APPENDICES

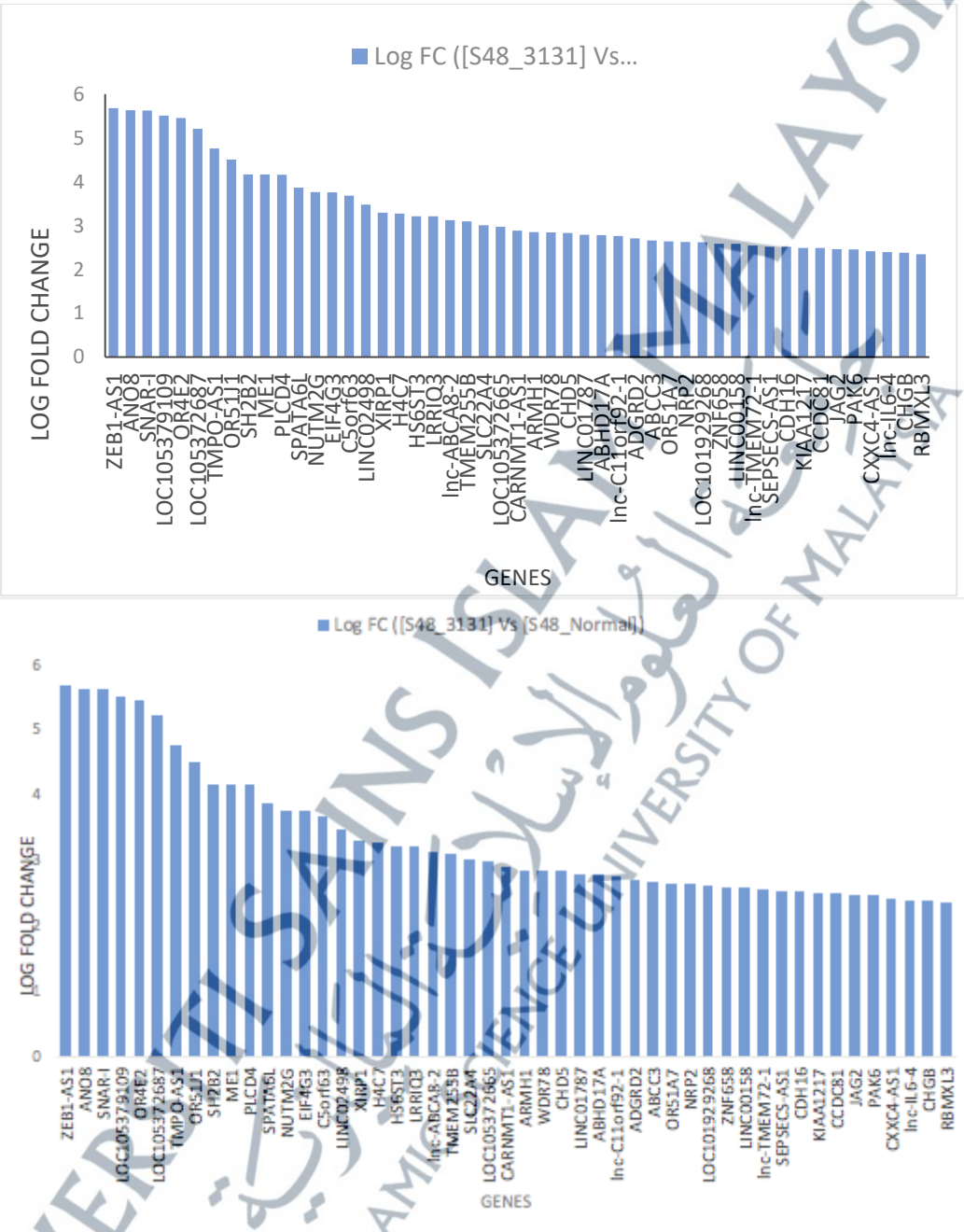
Appendix 1: List of MiRNA target genes.

List of target genes:

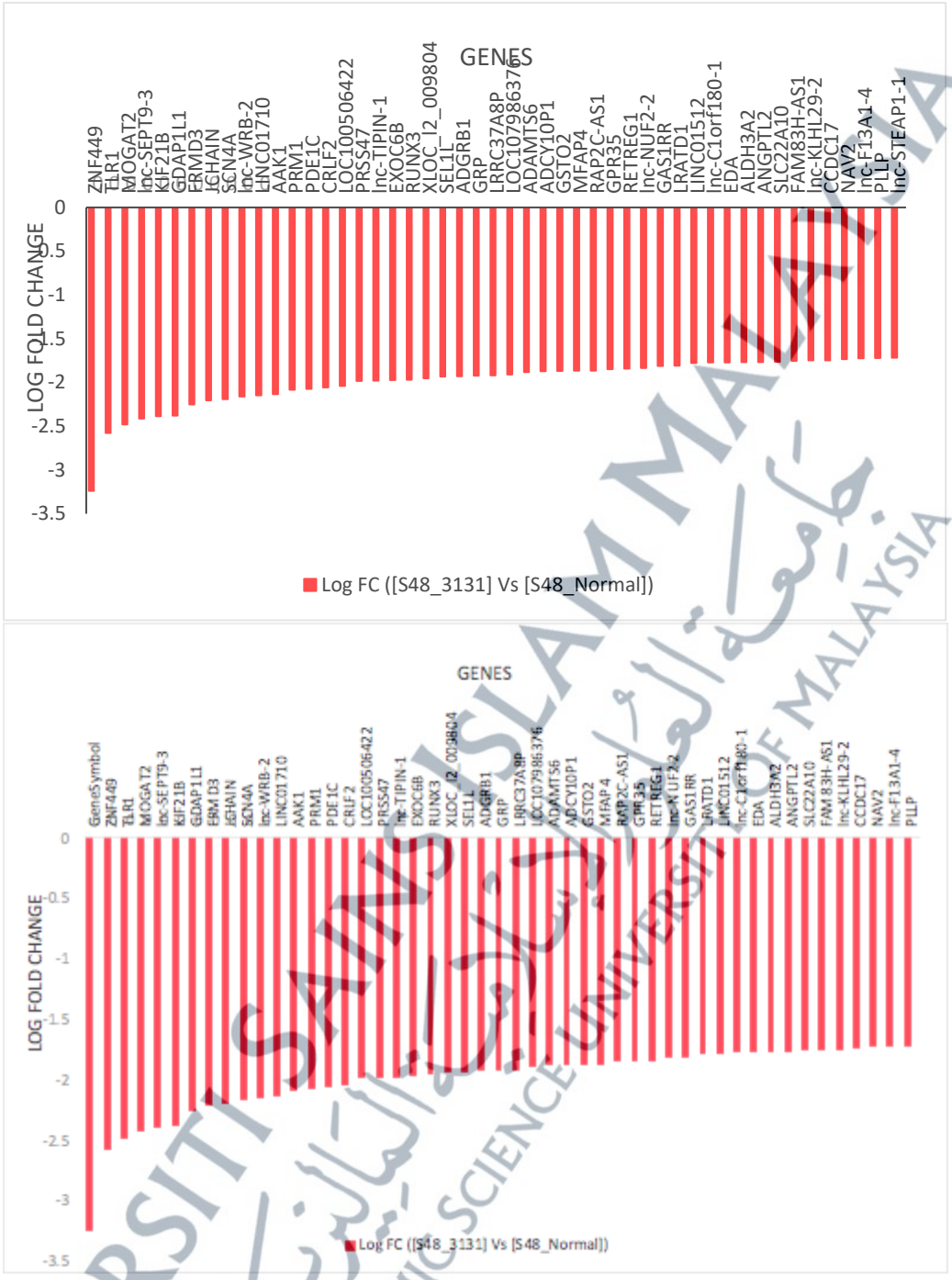
1) Full list of human miRNAs, hsa-miR-891a-3p, hsa-miR-3131, hsa-miR-6847-5p, hsa-miR-1185-1-3p, hsa-miR-1185-2-3p target genes: https://docs.google.com/spreadsheets/d/1-9r8nCmbfbIlkLq7WKgQ1tmXNOgf2ghh/edit?gid=951104886#gid=951104886

2) Full list of plant miRNAs, cpa-miR8154, mtr-miR2654, sof-miR159e, osa-miR1858a/b & osa-miR1858b target genes: https://docs.google.com/spreadsheets/d/1y1mps2-WINzGzqkzX6XyUAXO8FAvrQ1L/edit?gid=463210919#gid=463210919

