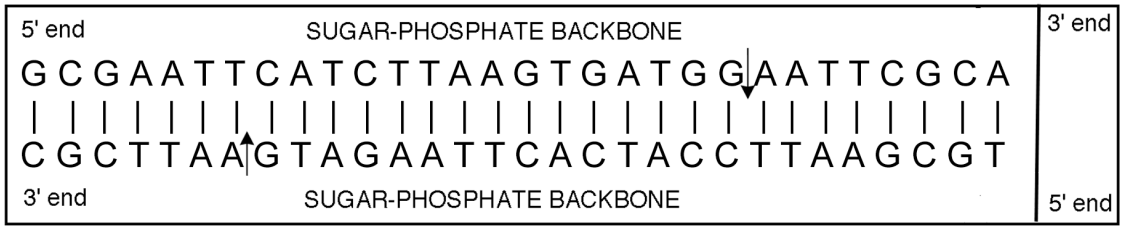
Practice with Genetic Engineering—Part 1.

Isolating a plasmid vector. Cut the three plasmid pieces on the solid lines and tape together to form a circular molecule of DNA. In DNA synthesis, the 5′ end of one sugar is attached to the 3′ end of the preceding sugar. This plasmid carries the (shaded) genes for resistance to the antibiotic ampicillin and for hydrolysis of lactose (*lac* Z).



Practice with Genetic Engineering—Part 2.

Isolating a gene. Genes can be isolated or, if small enough, synthesized. Cut out the gene on the solid lines.



Practice with Genetic Engineering—Part 3.

Restriction enzyme digestion. Your scissors are the restriction enzyme *Eco* R1 which cuts between the G and A in the sequence G↓AATTC on both strands of double-stranded DNA. The enzyme reads from the 5′ end of DNA. Note that the two strands of DNA are complementary so you will have staggered (or sticky) ends after your cuts. Cut the plasmid and gene. Although enzymes work by trial-and-error to find their substrate, the arrows (↑) will help you locate the correct sequence.

Ligation. You can use tape in place of the DNA ligase that covalently joins pieces of DNA. Match the complementary bases of the staggered ends to tape the gene into the plasmid.

Voilà! A recombinant plasmid to insert into a cell.

Questions: How will you identify cells carrying the recombinant plasmid? Cells carrying the original plasmid? Cells without a plasmid?