

Strain ID	MT1032
Strain Name	cyp19a1(K164X)

Mutated Gene	cyp19a1
Linkage Group	LG3
GenBank Accession No. for cDNA	D82968
Nucleotide Change (WT > MT)	A>T
Location of mutated nucleotide in wild-typegenomic sequence	TTCACC(A>T)AAGGTAAAA
Mutated Amino Acid (WT>MT)	K>X

Forward primer sequence (5'-3')	TCAGCTGCATCGGCATGAACGA	
Reverse primer sequence (5'-3')	CAGGTTTGGCCCAGTCAAAGCTATGTT	
PCR Product Size (bp)	MT	187
	WT	187
PCR Product Sequence (WT)	TCAGCTGCATCGGCATGAACGAGAAGGGAATCAT ATTTAACAACAATGTGGCTTTATGGAAAAAGATTC GTACCTATTTACCTAAGGTAAAATCTATCAACCA GAATTGCATTTTTCTGATTGAATATTGTACAATCTT TAATTATTGTTTTTTAACATAGCTTTGACTGGGCC AAACCTG	
PCR Reaction Conditions	PCR reactions were performed using Extaq (Takara Bio Inc, Japan, RR001A). 10x Extaq buffer: 1ul 2.5 mM dNTP mixture: 0.8ul 2.5uM primers: 0.2 ul Extaq: 0.05 ul template DNA: 1ul H2O: 6.95ul	
Thermocycler Program	95 °C 2 min (95 °C, 30 sec, 55 °C 30 sec, 72 °C 40 sec)x 30-35 cycles 72 °C 5 min	

Restriction enzyme(s)	DdeI	
Restriction digestion product sizes (bp)	WT	187
	MT	99, 83