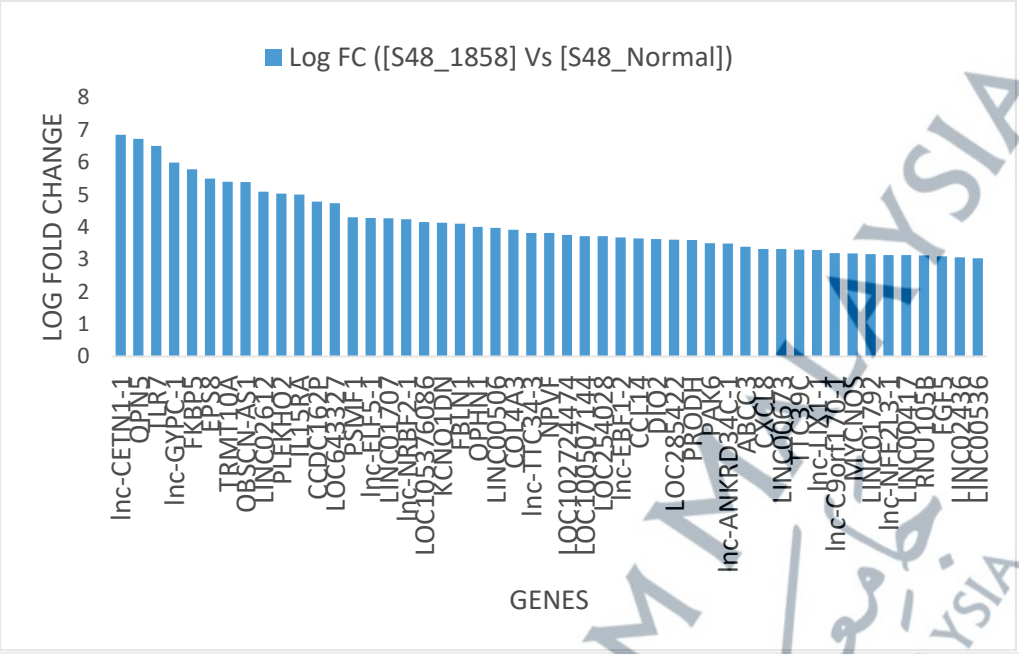
Appendix 2: Upregulated and downregulated microarray genes

