Gene: Slc13a5 ENSMUSG00000020805; Indy, NaC2/NaCT, Nact, mINDY

**Genotyping protocol for Slc13a5-R337X**

NM\_001004148.4:c.1009C>T

***\****Do NOT mix up WT and MU primers in one tube. One primer set each tube.

**Primers for the WT allele**

SLC13A5-Ex7-WT-F-634: CCGAGACCCCGGCTTCA

SLC13A5-Ex7-WT-R-634: CCCCAGCCTACACTGCTCCA; RC: TGGAGCAGTGTAGGCTGGGG

Amplicon size: 634 bp

**Primers for the MU allele (R337X)**

SLC13A5-Ex7-MU-F-420: GGTTCagCtGAGAtCCtGGaTTtAT

SLC13A5-Ex7-MU-R-420: CATACACGCCCTTCCTCCAA; RC: TTGGAGGAAGGGCGTGTATG

Amplicon size: 420 bp

Each PCR reaction contained 10 µL of Platinum Hot Start PCR Master Mix (2x) (Invitrogen, Carlsbad, CA, USA), 1 µL of 10 µM primer mix, 2 µL of DNA template (final conc. <500 ng/rxn) and raised to 20 µL with distilled water. The reactions were run using a GeneAmp PCR System 9700 (Applied Biosystems, Foster City, CA, USA) Thermocycler.

The reaction conditions were: template denaturation @ 94 °C for 2 min, then [30 cycles of 94 °C for 30 s (template denaturation) then 62 °C for 30 s (primer annealing) followed by 72 °C for 50 s (primer extension)], 72 °C for 2 min and then hold at 4 °C.

>*Slc13a5*-**WT**, 634 bp

CCGAGACCCCGGCTTCATGCCTGGCTGGCTGTCATTCGCCTGGGTCGAGGGAAACACCGTGTAAGTCGATGCAAAGGAAAGTCTTAGCTTAGGATAATGATGTCTAAATGTCACTGTTGTCAGCAACAGGCAGCAAGAGGACGGTATGACTTGGGCCCTGTGTTCTGGAGGAACATTCCGTTCTGTGGGCTTTGGGGTGCATTTGTTTTCAATCAATCTTCAGAATGAAACTACGGGTGCATTTGTTTCTGTGGCTCCCTCATGACCCCTAGAAAGCTGAACCAAGAAAGTTTATTGGGACGAGAGTAGGCTCCTTAGAGCCCTGATTTCTGCCTTCTTCCTTTCTTAAGAACCTATCTGAGACACATCTTGGTTACTTAGAAACCTAGTTGGAGGAAGGGCGTGTATGTGGATTCTCTCCTGCCTCTGTTAGGGAGACCTACAAGAAATACTGAAGAATGACTATCCTAGTAAAGGCAGAAGATGTGGGTGAGGAGGTATCTGTCTGCAACTGGCTACTTCATCTTGGTTACTTAGCTTAGCACTACAGTCCTAGAGGTTGACTGAATCCAGAAGTCTAAATCTCATGGTTGAGAAATCATGATGTTCACCCATGGAGCAGTGTAGGCTGGGG

>*Slc13a5*-**MU**, 420 bp

GGTTCagCtGAGAtCCtGGaTTtATGCCTGGCTGGCTGGCTGTCATTCGCCTGGGTCGAGGGAAACACCGTGTAAGTCGATGCAAAGGAAAGTCTTAGCTTAGGATAATGATGTCTAAATGTCACTGTTGTCAGCAACAGGCAGCAAGAGGACGGTATGACTTGGGCCCTGTGTTCTGGAGGAACATTCCGTTCTGTGGGCTTTGGGGTGCATTTGTTTTCAATCAATCTTCAGAATGAAACTACGGGTGCATTTGTTTCTGTGGCTCCCTCATGACCCCTAGAAAGCTGAACCAAGAAAGTTTATTGGGACGAGAGTAGGCTCCTTAGAGCCCTGATTTCTGCCTTCTTCCTTTCTTAAGAACCTATCTGAGACACATCTTGGTTACTTAGAAACCTAGTTGGAGGAAGGGCGTGTATG



M: 1 Kb Plus DNA Ladder (Invitrogen, Carlsbad, CA, USA)