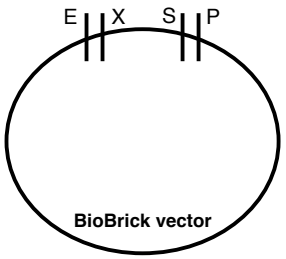
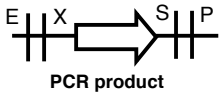
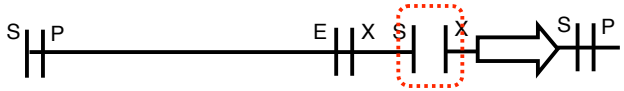
Cloning BioBrick parts by fusion PRC

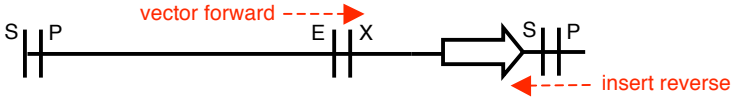




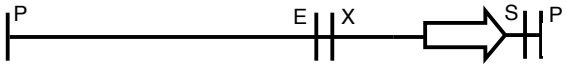
1. Digest BioBrick vector with SpeI and PCR product with XbaI. Heat treat the digests at 70°C for approximately 30 minutes to inactivate restriction enzymes.



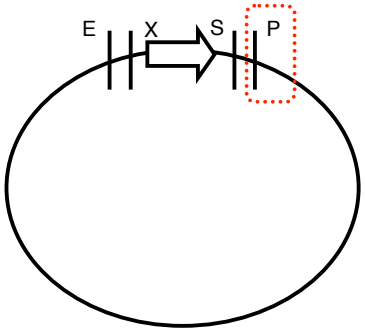
2. Set up overnight ligations of the digested, enzyme-deactivated, BioBrick vector and PCR product together.

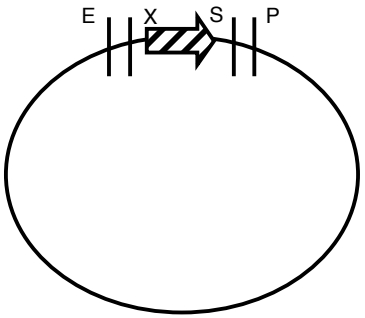
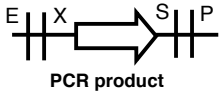
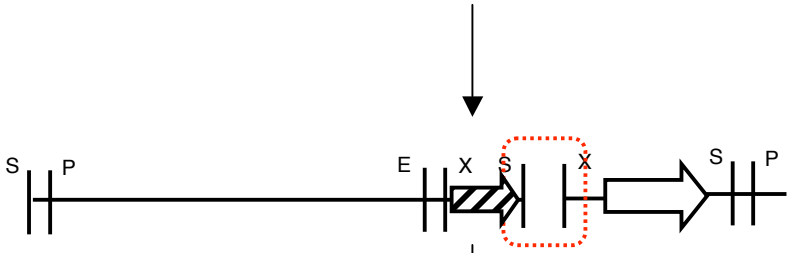
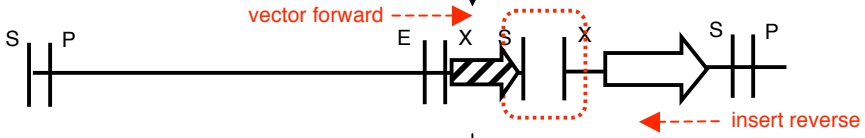
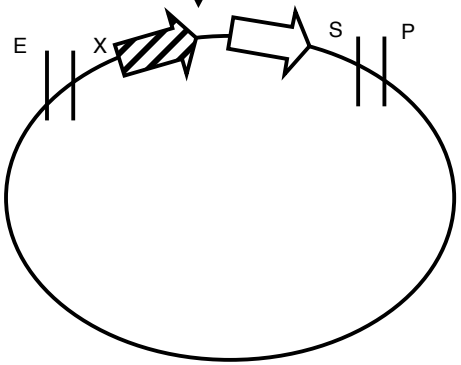


3. Set up a PCR reaction using the ligation as the template and vector (forward) primer and the reverse primer of the gene of interest.



4. Digest the product from the fusion PCR with PstI or SpeI to generate sticky ends. Heat treat at 70°C for approximately 30 minutes to inactivate restriction enzymes.



5. Set up overnight ligation and you're ready to transform cells!

The same procedure can be used to make composite BioBrick parts.