

Tel: (215) 573-7407

Fax: (215) 573-9327

E-mail: dnaseq@mail.med.upenn.edu FTP: dnaseq.med.upenn.edu

Web: www.med.upenn.edu/genetics/dnaseq/

Mailing Address:

B1 Richards Building

3700 Hamilton Walk

Philadelphia, PA 19104

REQUEST FOR CONSTRUCTION OF TARGETING VECTOR

PI _____

Fund No _____

Tel _____

Date _____

Contact person (full name) _____

Tel _____

*E-mail (Required) _____ Fax _____

Please attach a separate sheet detailing the targeting vector designed based on the recombinering method of Dr. Neal Copeland (Liu et. al. 2003, 13:476-484, Genome Research)

Only **final** vector design will be accepted. We strongly recommend not to synthesize any PCR primers before the strategy is completely laid out. Construction of a targeting vector is complex and therefore it is extremely important to describe the entire strategy clearly. Please take your time to figure out your strategy and re-check it several times until you are sure that it is correct. A meeting will be set up at this point to discuss your final design.

Please draw a detailed sketch of the final targeting vector on a separate sheet. **Please do not use copies of existing maps of cloning vectors with genomic arms pasted on. Rather draw all parts of the final targeting vector by hand or use software such as vector NTI to describe the final vector electronically. Use large letters that are easy to read. Your figure(s) should be sufficient to describe the entire procedure and a separate document should contain the following information:**

1. The genomic target and its full sequence printout.
2. The most suitable region to locate the two homology arms for target retrieval with your choice of restriction sites.
The latter have to be compatible with your southern blotting strategy.
3. For conditional knock-out construct, clearly identify the region to be knocked out and suggested locations of the homology arms of the two mini targeting vectors with your choice of restriction sites.
4. For knock-in construct clearly identify the location of mutation(s) on your target.
5. For knock-in construct suggested locations of the homology arms of the mini targeting vector with your choice of restriction sites.