

附錄二

以下為各引子與 DNA 模板 PCR 反應之產物片段大小

(1) Mutation: G382D

DNA template: Plasmid DNA (GI:1458227)

Forward primer: THU-MYH-5/ 5'-CCAACACTGGACAGTGCCACCT-3'

Reverse primer(WT): THU-MYH-6/ 5'-ACAGTCCTGCCAGCAGAC-3'

Reverse primer(MT): THU-MYH-7/ 5'-ACAGTCCTGCCAGCAGA **T**-3'

PCR production size: 204 bp

901 gaacagctct tagcctcagg gagcctgtcg ggcagtcctg acgtggagga gtgtgctccc
961 **aacactggac agtgccacct** gtgcctgcct cctcgggagc cctgggacca gaccctggga
1021 gtggtcaact tccccagaaa ggccagccgc aagcccccca gggaggagag ctctgccacc
1081 tgtgttctgg aacagcctgg ggccttggg gcccaaattc tgctggtgca gaggcccaac
1141 tcag**Gtctgc tggcaggact gt**gggagttc cegtccgtga cctgggagcc ctcagagcag

(2) Mutation: Y165C

DNA template: Plasmid DNA (GI:1458227)

Forward primer: THU-MYH-1/ 5'-CTACTATACCGGATGGATGC-3'

Reverse primer(WT): THU-MYH-2/ 5'-CGCCGGCCACGAGAATAGT-3'

Reverse primer(MT): THU-MYH-3/ 5'-CGCCGGCCACGAGAATAGC-3'

PCR production size: 114 bp

361 gtcatgctgc agcagacca ggttgccact gtgatcaact **actataccgg atggatgca**g
421 aagtggccta cactgcagga cctggccagt gcttccctgg aggaggtgaa tcaactctgg
481 gctggcctgg gct**Actattc tcgtggccgg cg**gctgcagg agggagctcg gaaggtggtg

(3) Mutation: V232F

DNA template: Plasmid DNA (GI:1458227)

Forward primer: THU-MYH-1/ 5'-CTACTATACCGGATGGATGC-3'

Reverse primer(WT): THU-MYH-12/ 5'-TCAGCACCAATGGCTCGGAC-3'

Reverse primer(MT): THU-MYH-13/ 5'-TCAGCACCAATGGCTCGGAA-3'

PCR production size: 315 bp

```
361 gtcattgctgc agcagaccca ggttgccact gtgatcaact actataccgg atggatgcag
421 aagtggccta cactgcagga cctggccagt gcttccctgg aggaggtgaa tcaactctgg
481 gctggcctgg gctactattc tcgtggccgg cggctgcagg agggagctcg gaaggtggta
541 gaggagctag gggggccacat gccacgtaca gcagagaccc tgcagcagct cctgcctggc
601 gtggggcgct acacagctgg ggccattgcc tctatcgctt ttggccaggc aaccggtgtg
661 gtggatggca acgtagcacg ggtgctgtgc cgtGtccgag ccattggtgc tgatcccagc
```

(4) Mutation: R227W

DNA template: Plasmid DNA (GI:1458227)

Forward primer: THU-MYH-1/ 5'-CTACTATACCGGATGGATGC-3'

Reverse primer(WT): THU-MYH-10/5'-CTCGGACACGGCACAGCACCCG-3'

Reverse primer(MT): THU-MYH-11/ 5'-CTCGGACACGGCACAGCACCCA-3'

PCR production size: 302 bp

```
361 gtcattgctgc agcagaccca ggttgccact gtgatcaact actataccgg atggatgcag
421 aagtggccta cactgcagga cctggccagt gcttccctgg aggaggtgaa tcaactctgg
481 gctggcctgg gctactattc tcgtggccgg cggctgcagg agggagctcg gaaggtggta
541 gaggagctag gggggccacat gccacgtaca gcagagaccc tgcagcagct cctgcctggc
601 gtggggcgct acacagctgg ggccattgcc tctatcgctt ttggccaggc aaccggtgtg
661 gtggatggca acgtagcaCg ggtgctgtgc cgtgtccgag ccattggtgc tgatcccagc
```

(5) Mutation: G382D

DNA template: Genomic DNA (GI:21902513)

Forward primer: THU-MYH-5/ 5'-CCAACACTGGACAGTGCCACCT-3'

Reverse primer(WT): THU-MYH-6/ 5'-ACAGTCCTGCCAGCAGAC-3'

Reverse primer(MT): THU-MYH-7/5'-ACAGTCCTGCCAGCAGA T-3'

PCR production size: 308 bp

```
9301 ctaaagccct cttggcttga gtagggttcg gggatctccg ttcccagctc ccaacactgg
9361 acagtgccac ctgtgcctgc ctccctcgga gccctgggac cagaccctgg gagtgggtcaa
9421 cttccccaga aaggccagcc gcaagccccc cagggaggag agctctgcca cctgtgttct
9481 ggaacagcct ggggcccttg gggcccaaat tctgctggtg cagaggccca actcaggtac
9541 ctggatactg ggcgtggagg gcagtgccat gagtaacaag agagaatgga gggaaatcggc
9601 agctgaggcc tgaccctcgc ctggctgccc tcctctcag Gtctgctggc aggactgtgg
```

(6) Mutation: Y165C

DNA template: Genomic DNA (GI:21902513)

Forward primer: THU-MYH-1/ 5'-CTACTATACCGGATGGATGC-3'

Reverse primer(WT): THU-MYH-2/ 5'-CGCCGGCCACGAGAATAGT-3'

Reverse primer(MT): THU-MYH-3/ 5'-CGCCGGCCACGAGAATAGC-3'

