RIKEN BRC

Sheet ID PS_00266 Print: 12/1/2011

RBRC00339 **Strain name** C57BL/6J-CSA/+

Primer

	primer_name	primer_seq	length
1	LCB19(CSP3)	GTGTGTTTTGCCGTTTCCAG	20
2	LCB20(CSP4)	GTTACCCTCCAGCCCTTGTA	20

Reaction Components

Enzyme Roche Taq (Cat. No. BM1418432)

	Conc	Run 1	Run 2	Run 3
H2O		17.2		
PCR buffer	10 X	2.5		
dNTP	2.5 mM	2.0		
primer 1	10 pmol/μl	1.0		
primer 2	10 pmol/μl	1.0		
DNA		1.0		
Taq		0.3		
	total	25		

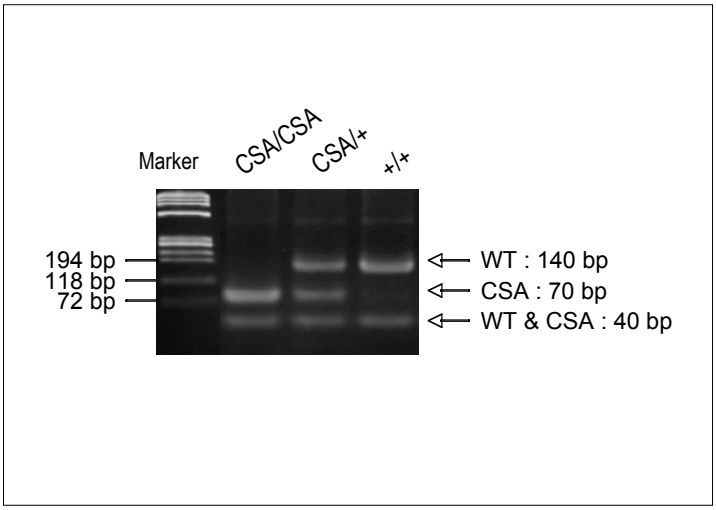
Reaction Conditions

step	temp (℃)	time (sec)	note
1	94	60	
2	94	30	*
3	55	60	*
4	72	30	* *Repeat for 35 cycles

Restriction_Reactions

PCR product 7.0 μL
10 X Buffer 1.0 μL
Taq I 0.2 μL
dH2O 1.8 μL
65℃, 60 min.

After Taq I (NIPPON GENE : Code 318-00372) digestion, run 10 μl of digested products on a 4% Nusieve in 0.5 X TBE .
CSA/CSA 70 , 40 bp
CSA/+ 140 , 70 , 40 bp
+/+ 140 , 40 bp



Marker

ϕX174 Hae III

Gel

4% Nusieve in 0.5 X TBE (100 V, 45 min)

primer_set		Band_size	product