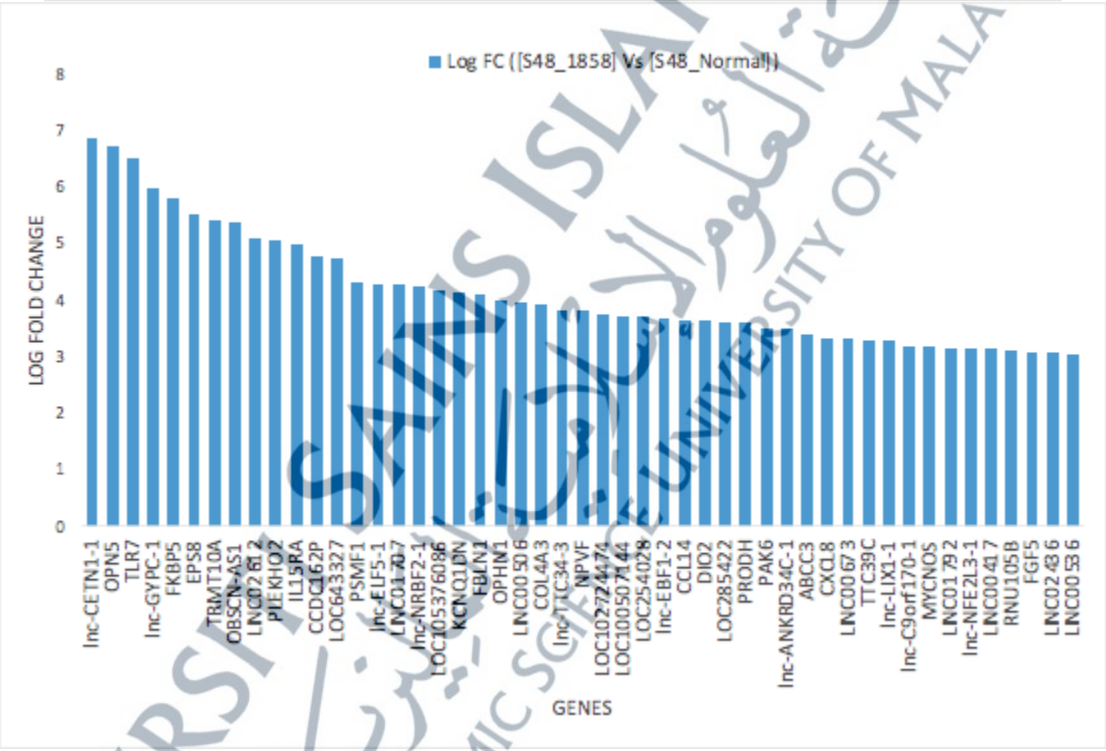


Log fold change of fifty upregulated genes from miR-3131 group of k562-s

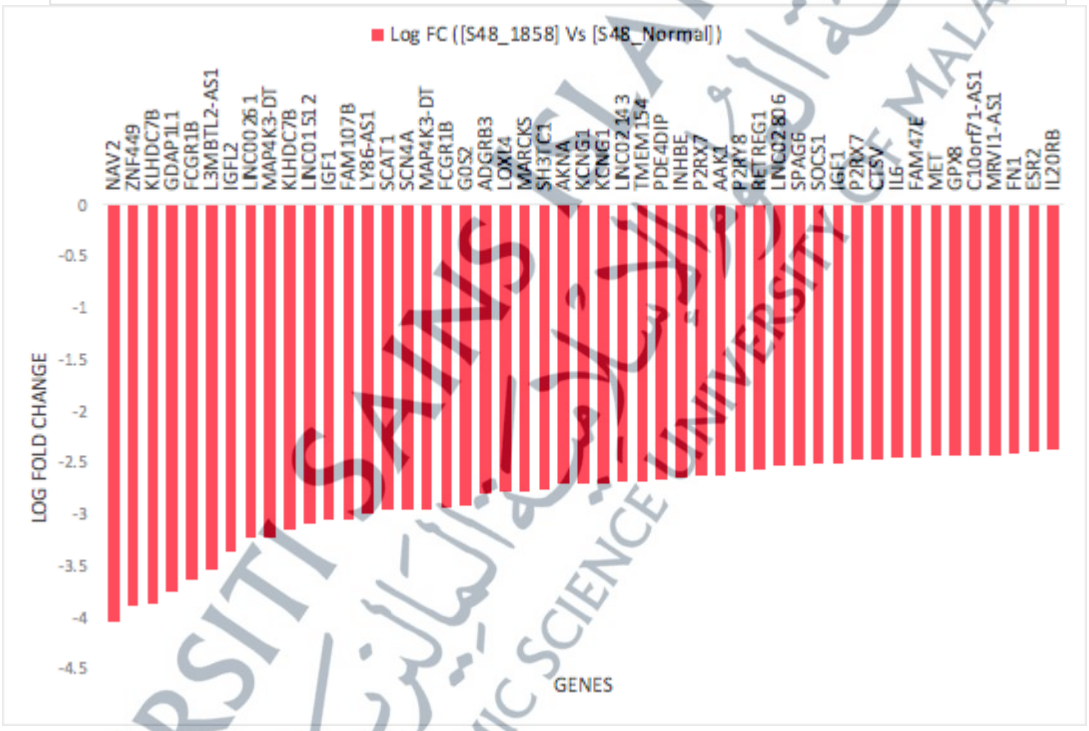


Log fold change of fifty downregulated genes from miR-3131 group of k562-s

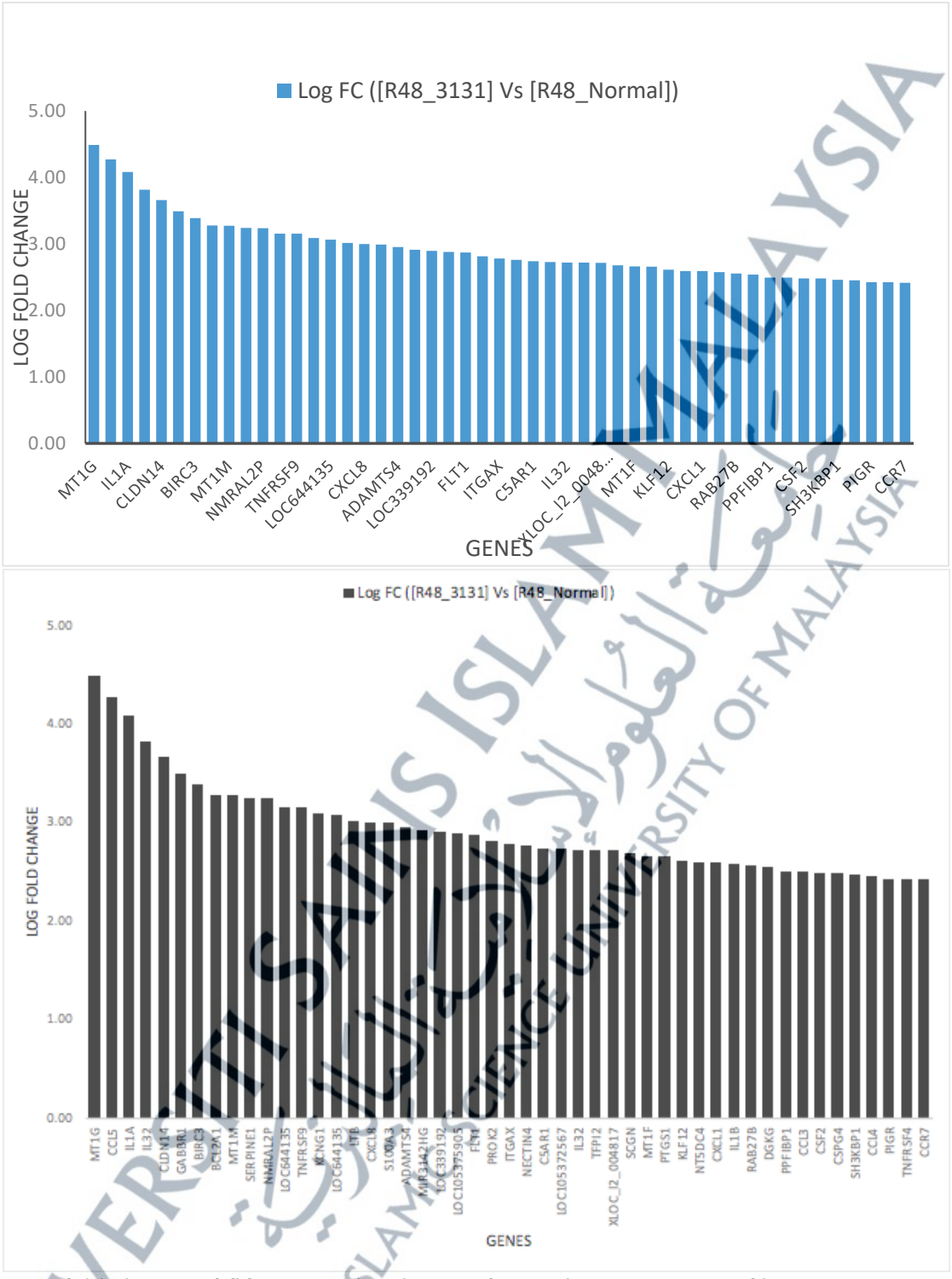




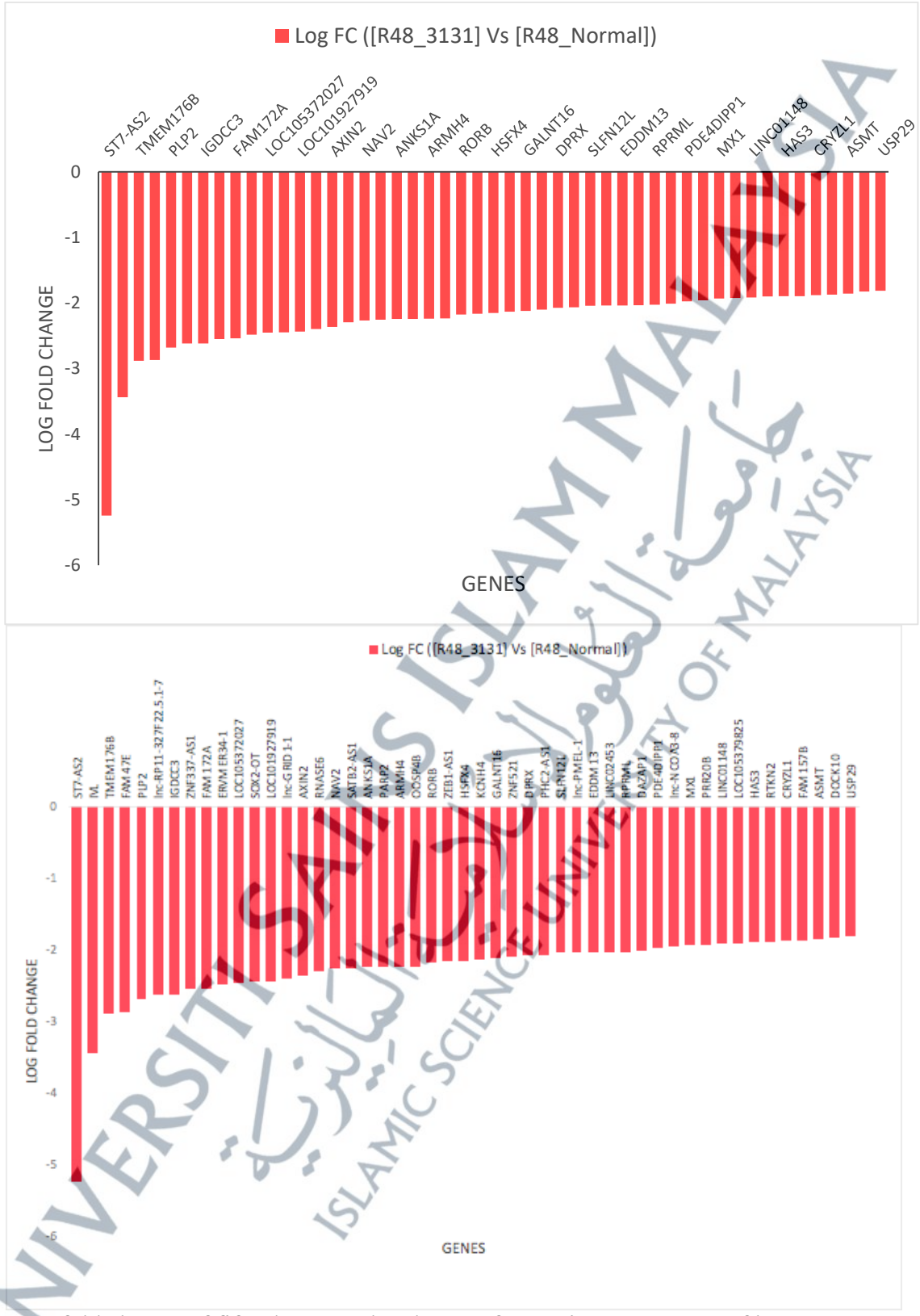
Log fold change of fifty upregulated genes from miR-1858a/b group of k562-s



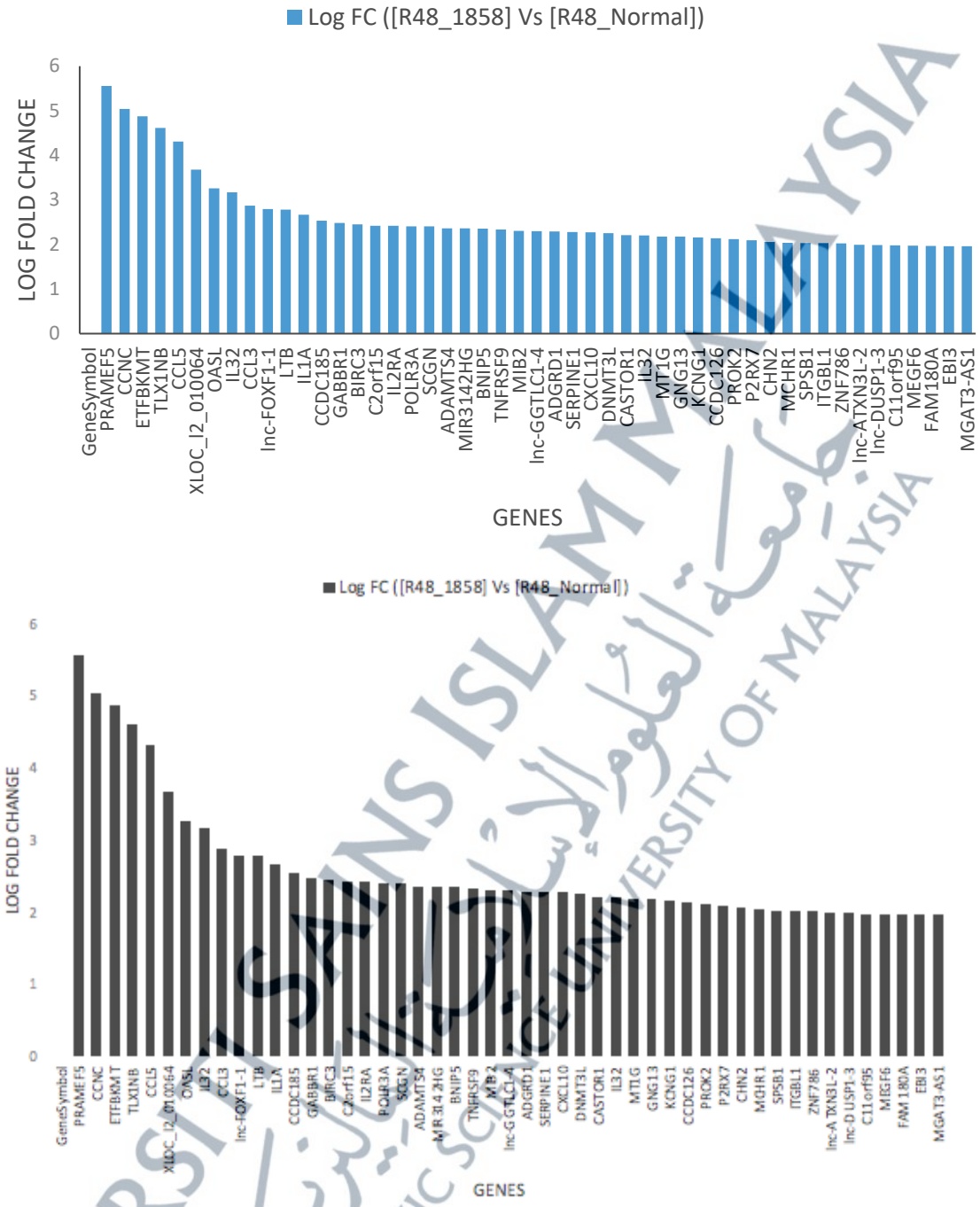
Log fold change of fifty downregulated genes from miR-1858a/b group of k562-s



