

p110d

PCR conditions: GEN65

1. 94 for 3 min
2. 94 for 30 sec
3. 65 for 30 sec
4. 72 for 30 sec
5. go to 2 39 times
6. 72 for 7 min

With cassette primers (lacZ)

For 1 ml stock (10X):

50 ul 100 pmol/ul stock of D149
100 ul 100 pmol/ul stock of D73
50 ul 100 pmol/ul stock of NeoF1
800 ul TE or H₂O

D73	CCTGCACAGAAATGCACTTCC
D149	AACGAAGCTCTCAGAGAAAGCTGG
Neo F1	CGCCTTCTATCGCCTTCTTGAC

WT- 312bp HET- 409bp + 312bp KO- 409bp

Without cassette primers

For 1 ml stock (10X):

50 ul 100 pmol/ul stock of D71
50 ul 100 pmol/ul stock of D146
900 ul TE or H₂O.

D71	GCGTAACAGAGAGCAAAGTCCC
D146	AGGGAACCGCCGTATGAC

WT- 232bp HET- 232bp + 404bp KO- 404bp