

RMRC Genotyping Protocol-PCR

種 原 編 號	RMRC13037	品 系 名 稱	FVB/N-Tg(PEPCK-Aurka)A79Tfts
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A. Primer

	Primer-name	Sequence	Length	Note
1	HS4 forward	cctccttgggcaacctgttcag	22 mers	
2	HS4 reverse	atgtggcactgagggacatggc	22 mers	

B. Reaction Components

Chemical	Conc.		Run1	Run2	Run3	Run4
H ₂ O	--		12.8 µl			
PCR buffer	10x		2 µl			
dNTP	2.5	mM	1.2 µl			
Primer 1	10	pmol/µl	1 µl			
Primer 2	10	pmol/µl	1 µl			
DNA	1~3 µg/µl		1 µl			
Tag		U/µl	1 µl			
Total			20µl			

C. Reaction Conditions

Step	Temp.(°C)	Time(Sec)	Note
1	95	300	
2	95	30	Repeat 2~4 for 35 times
3	66.5	30	
4	72	40	
5	72	300	
6	4	---	
7			
8			

D. PCR product

	Primer set		Product size	Product
1	HS4 forward	HS4 reverse	622 bps	HS4 insulator
2				

E. Gel photo