

A

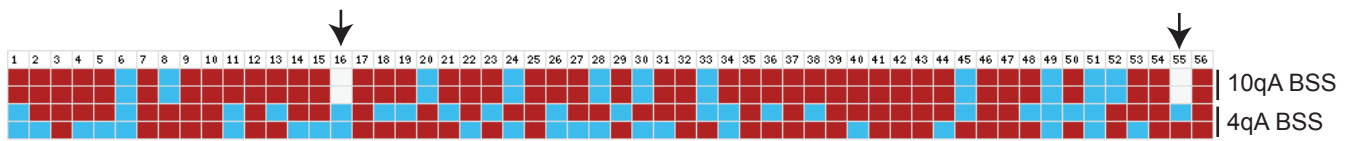
BSS 1438F
 5'-GTTTTGTTGGAGGAGTTTATAGGA CGCGGGGTTGGGACGGGGTCGGGTGGTTCGGGGTAGGGCGGTGGTTTTTTT
 TTCGCGGGGAA **TA** YG in 10A166 TTTTGGTTGGTTACGGAGGGGCGTGTTCGTTTCGTTTTTTTTATCGGGTTGATCGGTTTG
 DUX4 Exon 2 A in 10A166, 4A166
 GGATTTTTGTTTTTTAGGTTTAGGTTTCGGTGAGAGATTTTATAT **C** CGCGGAGAATTGTTATTTTTTTTTGGGTA
 DUX4 Exon 2 (CpG #16)
 TTTCGGGGATTTTAGAGTCGGTTTAGGTATTAGTAGGTTGGGTCGTTTATTGCGTACGCGCGGGTTTGCGGGTA
 GTCGTTTGGGTTGTGGGAGTAGTTCGGGTAGAGTTTTTTTTGTTTTTTTTATTAGTTTATTTTCGTCGTTTGATCG
 G in 10A166, 10A176T
 TTTTTTTTTTATTTTTATTTTTT **A** TTTTCGGAAAACGCGTCGTTTTTTGGGTTGGGTGGAGATTTTCGTTTCG
 CGAAATATCGGGTTTCGCGTAGCGTTCGGGTTTGATATCGTTTCGGCGGTTTCGTTTTTTTTTTCGTTTTTCGCGT
 7806 G 7812 G 7820 A 7827 YG
 TATCGTCGTTTCGTTTCGGGTTTTGTAGT **T** TTTTAG **T** TGTTAG **C** CGGAG **T** TTTTGGCGGTTAAAAGTAT
 (CpG #55) AAAACCGCCAATTTTCATA
 BSS 3626R
 ATTTTTGTT-3'
 TAAAACAA-5'

Non-permissive haplotype signatures
 7806 G = 4A166, 10A166, 10A180T, 10B161T
 7812 G = 4A166, 10A166, 10A180T
 7820 A = 4A160, 4C166, 10A166, 10A176T, 10A180T
 7827 G = 4A166, 4A166H, 10A176T, 10A166, 10B161T

B



C



D

BSS 4ALF
 5'-TTATTTATGAAGGGGTGGAGTTTGT TTGTTTGTGGGTTTTTATAAGGGCGGTTGGTTGGTTGGTTGGTTGTTTC
 GGGTAGGTTTTTTGGTTGTATTTGTCTAGTGTATAGTTCGGTTGAGGTGTACGGGAGTTCGTCGGTTTTTTTT
 TTGTTTCGCGTTCGTTTCGTGAAATTTTCGGTTCGGGTTTATCGCGATGGTTTTTTTCGATATTTTCGGATAGTATT
 TTTTTCGCGGAAGTTCGGGGACGAGGACGGCGACGGAGATTCGTTTGGATTTTCGAGTTAAAGCGAGGTTTTGC
 GAGTTTGTAGT **G** **G** **A** **YG**
 AAAACCGCCAATTTTCATATAAAAACAA-5'
 BSS 3626R

E

BSS +475

5'-**TTAGGAGGGAGGGAGGGAGGTAG**GGAGGTAGGGAGGAACGGAGGGAAAGATAGAGCGACGTAGGGATTGGGGG
 CGGGCGGGAGGGAGTCTGGGGACGGGGGAGGAAGGTAGGGAGGAAAAGCGGTTTTTCGGTTTTTCGGGAGTAGCG
 GGATTTTCGTTTTTTCGGGAAAACGGTTAGCGTTCGGCGCGGGTTGAGGGTTGGGTTTATAGTCGTCGCGTCGG
 TCGGCGGGGTATTATTTATTCGTTTTTCGGTTTCGTGGTTTAGGGAGTGGGCGGTTTTTTTCGGGATAAAAGATC
 GGGATTCGGGTTGTCTCGGGTTTTTATTCGCGCGGTTTATAGATCGTATATTTTTTAGGTTGAGTTTTGTAAC
 GCGGCGCGAGGTTCGATAGTTTTTCGGTTACGGAGGAGTTATACGTAGGACGACGGAGGCGTGATTTTTGGTTTTTCG
 CGTGTTTTTGTTTTTTCGTAAGGCGGTTTGTGTTTACGTTTTTTTCGGTTTTTCGAAAGGTTGGTTATGTCGATT
 GTTTGTTCGGAGTTTTGCGGGTATTCGAAATATGTAGGG**A**AGGGTGTAAGTTCGGTACGGTGTT -3'
TTCCACATTCAAGCCATGCCACAA -5'
 BSS -1036

Figure S1: BSS products. A) The 4qA BS-converted PCR product is shown. BSS primer sequences are highlighted in orange (forward) or blue (reverse). Base pair changes in the BS-converted sequence between the permissive 4A and nonpermissive 4A, 10A, and 10B haplotypes are highlighted in red (permissive) and yellow (nonpermissive). The CpG dinucleotides that would be missing from the analysis in the designated haplotypes are identified by number and are underlined. Y= C or T. B) 4qA BS PCR primers that have undergone freeze-thaw several times produce minor PCR products (*), using DNA from cells lacking permissive 4qA alleles. None of these products correspond to 4qA or 4qB and occasionally correspond to 10qA. C) Output analysis from BISMA comparing a typical 4qA BSS analysis with the rare nonpermissive 10A166 or 4A166 haplotype BSS outputs that may appear, as in B, above. These are readily recognized by the absence of CpGs #16 and 55 (black arrows) and eliminated from analysis. D) The 4qA-L BS-converted PCR product is shown. BSS primers are highlighted in orange (forward) or blue (reverse). Base pair changes between 4A -L and nonpermissive 4A and 10A haplotypes are highlighted in red (permissive) and yellow (nonpermissive). E) The DUX4 5' BS-converted PCR product. BSS primers are highlighted in orange (forward) or blue (reverse), with the 4q-specific D4Z4 polymorphism in highlighted in red and the 10q D4Z4 polymorphism highlighted in yellow.