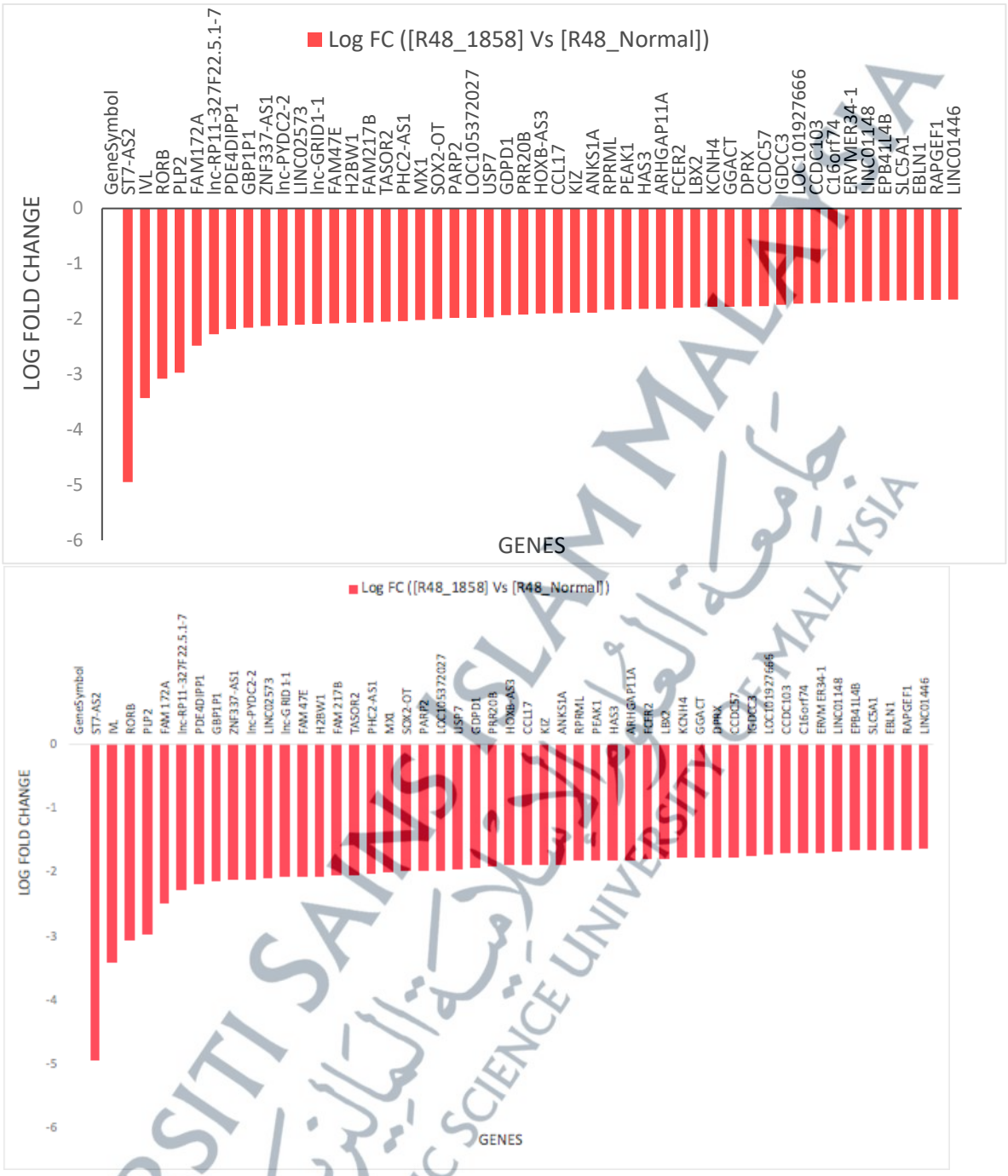
Log fold change of fifty upregulated genes from miR-3131 group of k562-r



Log fold change of fifty downregulated genes from miR-3131 group of k562-r



Log fold change of fifty upregulated genes from miR-1858a/b group of k562-r



Log fold change of fifty downregulated genes from miR-1858a/b group of k562-r

Appendix 3: Primer design for microarray gene validation

Primer design

IL6

```
PRIMER PICKING RESULTS FOR

Template masking selected
Using library: homo_sapiens with failure rate: 0.1
No mispriming library specified
Using 1-based sequence positions
OLIGO            start  len      tm      gc%  any_th  3' th hairpin
seq
LEFT PRIMER        190   20   60.32    60.00    0.00   0.00    0.00
CACAGACAGCCACTCACCTC
RIGHT PRIMER       314   20   59.33    50.00    0.00   0.00    0.00
GCCTCTTTGCTGCTTTCACA
SEQUENCE SIZE: 1127
INCLUDED REGION SIZE: 1127

PRODUCT SIZE: 125, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00
TARGETS (start len

    1 ATTCTGCCCTCGAGCCCACCGGGAACGAAAGAGAAGCTCTATCTCCCCTCCAGGAGCCCA

   61 GCTATGAACTCCTTCTCCACAAGCGCCTTCGGTCCAGTTGCCTTCTCCCTGGGGCTGCTC

  121 CTGGTGTTGCCTGCTGCCTTCCCTGCCCCAGTACCCCCAGGAGAAGATTCCAAAGATGTA

  181 GCCGCCCCACACAGACAGCCACTCACCTCTTCAGAACGAATTGACAAACAAATTCGGTAC
                >>>>>>>>>>>>>>>>>>>>                          *****

  241 ATCCTCGACGGCATCTCAGCCCTGAGAAAGGAGACATGTAACAAGAGTAACATGTGTGAA
      ********                                                <<<<<<

  301 AGCAGCAAAGAGGCACTGGCAGAAAACAACCTGAACCTTCCAAAGATGGCTGAAAAAGAT
      <<<<<<<<<<<<<<

  361 GGATGCTTCCAATCTGGATTCAATGAGGAGACTTGCCTGGTGAAAATCATCACTGGTCTT

  421 TTGGAGTTTGAGGTATACCTAGAGTACCTCCAGAACAGATTTGAGAGTAGTGAGGAACAA

  481 GCCAGAGCTGTGCAGATGAGTACAAAAGTCCTGATCCAGTTCCTGCAGAAAAAGGCAAAG

  541 AATCTAGATGCAATAACCACCCCTGACCCAACCACAAATGCCAGCCTGCTGACGAAGCTG

  601 CAGGCACAGAACCAGTGGCTGCAGGACATGACAACTCATCTCATTCTGCGCAGCTTTAAG

  661 GAGTTCCTGCAGTCCAGCCTGAGGGCTCTTCGGCAAATGTAGCATGGGCACCTCAGATTG
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 721 TTGTTGTTAATGGGCATTCCTTCTTCTGGTCAGAAACCTGTCCACTGGGCACAGAACTTA

 781 TGTTGTTCTCTATGGAGAACTAAAAGTATGAGCGTTAGGACACTATTTTAATTATTTTTA

 841 ATTTATTAATATTTAAATATGTGAAGCTGAGTTAATTTATGTAAGTCATATTTATATTTT

 901 TAAGAAGTACCACTTGAAACATTTTATGTATTAGTTTTGAAATAATAATGGAAAGTGGCT

 961 ATGCAGTTTGAATATCCTTTGTTTCAGAGCCAGATCATTTCTTGGAAAGTGTAGGCTTAC

1021 CTCAAATAAATGGCTAACTTATACATATTTTTAAAGAAATATTTATATTGTATTTATATA

1081 ATGTATAAATGGTTTTTATACCAATAAATGGCATTTTAAAAAATTCA
```

```
KEYS (in order of precedence):
****** target
>>>>>> left primer
<<<<<< right primer

ADDITIONAL OLIGOS
                    start  len      tm      gc%   any th   3' th hairpin seq

 1 LEFT PRIMER        168   20   60.39   55.00     0.00    0.00    0.00
TTCCAAAGATGTAGCCGCCC
   RIGHT PRIMER       317   20   59.04   50.00     0.00    0.00    0.00
AGTGCCTCTTTGCTGCTTTC
   PRODUCT SIZE: 150, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00

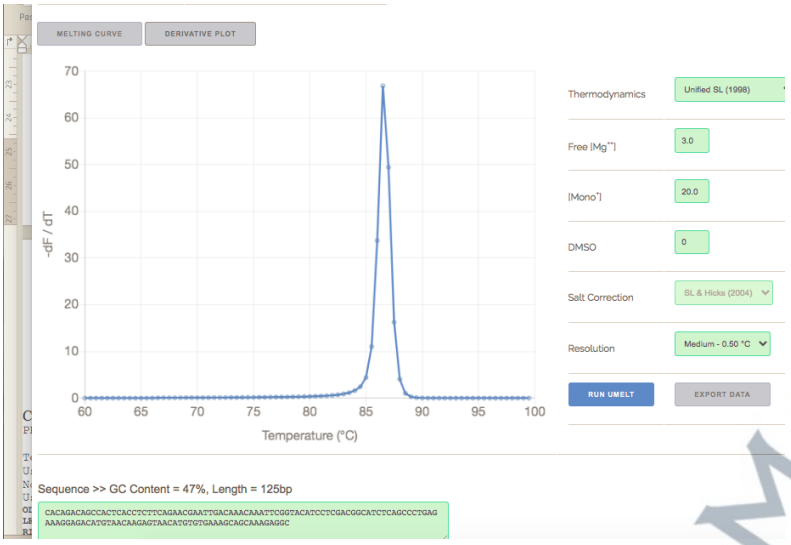
 2 LEFT PRIMER        194   20   59.32   55.00     0.00    0.00    0.00
GACAGCCACTCACCTCTTCA
   RIGHT PRIMER       319   19   60.60   57.89     0.00    0.00    0.00
CCAGTGCCTCTTTGCTGCT
   PRODUCT SIZE: 126, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00

 3 LEFT PRIMER        187   20   60.88   60.00     0.00    0.00    0.00
CCACACAGACAGCCACTCAC
   RIGHT PRIMER       334   21   58.98   47.62     0.00    0.00    0.00
TCAGGTTGTTTTCTGCCAGTG
   PRODUCT SIZE: 148, PAIR ANY_TH COMPL: 7.45, PAIR 3'_TH COMPL: 7.45

 4 LEFT PRIMER        196   21   59.65   52.38     0.00    0.00    0.00
CAGCCACTCACCTCTTCAGAA
   RIGHT PRIMER       337   21   59.25   47.62     0.00    0.00    0.00
GGTTCAGGTTGTTTTCTGCCA
   PRODUCT SIZE: 142, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.47
```

