

HABC Gentoype Data Submission Form

Submitting Investigator	
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Date Submitted	10/06/06
Polymorphism Information	
Name	R577X
RS# or Unique Identifier	rs2228325
Gene	Alpha-actinin-3
Chromosome position	11q13-q14
Alleles	R, X (for amino acid); C (G), T(A) for nucleotides; () = nucleotide for complementary strand
Assay Information	
Genotyping Method	TaqMan allele discrimination assay
Genotypes in HWE (Y/N) (attach HWE Form)	Yes for Caucasians, No for Blacks.
Amount of DNA used	~ 20 ng
PCR primers	F: ACGATCAGTTCAAGGCAACACT R: ACCCTGGATGCCCATGATG
PCR Components and Concentrations (Taq, buffer, MgCl ₂ , primers, DMSO, other reagents)	1.5 mM MgCl ₂ , 200 μM each dNTPs, 1 unit of Taq and 50 ng of each primer
PCR Cycling conditions (time and temperature for each step, # of cycles, etc.)	40 cycles of denaturation at 94°C for 30 sec, annealing at 58°C for 30 sec, and extension at 72°C for 30 sec, and a final extension step of 72°C for 5 min
Detection Oligo(s) (if applicable)	R allele: FAM-CGCTCTCGGTCAGC X allele: VICTCGCTCTCAGTCAGC
Other Reaction Conditions (detection reaction components, incubation conditions, gel %, etc.)	TaqMan allele discrimination assay was used that employed the 5' nuclease activity of Taq polymerase to detect a fluorescent reporter signal generated during PCR reactions. Both alleles were detected simultaneously using allele-specific oligonucleotides labeled with different fluorophores, and genotypes determined by the ratio of the two fluorophores used.
Other Assay Info	

