

PCR DNA pol mu

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 Pol mu a (200 ng/µl)
 0,20 µl primer Pol mu b (200 ng/µl)
 0,20 µl primer Pol neo (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	
1 min	94°C	35 cycles
1 min	60°C	
2 min	72°C	
5 min	72°C	

4- Primers:

- Primer Pol mu a 5' GGGTTGGGGTGAAGACTGC 3'
 - Primer Pol mu b 5' CTCATGGCCAACCCTGGGTC 3'
 - Primer Pol neo 5' CATAGCGTTGGCTACCCGTG 3'

5- Amplifications:

wild-type allele: 250 bp
 mutant allele: 500 bp