IDT OligoAnalyzer

	Forward	Reverse
Hairpin **ΔG (kcal.mole-1)**	1.29	0.34
Self-dimer **ΔG (kcal.mole-1)**	-3.14	-3.14
Hetero-dimer **ΔG (kcal.mole-1)**	-6.69	



NCBI BLAST- PASSED

CSF1

PRIMER PICKING RESULTS FOR

Template masking selected
Using library: homo_sapiens with failure rate: 0.1
No mispriming library specified
Using 1-based sequence positions

OLIGO	start	len	tm	gc%	any th	3' th	hairpin	seq
LEFT PRIMER	**202**	**18**	**60.12**	**66.67**	**0.00**	**0.00**	**0.00**	**CTGCCCTCCCACGACATG**
RIGHT PRIMER	**276**	**20**	**59.74**	**55.00**	**0.00**	**0.00**	**0.00**	**TCCTCGGTGATACTCCTGCT**

SEQUENCE SIZE: 3994
INCLUDED REGION SIZE: 3994

PRODUCT SIZE: 75, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00

```
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   61 CCGCATCCCAGGACAGCGGTGCGGCCCTCGGCCGGGGCGCCCACTCCGCAGCAGCCAGCG

  121 AGCGAGCGAGCGAGCGAGGGCGGCCGACGCGCCCGGCCGGGACCCAGCTGCCCGTATGAC

  181 CGCGCCGGGCGCCGCCGGGCGCTGCCCTCCCACGACATGGCTGGGCTCCCTGCTGTTGTT
                          >>>>>>>>>>>>>>>>>>

  241 GGTCTGTCTCCTGGCGAGCAGGAGTATCACCGAGGAGGTGTCGGAGTACTGTAGCCACAT
                      <<<<<<<<<<<<<<<<<<<

  301 GATTGGGAGTGGACACCTGCAGTCTCTGCAGCGGCTGATTGACAGTCAGATGGAGACCTC

  361 GTGCCAAATTACATTTGAGTTTGTAGACCAGGAACAGTTGAAAGATCCAGTGTGCTACCT

  421 TAAGAAGGCATTTCTCCTGGTACAAGACATAATGGAGGACACCATGCGCTTCAGAGATAA
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 481 CACCCCCAATGCCATCGCCATTGTGCAGCTGCAGGAACTCTCTTTGAGGCTGAAGAGCTG
 541 CTTCACCAAGGATTATGAAGAGCATGACAAGGCCTGCGTCCGAACTTTCTATGAGACACC
 601 TCTCCAGTTGCTGGAGAAGGTCAAGAATGTCTTTAATGAAACAAAGAATCTCCTTGACAA
 661 GGACTGGAATATTTTCAGCAAGAACTGCAACAACAGCTTTGCTGAATGCTCCAGCCAAGA
 721 TGTGGTGACCAAGCCTGATTGCAACTGCCTGTACCCCAAAGCCATCCCTAGCAGTGACCC
 781 GGCCTCTGTCTCCCCTCATCAGCCCCTCGCCCCCTCCATGGCCCCTGTGGCTGGCTTGAC
 841 CTGGGAGGACTCTGAGGGAACTGAGGGCAGCTCCCTCTTGCCTGGTGAGCAGCCCCTGCA
 901 CACAGTGGATCCAGGCAGTGCCAAGCAGCGGCCACCCAGGAGCACCTGCCAGAGCTTTGA
 961 GCCGCCAGAGACCCCAGTTGTCAAGGACAGCACCATCGGTGGCTCACCACAGCCTCGCCC
1021 CTCTGTCGGGGCCTTCAACCCCGGGATGGAGGATATTCTTGACTCTGCAATGGGCACTAA
1081 TTGGGTCCCAGAAGAAGCCTCTGGAGAGGCCAGTGAGATTCCCGTACCCCAAGGGACAGA
1141 GCTTTCCCCCTCCAGGCCAGGAGGGGGCAGCATGCAGACAGAGCCCGCCAGACCCAGCAA
1201 CTTCCTCTCAGCATCTTCTCCACTCCCTGCATCAGCAAAGGGCCAACAGCCGGCAGATGT
1261 AACTGGTACCGCCTTGCCCAGGGTGGGCCCCGTGAGGCCCACTGGCCAGGACTGGAATCA
1321 CACCCCCCAGAAGACAGACCATCCATCTGCCCTGCTCAGAGACCCCCCGGAGCCAGGCTC
1381 TCCCAGGATCTCATCACTGCGCCCCCAGGGCCTCAGCAACCCCTCCACCCTCTCTGCTCA
1441 GCCACAGCTTTCCAGAAGCCACTCCTCGGGCAGCGTGCTGCCCCTTGGGGAGCTGGAGGG
1501 CAGGAGGAGCACCAGGGATCGGAGGAGCCCCGCAGAGCCAGAAGGAGGACCAGCAAGTGA
1561 AGGGGCAGCCAGGCCCCTGCCCCGTTTTAACTCCGTTCCTTTGACTGACACAGGCCATGA
1621 GAGGCAGTCCGAGGGATCCTTCAGCCCGCAGCTCCAGGAGTCTGTCTTCCACCTGCTGGT
```

