

PCR SST2KO

1- PCR mix

2,50 µl 10X buffer(Q BIOgene ; 1,5 mM MgCl₂)
 0,25 µl dNTP (20 mM total)
 0,40 µl primer SST2 a (200 ng/µl)
 0,20 µl primer SST2 b (200 ng/µl)
 0,20 µl primer SST2 c (200 ng/µl)
 0,20 µl Taq polymerase (Q BIOgene ; 5U/µl)
 19,25 µl distilled water

2- PCR reaction :

23,0 µl mix
 2,0 µl DNA (200ng/µl)

3- PCR program :

5 min	94°C	
30 sec	94°C	
30 sec	62°C	35 cycles
1 min	72°C	
5 min	72°C	

4- Primers:

- Primer SST2 a 5' CTCACTGAACAGCTAGCCC 3'
 - Primer SST2 b 5' CCCATTCAACTGCTCAGAGC 3'
 - Primer SST2 c 5' TGGATGTGGAATGTGTGCGAG 3'

5- Amplifications:

wild-type allele: about 350 bp
 mutant allele: about 410 bp