

Gene symbol	<i>Cacna1a</i> <sup>gry</sup>			
primer	Sequence(5'-3')	Size	target gene	accession
rCacna1a-14-F rCacna1a-14-R	CGTCCTGAAGTCATCATGAAG AGTCTCAGCTGCTCTGTGGTT	256bp	<i>Cacna1a</i>	NM_012918.1
comment				

PCR condition						
Taq polymerase	Nova Taq Hot start DNA polymerase					
Thermal cycler	PC-808(ASTEC)					
PCR buffer	Ampdirect Plus					
	first denature	denature	anneal	extension	cycle	final extension
PCR	°C	94	94	60	72	72
	min	3	0.5	0.5	1	3
comment	FTAカードに採取した血液を未精製のままテンプレートとして用いる。 PCR Productを制限酵素 <i>Pst</i> Iで処理(37°C、2時間)					

Electrophoresis ; 4% agarose gel 300V, 30min						
<p>The image shows an agarose gel electrophoresis (4% agarose) result. Lanes are labeled from left to right: 1, 2, 3, gry/gry, gry/+ and +/+ genotypes, and M (marker). Lanes 1, 2, and 3 show bands at approximately the same position, indicating they are homozygous for the gry mutation. The gry/gry lane shows a single prominent band. The gry/+ and +/+ lanes show two bands, with the lower band being more intense in the +/+ lane. Lane M shows a ladder of bands.</p>						
<p>M: (φX174/Hae III digest)</p>						
Strain harboring mutation	GRY/Idr, WTC.GRY-Cacna1a <sup>gry</sup> /Kyo					