1681 GCCCAGTGTCATCCTGGTCTTGCTGGCCGTCGGAGGCCTCTTGTTCTACAGGTGGAGGCG

1741 GCGGAGCCATCAAGAGCCTCAGAGAGCGGATTCTCCCTTGGAGCAACCAGAGGGCAGCCC

1801 CCTGACTCAGGATGACAGACAGGTGGAACTGCCAGTGTAGAGGGAATTCTAAGCTGGACG

1861 CACAGAACAGTCTCTCCGTGGGAGGAGACATTATGGGGCGTCCACCACCACCCCTCCCTG

1921 GCCATCCTCCTGGAATGTGGTCTGCCCTCCACCAGAGCTCCTGCCTGCCAGGACTGGACC

1981 AGAGCAGCCAGGCTGGGGCCCCTCTGTCTCAACCCGCAGACCCTTGACTGAATGAGAGAG

2041 GCCAGAGGATGCTCCCCATGCTGCCACTATTTATTGTGAGCCCTGGAGGCTCCCATGTGC

2101 TTGAGGAAGGCTGGTGAGCCCGGCTCAGGACCCTCTTCCCTCAGGGGCTGCACCCTCCTC

2161 TCACTCCCTTCCATGCCGGAACCCAGGCCAGGGACCCACCGGCCTGTGGTTTGTGGGAAA

2221 GCAGGGTGGACGCTGAGGAGTGAAAGAACCCTGCACCCAGAGGGCCTGCCTGGTGCCAAG

2281 GTATCCCAGCCTGGACAGGCATGGACCTGTCTCCAGAGAGAGGAGCCTGAAGTTCGTGGG

2341 GCGGGACAGCGTCGGCCTGATTTCCCGTAAAGGTGTGCAGCCTGAGAGACGGGAAGAGGA

2401 GGCCTCTGGACCTGCTGGTCTGCACTGACAGCCTGAAGGGTCTACACCCTCGGCTCACCT

2461 AAGTGCCCTGTGCTGGTTGCCAGGCGCAGACGGGGAGGCCAGCCCTGCCCTCAGGACCTGC

2521 CTGACCTGCCAGTGATGCCAAGAGGGGGATCAAGCACTGGCCTCTGCCCCTCCTCCTTCC

2581 AGCACCTGCCAGAGCTTCTCCAGGAGGCCAAGCAGAGGCTCCCCTCATGAAGGAAGCCAT

2641 TGCACTGTGAACACTGTACCTGCCTGCTGAACAGCCTGCCCCCGTCCATCCATGAGCCAG

2701 CATCCGTCCGTCCTCCACTCTCCAGCCTCTCCCCAGCCTCCTGCACTGAGCTGGCCTCAC

2761 CAGTCGACTGAGGGAGCCCCTCAGCCCTGACCTTCTCCTGACCTGGCCTTTGACTCCCCG

2821 GAGTGGAGTGGGGTGGGAGAACCTCCTGGGCCGCCAGCCAGAGCCGGTCTTTAGGCTGTG

2881 TTGTTCGCCCAGGTTTCTGCATCTTGCACTTTGACATTCCCAAGAGGGAAGGGACTAGTG

2941 GGAGAGAGCAAGGGAGGGGAGGGCACAGACAGAGAGGCTACAGGGCGAGCTCTGACTGAA

3001 GATGGGCCTTTGAAATATAGGTATGCACCTGAGGTTGGGGGAGGGTCTGCACTCCCAAAC

3061 CCCAGCGCAGTGTCCTTTCCCTGCTGCCGACAGGAACCTGGGGCTGAACAGGTTATCCCT

3121 GTCAGGAGCCCTGGACTGGGCTGCATCTCAGCCCCACCTGCATGGTATCCAGCTCCCATC

3181 CACTTCTCACCCTTCTTTCCTCCTGACCTTGGTCAGCAGTGATGACCTCCAACTCTCACC

3241 CACCCCCTCTACCATCACCTCTAACCAGGCAAGCCAGGGTGGGAGAGCAATCAGGAGAGC

3301 CAGGCCTCAGCTTCCAATGCCTGGAGGGCCTCCACTTTGTGGCCAGCCTGTGGTGGTGGC

3361 TCTGAGGCCTAGGCAACGAGCGACAGGGCTGCCAGTTGCCCCTGGGTTCCTTTGTGCTGC

3421 TGTGTGCCTCCTCTCCTGCCGCCCTTTGTCCTCCGCTAAGAGACCCTGCCCTACCTGGCC

3481 GCTGGGCCCCGTGACTTTCCCTTCCTGCCCAGGAAAGTGAGGGTCGGCTGGCCCCACCTT

3541 CCCTGTCCTGATGCCGACAGCTTAGGGAAGGGCAGTGAACTTGCATATGGGGCTTAGCCT

3601 TCTAGTCACAGCCTCTATATTTGATGCTAGAAAACACATATTTTTAAATGGAAGAAAAAT

3661 AAAAAGGCATTCCCCCTTCATCCCCCTACCTTAAACATATAATATTTTAAAGGTCAAAAA

3721 AGCAATCCAACCCACTGCAGAAGCTCTTTTTGAGCACTTGGTGGCATCAGAGCAGGAGGA

3781 GCCCCAGAGCCACCTCTGGTGTCCCCCCAGGCTACCTGCTCAGGAACCCCTTCTGTTCTC

3841 TGAGAAGTCAAGAGAGGACATTGGCTCACGCACTGTGAGATTTTGTTTTTATACTTGGAA

3901 GTGGTGAATTATTTTATATAAAGTCATTTAAATATCTATTTAAAAGATAGGAAGCTGCTT

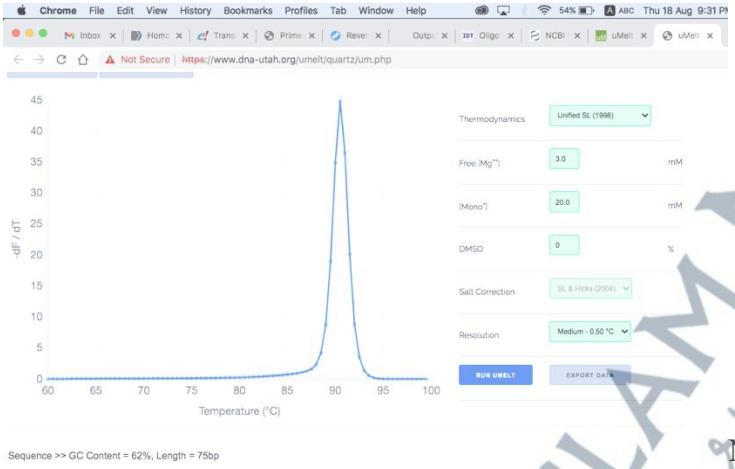
3961 ATATATTTAATAATAAAAGAAGTGCACAAGCTGC

KEYS (in order of precedence):
>>>>>> left primer

<<<<<< right primer

IDT OligoAnalyzer

	Forward	Reverse
Hairpin ΔG (kcal.mole-1)	0.61	0.43
Self-dimer ΔG (kcal.mole-1)	-5.38	-3.61
Hetero-dimer ΔG (kcal.mole-1)	-5.19	



NCBI BLAST- PASSED

TNF

PRIMER PICKING RESULTS FOR

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Using library: homo_sapiens with failure rate: 0.1
No mispriming library specified
Using 1-based sequence positions

OLIGO	start	len	tm	gc%	any_th	3'_th	hairpin	seq
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RIGHT PRIMER	470	21	59.05	47.62	0.00	0.00	0.00	GCTTGAGGGTTTGCTACAACA

PRODUCT SIZE: 70, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00

```
   1 AGCAGACGCTCCCTCAGCAAGGACAGCAGAGGACCAGCTAAGAGGGAGAGAAGCAACTAC

  61 AGACCCCCCCTGAAAACAACCCTCAGACGCCACATCCCCTGACAAGCTGCCAGGCAGGTT

 121 CTCTTCCTCTCACATACTGACCCACGGCTCCACCCTCTCTCCCCTGGAAAGGACACCATG

 181 AGCACTGAAAGCATGATCCGGGACGTGGAGCTGGCCGAGGAGGCGCTCCCCAAGAAGACA

 241 GGGGGGCCCCAGGGCTCCAGGCGGTGCTTGTTCCTCAGCCTCTTCTCCTTCCTGATCGTG

 301 GCAGGCGCCACCACGCTCTTCTGCCTGCTGCACTTTGGAGTGATCGGCCCCCAGAGGGAA
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 361 GAGTTCCCCAGGGACCTCTCTCTAATCAGCCCTCTGGCCCAGGCAGTCAGATCATCTTCT
                                                >>>>>>>>>>>>>>>>>>>>

 421 CGAACCCCGAGTGACAAGCCTGTAGCCCATGTTGTAGCAAACCCTCAAGCTGAGGGGCAG
     >>                          <<<<<<<<<<<<<<<<<<<

 481 CTCCAGTGGCTGAACCGCCGGGCCAATGCCCTCCTGGCCAATGGCGTGGAGCTGAGAGAT

 541 AACCAGCTGGTGGTGCCATCAGAGGGCCTGTACCTCATCTACTCCCAGGTCCTCTTCAAG

 601 GGCCAAGGCTGCCCCTCCACCCATGTGCTCCTCACCCACACCATCAGCCGCATCGCCGTC

 661 TCCTACCAGACCAAGGTCAACCTCCTCTCTGCCATCAAGAGCCCCTGCCAGAGGGAGACC

 721 CCAGAGGGGGCTGAGGCCAAGCCCTGGTATGAGCCCATCTATCTGGGAGGGGTCTTCCAG

 781 CTGGAGAAGGGTGACCGACTCAGCGCTGAGATCAATCGGCCCGACTATCTCGACTTTGCC

 841 GAGTCTGGGCAGGTCTACTTTGGGATCATTGCCCTGTGAGGAGGACGAACATCCAACCTT

 901 CCCAAACGCCTCCCCTGCCCCAATCCCTTTATTACCCCCTCCTTCAGACACCCTCAACCT

 961 CTTCTGGCTCAAAAAGAGAATTGGGGGCTTAGGGTCGGAACCCAAGCTTAGAACTTTAAG

1021 CAACAAGACCACCACTTCGAAACCTGGGATTCAGGAATGTGTGGCCTGCACAGTGAAGTG

1081 CTGGCAACCACTAAGAATTCAAACTGGGGCCTCCAGAACTCACTGGGGCCTACAGCTTTG

1141 ATCCCTGACATCTGGAATCTGGAGACCAGGGAGCCTTTGGTTCTGGCCAGAATGCTGCAG

1201 GACTTGAGAAGACCTCACCTAGAAATTGACACAAGTGGACCTTAGGCCTTCCTCTCTCCA

1261 GATGTTTCCAGACTTCCTTGAGACACGGAGCCCAGCCCTCCCCATGGAGCCAGCTCCCTC

1321 TATTTATGTTTGCACTTGTGATTATTTATTATTTATTTATTATTTATTTATTTACAGATG

1381 AATGTATTTATTTGGGAGACCGGGGTATCCTGGGGGACCCAATGTAGGAGCTGCCTTGGC

1441 TCAGACATGTTTTCCGTGAAAACGGAGCTGAACAATAGGCTGTTCCCATGTAGCCCCCTG

1501 GCCTCTGTGCCTTCTTTTGATTATGTTTTTTAAAATATTTATCTGATTAAGTTGTCTAAA
```

