**Genotyping Primers**

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| GENE | Sequence 5' --> 3' |
| STOCK Piezo1- Forward | GCCTAGATTCACCTGGCTTC |
| STOCK Piezo1- Reverse | GCTCTTAACCATTGAGCCATCT |
| Piezo1- Wild type Forward | ACGCCAAGCTCATCTTCCT |
| Piezo1- Common | GTCCCTTTGACAGCAGCATC |
| Piezo1- Mutant Forward | CACCTGTTCCTGTACGGCATGGAC |

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| GENE | Sequence 5' --> 3' | |
| M-GAPDH | Forward | ACGGCCGCATCTTCTTGTGCA |
| Reverse | AATGGCAGCCCTGGTGACCA |
| M-Piezo1 | Forward | CCGTGAATGTGATTGGGCAG |
| Reverse | GGAACAGCGTGAGGAACAGA |
| M-Calpain-2 | Forward | CCCCAGTTCATTATTGGAGG |
| Reverse | AAGCTCTGATCTGGAGGCAC |
| M-STAT3 | Forward | CTTGTCTACCTCTACCCCGACAT |
| Reverse | GATCCATGTCAAACGTGAGCG |
| M-IL-33 | Forward | TCCAACTCCAAGATTTCCCCG |
| Reverse | CATGCAGTAGACATGGCAGAA |
| M-ST2L | Forward | TGTATTTGACAGTTACGGAGGGC |
| Reverse | ACTTCAGACGATCTCTTGAGACA |
| M-Collagen I | Forward | GCTCCTCTTAGGGGCCACT |
| Reverse | CCACGTCTCACCATTGGGG |
| M-TGF-β1 | Forward | CCACCTGCAAGACCATCGAC |
| Reverse | CTGGCGAGCCTTAGTTTGGAC |
| M-MMP2 | Forward | ACCTGAACACTTTCTATGGCTG |
| Reverse | CTTCCGCATGGTCTCGATG |
| M-MMP9 | Forward | CTGGACAGCCAGACACTAAAG |
| Reverse | CTCGCGGCAAGTCTTCAGAG |
| M-CTGF | Forward | GGGCCTCTTCTGCGATTTC |
| Reverse | ATCCAGGCAAGTGCATTGGTA |
| H-GAPDH | Forward | TCAACGGGAAGCCCATCA |
| Reverse | CTCGTGGTTCACACCCATCA |
| H-Piezo1 | Forward | GAGGGTGGAGGTGGAACAACCATGGTGAGCAAGGGCGCC |
| Reverse | TCCACCTCCACCCTCCTTCTCACGAGTCCAC |
| H-Calpain-2 | Forward | GTTCTGGCAATACGGCGAGT |
| Reverse | CTTCGGCTGAATGCACAAAGA |
| H-STAT3 | Forward | GGCATTCGGGAAGTATTGTCG |
| Reverse | GGTAGGCGCCTCAGTCGTATC |
| H-IL-33 | Forward | GTGACGGTGTTGATGGTAAGAT |
| Reverse | AGCTCCACAGAGTGTTCCTTG |
| ShRNA Primers | | |
| GENE | Sequence 5' --> 3' | |
| shNC | Forward | CCGGGCTTCTCCGAACGTGTCACGTCTCGAGACGTGACACGTTCGGAGAATTTTTG |
| Reverse | AATTCAAAAAGCTTCTCCGAACGTGTCACGTCTCGAGACGTGACACGTTCGGAGAA |
| shPiezo1 | Forward | CCGGTCCCTGTGCATTGATTATCCCTCTCGAGAGGGATAATCAATGCACAGGGTTTTTTG |
| Reverse | AATTCAAAAAACCCTGTGCATTGATTATCCCTCTCGAGAGGGATAATCAATGCACAGGGA |
| shCalpain2 | Forward | CCGGGCGCTCAGACACCTTCATCAACTCGAGTTGATGAAGGTGTCTGAGCGCTTTTTG |
| Reverse | GATCCAAAAAGCGCTCAGACACCTTCATCAACTCGAGTTGATGAAGGTGTCTGAGCGC |
| shSTAT3 | Forward | CCGGGCACAATCTACGAAGAATCAACTCGAGTTGATTCTTCGTAGATTGTGCTTTTTG |
| Reverse | AATTCAAAAAGCACAATCTACGAAGAATCAACTCGAGTTGATTCTTCGTAGATTGTGC |

PCR conditions

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| --- | --- | --- | --- |
| Step # | Temp °C | Time | Note |
| 1 | 94 | 2 min |  |
| 2 | 94 | 20sec |  |
| 3 | 65 | 15sec | -0.5 C per cycle decrease |
| 4 | 68 | 10sec |  |
| 5 |  |  | repeat steps 2-4 for 10cycles(Touchdown) |
| 6 | 94 | 15sec |  |
| 7 | 60 | 15sec |  |
| 8 | 72 | 10sec |  |
| 9 |  |  | repeat steps 6-8 for 28 cycles |
| 10 | 72 | 2 min |  |
| 11 | 10 |  | hold |