

Centro Nacional de Biotecnología





CNB-CSIC Mouse Embryo Cryopreservation Facility – Spanish EMMA node

GENOTYPING PROTOCOL

EMMA ID number	EM:04897
Mouse mutant strain	STOCK Myo7a4626SB/WtsiCnbc
Gene/Locus affected	Муо7а
MGI ID number	MGI:104510
Mouse strain common name	Myo7a4626SB
Owner/Provider of this mouse strain	Karen Steel
DNA primers for wild-type allele	Forward: 5'-GTGACCTCCGAGGGACATG-3' Reverse: 5'-AGAAAGATCTTGGTTTTGCCAGTC-3'
PCR conditions for wild-type allele	Program: 5'94°;35x(30''95°,30''57°,45''72°);10'72° PCR product: bp
DNA primers for mutant allele	
PCR conditions for mutant allele	
Source of genotyping information	Provider or PubMed ID: 14648237, 9435277, 9186010, 7870172, 18160714, 11753415, 12121736, 11222540, 10224267
Comments	Genotyping is performed by PCR followed by sequencing. These can also be genotyped by PCR followed by restriction digestion, but we have found this to be somewhat unreliable a times The Myo7a4626SB mutation was genotyped by PCR using the indicated forward and reverse primers annealing at 57°C. Use platinum PCR Supermix (Invitrogen) mix. The reverse primer introduces a base change that in conjunction with the Myo7a4626SB mutation creates an AccI restriction site. Thus, upon AccI digestion (at 37°C) the mutant PCR product is cleaved into a 63 and 25 bp fragment, and the wild type product is not cut.