1561 CAATGCTGATTTGGTGACCAACTGTCACTCATTGCTGAGCCTCTGCTCCCCAGGGGAGTT

1621 GTGTCTGTAATCGCCCTACTATTCAGTGGCGAGAAATAAAGTTTGCTTAGAAAAGAAA

KEYS (in order of precedence):
>>>>>> left primer
<<<<<< right primer

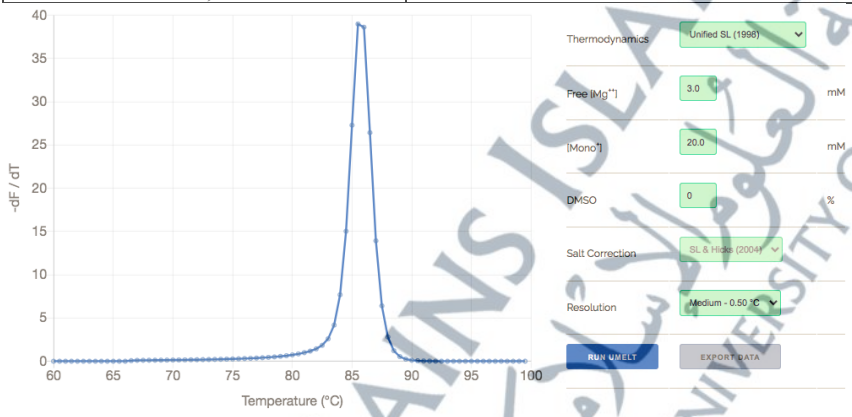
ADDITIONAL OLIGOS

	start	len	tm	gc%	any_th	3'_th	hairpin	seq
1 LEFT PRIMER	398	20	58.87	55.00	0.00	0.00	0.00	CCCAGGCAGTCAGATCATCT
RIGHT PRIMER	545	21	59.25	52.38	0.00	0.00	0.00	TGGTTATCTCTCAGCTCCACG

PRODUCT SIZE: 148, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00

IDT OligoAnalyzer

	Forward	Reverse
Hairpin ΔG (kcal.mole-1)	0.69	-0.19
Self-dimer ΔG (kcal.mole-1)	-4.65	-3.9
Hetero-dimer ΔG (kcal.mole-1)	-5.09	



Sequence >> GC Content = 52%, Length = 70bp

AGGCAGTCAGATCATCTTCTCGAACCCCGAGTGACAAGCCTGTAGCCCATGTTGTAGCAAACCCTCAAGC

NCBI BLAST-PASSED

MAP4K1

PRIMER PICKING RESULTS FOR

Template masking selected
Using library: homo_sapiens with failure rate: 0.1
No mispriming library specified
Using 1-based sequence positions

OLIGO	start	len	tm	gc%	any_th	3'_th	hairpin	seq
LEFT PRIMER	365	20	60.18	55.00	0.00	0.00	0.00	TACCAAGTGACAGGCTCCCT
RIGHT PRIMER	512	20	59.74	55.00	0.00	0.00	0.00	TGACCTCCCCAGCATCATTG

SEQUENCE SIZE: 2729
INCLUDED REGION SIZE: 2729

PRODUCT SIZE: 148, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00

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   1 AGTTGAGCCCTGAGGCCCTGCTGGCCCCTGGGCGCAGGCCCAGCTCAGGCCCCCAGGGAT

  61 GGACGTCGTGGACCCTGACATTTTCAATAGAGACCCCCGGGACCACTATGACCTGCTACA

 121 GCGGCTGGGTGGCGGCACGTATGGGGAAGTCTTTAAGGCTCGAGACAAGGTGTCAGGGGA

 181 CCTGGTGGCACTGAAGATGGTGAAGATGGAGCCTGATGATGATGTCTCCACCCTTCAGAA

 241 GGAAATCCTCATATTGAAAACTTGCCGGCACGCCAACATCGTGGCCTACCATGGGAGTTA

 301 TCTCTGGTTGCAGAAACTCTGGATCTGCATGGAATTCTGTGGGGCTGGTTCTCTCCAGGA

 361 CATCTACCAAGTGACAGGCTCCCTGTCAGAGCTCCAGATTAGCTATGTCTGCCGGGAAGT
         >>>>>>>>>>>>>>>>>>>>

 421 GCTCCAGGGACTGGCCTATTTGCACTCACAGAAGAAGATACACAGGGACATCAAGGGAGC

 481 TAACATCCTCATCAATGATGCTGGGGAGGTCAGATTGGCTGACTTTGGCATCTCGGCCCA
                 <<<<<<<<<<<<<<<<<<<<

 541 GATTGGGGCTACACTGGCCAGACGCCTCTCTTTCATTGGGACACCCTACTGGATGGCTCC

 601 GGAAGTGGCAGCTGTGGCCCTGAAGGGAGGATACAATGAGCTGTGTGACATCTGGTCCCT

 661 GGGCATCACGGCCATCGAACTGGCCGAGCTACAGCCACCGCTCTTTGATGTGCACCCTCT

 721 CAGAGTTCTCTTCCTCATGACCAAGAGTGGCTACCAGCCTCCCCGACTGAAGGAAAAAGG

 781 CAAATGGTCGGCTGCCTTCCACAACTTCATCAAAGTCACTCTGACTAAGAGTCCCAAGAA

 841 ACGACCCAGCGCCACCAAGATGCTCAGTCATCAACTGGTATCCCAGCCTGGGCTGAATCG

 901 AGGCCTGATCCTGGATCTTCTTGACAAACTGAAGAATCCCGGGAAAGGACCCTCCATTGG

 961 GGACATTGAGGATGAGGAGCCCGAGCTACCCCCTGCTATCCCTCGGCGGATCAGATCCAC

1021 CCACCGCTCCAGCTCTCTGGGGATCCCAGATGCAGACTGCTGTCGGCGGCACATGGAGTT

1081 CAGGAAGCTCCGAGGAATGGAGACCAGACCCCCAGCCAACACCGCTCGCCTACAGCCTCC

1141 TCGAGACCTCAGGAGCAGCAGCCCCAGGAAGCAACTGTCAGAGTCGTCTGACGATGACTA
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1201 TGACGACGTGGACATCCCCACCCCTGCAGAGGACACACCTCCTCCACTTCCCCCCAAGCC

1261 CAAGTTCCGTTCTCCATCAGACGAGGGTCCTGGGAGCATGGGGGATGATGGGCAGCTGAG

1321 CCCGGGGGTGCTGGTCCGGTGTGCCAGTGGGCCCCCACCAAACAGCCCCCGTCCTGGGCC

1381 TCCCCCATCCACCAGCAGCCCCCACCTCACCGCCCATTCAGAACCCTCACTCTGGAACCC

1441 ACCCTCCCGGGAGCTTGACAAGCCCCCACTTCTGCCCCCCAAGAAGGAAAAGATGAAGAG

1501 AAAGGGATGTGCCCTTCTCGTAAAGTTGTTCAATGGCTGCCCCCTCCGGATCCACAGCAC

1561 GGCCGCCTGGACACATCCCTCCACCAAGGACCAGCACCTGCTCCTGGGGGCAGAGGAAGG

1621 CATCTTCATCCTGAACCGGAATGACCAGGAGGCCACGCTGGAAATGCTCTTTCCTAGCCG

1681 GACTACGTGGGTGTACTCCATCAACAACGTTCTCATGTCTCTCTCAGGAAAGACCCCCCA

1741 CCTGTATTCTCATAGCATCCTTGGCCTGCTGGAACGGAAAGAGACCAGAGCAGGAAACCC

1801 CATCGCTCACATTAGCCCCCACCGCCTACTGGCAAGGAAGAACATGGTTTCCACCAAGAT

1861 CCAGGACACCAAAGGCTGCCGGGCGTGCTGTGTGGCGGAGGGTGCGAGCTCTGGGGGCCC

1921 GTTCCTGTGCGGTGCATTGGAGACGTCCGTTGTCCTGCTTCAGTGGTACCAGCCCATGAA

1981 CAAATTCCTGCTTGTCCGGCAGGTGCTGTTCCCACTGCCGACGCCTCTGTCCGTGTTCGC

2041 GCTGCTGACCGGGCCAGGCTCTGAGCTGCCCGCTGTGTGCATCGGCGTGAGCCCCGGGCG

2101 GCCGGGGAAGTCGGTGCTCTTCCACACGGTGCGCTTTGGCGCGCTCTCTTGCTGGCTGGG

2161 CGAGATGAGCACCGAGCACAGGGGACCCGTGCAGGTGACCCAGGTAGAGGAAGATATGGT

2221 GATGGTGTTGATGGATGGCTCTGTGAAGCTGGTGACCCCGGAGGGGTCCCCAGTCCGGGG

2281 ACTTCGCACACCTGAGATCCCCATGACCGAAGCGGTGGAGGCCGTGGCTATGGTTGGAGG

2341 TCAGCTTCAGGCCTTCTGGAAGCATGGAGTGCAGGTGTGGGCTCTAGGCTCGGATCAGCT

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2401 GCTACAGGAGCTGAGAGACCCTACCCTCACTTTCCGTCTGCTTGGCTCCCCCAGGCTGGA

2461 GTGCAGTGGCACGATCTCGCCTCACTGCAACCTCCTCCTCCCAGGTTCAAGCAATTCTCC

2521 TGCCTCAGCCTCCCGAGTAGCTGGGATTACAGGCCTGTAGTGGTGGAGACACGCCCAGTG

2581 GATGATCCTACTGCTCCCAGCAACCTCTACATCCAGGAATGAGTCCCTAGGGGGGTGTCA

2641 GGAACTAGTCCTTGCACCCCCTCCCCCATAGACACACTAGTGGTCATGGCATGTCCTCAT

2701 CTCCCAATAAACATGACTTTAGCCTCTGC


KEYS (in order of precedence):
>>>>>> left primer
<<<<<< right primer

ADDITIONAL OLIGOS
                    start  len      tm      gc%   any_th  3'_th  hairpin  seq

 1 LEFT PRIMER        360   22    59.77    50.00    0.00    0.00     0.00
ACATCTACCAAGTGACAGGCTC
   RIGHT PRIMER       480   20    60.39    60.00    0.00    0.00     0.00
GCTCCCTTGATGTCCCTGTG
   PRODUCT SIZE: 121, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00