PCR production size: 325 bp

```
8041 gtcatgctgc agcagaccca ggttgccact gtgatcaact actataccgg atggatgcag
8101 gtgactccag gggaggaagg gaagggcat gggtcagacc ccagatgaga gcctctactt
8161 tgggggtgggt gtagagaagg cttcctctac caccttcacc cttgaccttg tctctttctg
8221 cctgcctgtg gctatagaag tggcctacac tgcaggacct ggccagtgct tcctggagg
8281 tgagagccac cctagggtag gggaaatagg aacgatagag ggactgacgg gtgatctctt
8341 tgacctctga tctaccac aggaggtgaa tcaactctgg gctggcctgg gctActatctc
8401 tcgtggccgg cggctgcagg agggagctcg gaaggtaagg ggatggcagg agggtaggaa
```

(7) Mutation: V232F

DNA template: Genomic DNA (GI:21902513)

Forward primer: THU-MYH-1/ 5'-CTACTATACCGGATGGATGC-3'

Reverse primer(WT): THU-MYH-12/ 5'-TCAGCACCAATGGCTCGGAC-3'

Reverse primer(MT): THU-MYH-13/ 5'-TCAGCACCAATGGCTCGGAA-3'

PCR production size: 695 bp

8041 **gtcatgctgc agcagaccca ggttgccact gtgatcaact actataccgg atggatgcag**
8101 gtgactccag gggaggaagg gaaggggtcat gggtcagacc ccagatgaga gcctctactt
8161 tgggggtgggt gtagagaagg cttcctctac caccttcacc cttgaccttg tctctttctg
8221 cctgcctgtg gctataga**aag tggcctacac tgcaggacct ggccagtgct tccctggagg**
8281 **tg**agagccac cctagggtag gggaaatagg aacgatagag ggactgacgg gtgatctctt
8341 tgacctctga tcctaccac aggaggtg**aa tcaactctgg gctggcctgg gctactattc**
8401 **tcgtggccgg cggctgcagg agggagctcg gaag**gtaagg ggatggcagg agggtaggaa
8461 cccaggagtc ttgggtgtct tataatcttg agtcttgca tccaatcagg **tg**gtagagga
8521 **gctagggggc cacatgccac gtacagcaga gaccctgcag cagctcctgc ctggcgtggg**
8581 **gc**gctacaca gctggggcca ttgcctctat **cg**cctttggc **cag**gtgatct cacagcccac
8641 ccccactttg tgcgtgccca gctccttcc tcccagccca ggctaactct ttggcccctc
8701 tgtgccag**gc aaccgggtgtg gtggatggca acgtagcacg ggtgctgtgc cgt**Gtccgag
8761 ccattggtgc tgatcccagc agcacccttg tttcccagca gctctggtag gatggtgggg

(8) Mutation: G382D (Sequencing)

DNA template: Genomic DNA (GI:21902513)

Forward primer: THU-MYH-5/ 5'-CCAACACTGGACAGTGCCACCT-3'

Reverse primer: THU-MYH-8/ 5'-CCTTGCTGGGCTACTATTCT-3'

PCR production size: 359 bp

```
9301 ctaaagccct cttggcctga gtagggttcg gggatctccg ttcccagctc ccaacactgg
9361 acagtgccac ctgtgcctgc ctccctcgga gccctgggac cagaccctgg gagtgggtcaa
9421 cttccccaga aaggccagcc gcaagccccc cagggaggag agctctgcc aactgtgtct
9481 ggaacagcct ggggcccttg gggcccaaat tctgctggtg cagaggcca actcaggtac
9541 ctggatactg ggcgtggagg gcagtggcat gagtaacaag agagaatgga gggaaatcggc
9601 agctgaggcc tgacccttgc ctggctgccc tccctctcag Gtctgctggc aggactgtgg
9661 gagttcccgt ccgtgacctg ggagccctca gacgagcttc agcgcaaggc cctgctgcag
```

(9) Mutation: Y165C and V232F (Sequencing)

DNA template: Genomic DNA (GI:21902513)

Forward primer: THU-MYH-53/ 5'-AAGTGGCCTACACTGCAGGA -3'

Reverse primer: THU-MYH-54/ 5'-CGCTGTGGGTACACACTGT -3'

PCR production size: 742 bp

```
8221 cctgcctgtg gctatagaag tggcctacac tgcaggacct ggccagtgct tcctggagg
8281 tgagagccac cctagggtag gggaaatagg aacgatagag ggactgacgg gtgatctctt
8341 tgacctctga tccatccac aggaggtgaa tcaactctgg gctggcctgg gctActattc
8401 tcgtggcccg cggctgcagg agggagctcg gaaggtaagg ggatggcagg agggtaggaa
8461 cccaggagtc ttgggtgtct tataatcttg agtcttgca tccaatcagg tggtagagga
8521 gctagggggc cacatgccac gtacagcaga gaccctgcag cagctcctgc ctggcgtggg
8581 gcgctacaca gctggggcca ttgcctctat cgcctttggc cagggtgatct cacagccac
8641 ccccactttg tgcgtgcccc gcctccttcc tccagcccc ggctaactct ttggccccctc
8701 tgtgccaggc aaccgggtgtg gtggatggca acgtagcacg ggtgctgtgc cgtGtccgag
8761 ccattgggtg tgatcccagc agcacccttg tttcccagca gctctggtag gatgttgggg
8821 taacaagggg gcttcagggg tgtctgcaaa ggagctctgc ttcacagcag tgttcccttc
8881 tttttagggg ctagcccagc agctgggtgga cccagcccgg ccaggagatt tcaaccaagc
8941 agccatggag ctaggggcca cagtggtgtac cccacagcgc ccactgtgca gccagtgccc
```