

PROTOCOL; PDGF-A ex6

Primers

5' forward primer (PDGF-A ex6 fwd) - 5' **aac aac att cag gca gcc c** 3'

3' reverse primer (PDGF-A ex6 rev) - 5' **tta cct gat tcc cta cgc c** 3'

PCR profile

93 °C, 3 min

93 °C, 30 s 40 cycles

55 °C, 30 s

65 °C, 20 s

65 °C, 10 min

4 °C, ∞

PCR mix

10 x PCR Buffer	3.0 µl
MgCl ₂ (25 mM)	1.8 µl
dNTP (25 mM)	0.24 µl
forward primer (100 µM)	0.15 µl
reverse primer (100 µM)	0.15 µl
Taq polymerase (5 U/µl) (Invitrogen, cat#10342-053)	0.2 µl
DNA template (~ 0.5 µg tail DNA)	2.0 µl
H ₂ O	<u>22.46 µl</u>
	30 µl

Post-PCR analysis

Load 12 µl of the PCR reaction on a 2-3% agarose gel.

Expected pattern is mutant; 147 bp

Wt: 185 bp