

Primer	Sequence	Restriction sites	Purpose
1	5'-GACGAAAAGGGTGACCGTGATAAGGAG-3'		Generate KDEL. Used with primer 2 and T4 polymerase. The fragment generated was cloned in pBLUESCRIPT digested with Xho I-Hinc II
2	5'-CCGCTCGAGTTACAAATCCTCCTTATC-3'	XhoI	see primer 1
3	5'-CCC <u>AAGCTT</u> ATCTTTTACCCA 3'	HindIII	Amplification of 3 X HA tag from pSLF-72 vector. Used with primer 4. The product was cloned in pBLUESCRIPT:KDEL digested with Hind III-EcoR V
4	5'-CCGGGTCGAGATATCAGCGTA-3'	EcoR V	see primer 3
5	5'-CCC <u>AAGCTT</u> TTCCAACCTCTTCTTC-3'	Hind III	Amplification of CRT. Used with primer 7. The fragment was cloned in pBLUESCRIPT:HA-KDEL digested with Hind III-EcoR I
6	5'-CCC <u>AAGCTT</u> TCCGCCTTCTTCAGG-3'	Hind III	Amplification of CRT-ΔC. Used with primer 7. The fragment was cloned in pBLUESCRIPT:HA-KDEL digested with Hind III-EcoR I
7	5'-GGAATTCATGCGTGACAGCA-3'	EcoR I	see primer 5
8	5'-CCC <u>AAGCTT</u> TTCCAACCTCTTCTTC-3'	Hind III	Amplification of CRT-C. Used with primer 9. The product was cloned in pBLUESCRIPT:KEDL digested with EcoR V-Hind III
9	5'-ATCGGCCTGAAGAAAGCG-3'		see primer 8
10	5'-GGAATTCATGGTGAGCAAG-3'	EcoR I	Amplification of GFP. Used with primer 11. The fragment was cloned in pBLUESCRIPT: KEDL or pBLUESCRIPT:CRT-C-KDEL, both previously digested with EcoR I-EcoR V
11	5'-CTTGACAGCTCGTCCATC-3'		see primer 10
12	5'-GGCCTTGCGACACTGTCAGCCGTGCA CGGCACGGTGAGCAAGGGCGAGGAGC-3'		Introduction of signal peptide by partial annealing with templates pBLUESCRIPT:GFP-KEDL or pBLUESCRIPT:GFP-CRT-C-KEDL. Used with primer 13
13	5'-CAAAAGGGCACAGAAAAAATTGCTG CACGCATGAATTCCTGCAGCCCGGG-3'		see primer 12