## **1** Supplemental file 2. Assay interpretation

2 Comparison of the amplification curve generated by the ancestral primer (blue,

3 Diagram 1) and the derived primer (red, Diagram 2). Since the ancestral primers

4 matches the SNP at the ultimate 3' end nucleotide the Cq value will be significantly

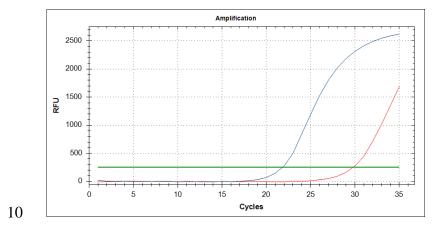
5 lower compared to the non-complementary derived primer. Thus, the template

6 exhibits the ancestral SNP state in this example. The products can be distinguished by

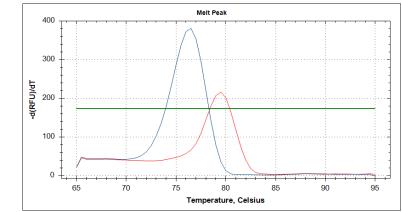
7 comparing their Tm-values (Diagram 2).



## 9 Diagram 1



## 11 Diagram 2



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