PCR: Cbl-b KO

Amplifluor: No

Primers

Nucleotide sequence (5` --> 3`)

Primer	Sequence
Cbl-b KO	TTCCTCCCACTCATGATCTATAG
Cbl-b Rev	GGAAAAATATTAGTTACAACTGG

Reagents

Reagents/Constituents	Commercial name	Stock Concentration	Volume (µL)
DNA Sample			1.0
10x Buffer	10x 0289 PCR Buffer	0.0 mM MgCl2	4.0
dNTPs		10.0 mM	1.6
Primer: **Cbl-b KO**		10.0 µM	
Primer: **Cbl-b Rev**		10.0 µM	
Water			26.05
Taq Polymerase			0.05
Reagent: 25mM MgCl2			4.8

Comments on protocol: DNA Sample (ear punch diluted 1:1 with water)

Strategy

Steps	Temp (°C)	Time (seconds)
Hot start: **No**		
Initiation/Melting	94.0	120
Denaturation	94.0	30
Annealing	55.0	30
Elongation	72.0	30
Number of cycles (repeat denature, anneal & elgonate): **32**		
Strand completion (i.e. 72°C, 10 min)	72.0	10
Finish (i.e. 4°C, indefinite)	10.0	Hold

Electrophoresis

Running buffer:	
% Agarose:	2.5
Volt:	400.0
Estimated running time (min):	10.0

Bands

Number	Band (bp)	Genotype
1.	550	Cbl-b KO