Primer design for semi-quantitative RT-PCR.v1

1. Obtain mRNA sequence from your source or online and save it in a word file. Print the sequence as a hard copy.

2. Paste the sequence to the query window at http://frodo.wi.mit.edu/cgi-bin/primer3/primer3_www.cgi.

3. Make changes to the "Product Size Ranges" to 401-500 (Delete the rest and leave 401-500).

4. Hit "Pick Primers" to obtain the sequence information. The search engine will give you the best choice and some other choices.

5. Decide which primer set you want and save the sequence information in a word file. Give each of the primers a name, for example, Igf1r F for IGF-1R forward primer. Mark the primer positions on the hard copy of your sequence.

6. You may also use http://scitools.idtdna.com/Primerquest/ to design your primers (Please try it on your own).

7. Give the sequence information to Mei for her to order the primers. File everything in two places in Zhen Yan's office: Oligo folder and Gene folder.

8. Resuspend the oligos in 1X TE at 200 μM stock concentration and store at –20°C when the oligoes are received,. For working stock, dilute the oligos in 1X TE to a final concentration of 10 μM and store at 4°C. Update the information in the database. Please add the complete information to the database.