| Gene symbol | Kcna1 <sup>m1Kyo</sup> |
|-------------|------------------------|
|             |                        |

| primer        | Sequence(5'-3')                                     | Size  | target gene | accession |
|---------------|---|-------|-------------|-----------|
| Kcna1_Ex1-2-F | GCCATTGTATCCGTCATGGT                                | 575bp | Kcna1       | NC_005103 |
| Kcna1_Ex1-2-R | TCGGGGATACTGGAGAAGTG                                | 373pb |             |           |
| comment       | Kcna1 mutation (T925A) is determined by sequencing. |       |             |           |
|               |   |       |             |           |
|               |   |       |             |           |
|               |   |       |             |           |

| PCR condition  | า   |  |          |        |           |       |                  |  |
|--|-----|--|----------|--------|-----------|-------|------------------|--|
| Taq polymera   | se  | BIOTAQTM HS DNA Polymerase (Bioline Reagents Ltd., UK) |          |        |           |       |                  |  |
| Thermal cycle  | er  | PC-808(ASTEC)  |          |        |           |       |                  |  |
| PCR buffer   |     | Ampdirect Plus(Shimadzu.Co)                            |          |        |           |       |                  |  |
|  |     | first denature   | denature | anneal | extension | cycle | final extension  |  |
| PCR  | °C  | 94   | 94       | 60     | 72        | 40    | 72               |  |
|  | min | 3  | 0.5      | 1      | 0.75      |       | 3                |  |
| commentPCR method; Blood was applied to FTA®card (GE Healthcare UK Ltd.,UK) and dried. 1.5mmFTA disc was removed from the bloody-stanined region on FTA®card. Untreated sample<br>discs were placed directly in 15 $\mu$ L PCR mixture containing 1 × Ampdirect®Plus, 0.2 $\mu$ M eac<br>primer and 0.4 units of BIOTAQTM HS DNA Polymerase. |     |  |          |        |           |       | Untreated sample |  |