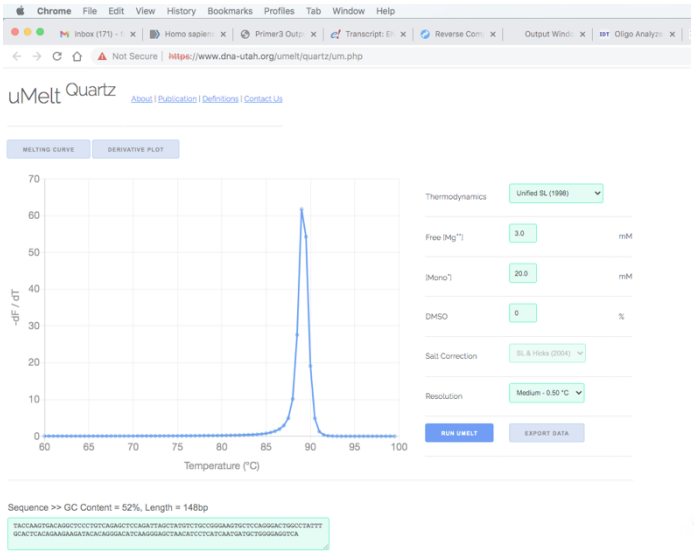
 2 LEFT PRIMER        249   22    59.18    40.91   29.03   29.03     0.00
TCATATTGAAAACTTGCCGGCA
   RIGHT PRIMER       383   20    59.75    60.00    0.00    0.00     0.00
GGGAGCCTGTCACTTGGTAG
   PRODUCT SIZE: 135, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00

 3 LEFT PRIMER        355   22    59.43    50.00    0.00    0.00     0.00
CCAGGACATCTACCAAGTGACA
   RIGHT PRIMER       450   21    59.93    47.62    5.01    0.00     0.00
TGTGAGTGCAAATAGGCCAGT
   PRODUCT SIZE: 96, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00

 4 LEFT PRIMER        312   22    59.16    45.45    0.00    0.00     0.00
AGAAACTCTGGATCTGCATGGA
   RIGHT PRIMER       381   22    59.77    50.00    0.00    0.00     0.00
GAGCCTGTCACTTGGTAGATGT
   PRODUCT SIZE: 70, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00
```

IDT OligoAnalyzer

	Forward	Reverse
Hairpin **ΔG (kcal.mole-1)**	0.25	0.77
Self-dimer **ΔG (kcal.mole-1)**	-4.67	-3.53
Hetero-dimer **ΔG (kcal.mole-1)**	-4.74	



NCBI BLAST-PASSED

IL1B

PRIMER PICKING RESULTS FOR

Template masking selected
Using library: homo_sapiens with failure rate: 0.1
No mispriming library specified
Using 1-based sequence positions

OLIGO	start	len	tm	gc%	any_th	3'_th	hairpin	seq
LEFT PRIMER	639	20	59.41	55.00	0.00	0.00	0.00	CCTGTCCTGCGTGTTGAAAG
RIGHT PRIMER	788	20	59.89	55.00	0.00	0.00	0.00	GGGAACTGGGCAGACTCAAA

SEQUENCE SIZE: 1507
INCLUDED REGION SIZE: 1507

PRODUCT SIZE: 150, PAIR ANY_TH COMPL: 0.73, PAIR 3'_TH COMPL: 0.00
TARGETS (start len

```
    1 ACCAAACCTCTTCGAGGCACAAGGCACAACAGGCTGCTCTGGGATTCTCTTCAGCCAATC

   61 TTCATTGCTCAAGTGTCTGAAGCAGCCATGGCAGAAGTACCTGAGCTCGCCAGTGAAATG

  121 ATGGCTTATTACAGTGGCAATGAGGATGACTTGTTCTTTGAAGCTGATGGCCCTAAACAG

  181 ATGAAGTGCTCCTTCCAGGACCTGGACCTCTGCCCTCTGGATGGCGGCATCCAGCTACGA

  241 ATCTCCGACCACCACTACAGCAAGGGCTTCAGGCAGGCCGCGTCAGTTGTTGTGGCCATG

  301 GACAAGCTGAGGAAGATGCTGGTTCCCTGCCCACAGACCTTCCAGGAGAATGACCTGAGC

  361 ACCTTCTTTCCCTTCATCTTTGAAGAAGAACCTATCTTCTTCGACACATGGGATAACGAG
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 421 GCTTATGTGCACGATGCACCTGTACGATCACTGAACTGCACGCTCCGGGACTCACAGCAA

 481 AAAAGCTTGGTGATGTCTGGTCCATATGAACTGAAAGCTCTCCACCTCCAGGGACAGGAT

 541 ATGGAGCAACAAGTGGTGTTCTCCATGTCCTTTGTACAAGGAGAAGAAAGTAATGACAAA

 601 ATACCTGTGGCCTTGGGCCTCAAGGAAAAGAATCTGTACCTGTCCTGCGTGTTGAAAGAT
                                                >>>>>>>>>>>>>>>>>>>

 661 GATAAGCCCACTCTACAGCTGGAGAGTGTAGATCCCAAAAATTACCCAAAGAAGAAGATG
                      **********

 721 GAAAAGCGATTTGTCTTCAACAAGATAGAAATCAATAACAAGCTGGAATTTGAGTCTGCC
                                                  <<<<<<<<<<<

 781 CAGTTCCCCAACTGGTACATCAGCACCTCTCAAGCAGAAAACATGCCCGTCTTCCTGGGA
     <<<<<<<<

 841 GGGACCAAAGGCGGCCAGGATATAACTGACTTCACCATGCAATTTGTGTCTTCCTAAAGA

 901 GAGCTGTACCCAGAGAGTCCTGTGCTGAATGTGGACTCAATCCCTAGGGCTGGCAGAAAG

 961 GGAACAGAAAGGTTTTTGAGTACGGCTATAGCCTGGACTTTCCTGTTGTCTACACCAATG

1021 CCCAACTGCCTGCCTTAGGGTAGTGCTAAGAGGATCTCCTGTCCATCAGCCAGGACAGTC

1081 AGCTCTCTCCTTTCAGGGCCAATCCCCAGCCCTTTTGTTGAGCCAGGCCTCTCTCACCTC

1141 TCCTACTCACTTAAAGCCCGCCTGACAGAAACCACGGCCACATTTGGTTCTAAGAAACCC

1201 TCTGTCATTCGCTCCCACATTCTGATGAGCAACCGCTTCCCTATTTATTTATTTATTTGT

1261 TTGTTTGTTTTATTCATTGGTCTAATTTATTCAAAGGGGGCAAGAAGTAGCAGTGTCTGT

1321 AAAAGAGCCTAGTTTTTAATAGCTATGGAATCAATTCAATTTGGACTGGTGTGCTCTCTT

1381 TAAATCAAGTCCTTTAATTAAGACTGAAAATATATAAGCTCAGATTATTTAAATGGGAAT

1441 ATTTATAAATGAGCAAATATCATACTGTTCAATGGTTCTGAAATAAACTTCACTGAAGAA

1501 AAAAAAA
```

KEYS (in order of precedence):

```
****** target
>>>>>> left primer
<<<<<< right primer

ADDITIONAL OLIGOS
                    start  len      tm      gc%   any_th   3'_th  hairpin  seq

 1 LEFT PRIMER        636   20   59.97    55.00     0.00    0.00     0.00
GTACCTGTCCTGCGTGTTGA
   RIGHT PRIMER       781   20   59.19    55.00     0.00    0.00     0.00
GGGCAGACTCAAATTCCAGC
   PRODUCT SIZE: 146, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00

 2 LEFT PRIMER        632   20   59.47    55.00     0.00    0.00     0.00
ATCTGTACCTGTCCTGCGTG
   RIGHT PRIMER       780   22   60.09    50.00     0.00    0.00     0.00
GGCAGACTCAAATTCCAGCTTG
   PRODUCT SIZE: 149, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00

 3 LEFT PRIMER        642   21   58.86    47.62     0.00    0.00     0.00
GTCCTGCGTGTTGAAAGATGA
   RIGHT PRIMER       791   19   60.77    57.89     0.00    0.00     0.00
TTGGGGAACTGGGCAGACT
   PRODUCT SIZE: 150, PAIR ANY_TH COMPL: 3.52, PAIR 3'_TH COMPL: 0.00

 4 LEFT PRIMER        647   22   58.31    45.45     0.00    0.00     0.00
GCGTGTTGAAAGATGATAAGCC
   RIGHT PRIMER       786   22   59.51    50.00     0.00    0.00     0.00
GAACTGGGCAGACTCAAATTCC
   PRODUCT SIZE: 140, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00
```

IDT OligoAnalyzer

	Forward	Reverse
Hairpin **ΔG (kcal.mole-1)**	0.57	-0.84
Self-dimer **ΔG (kcal.mole-1)**	-3.61	-3.55
Hetero-dimer **ΔG (kcal.mole-1)**	-3.89	

NCBI BLAST-PASSED

MYC

PRIMER PICKING RESULTS FOR

Template masking selected
Using library: homo_sapiens with failure rate: 0.1
No mispriming library specified
Using 1-based sequence positions

OLIGO	start	len	tm	gc%	any_th	3'_th	hairpin	seq
LEFT PRIMER	1177	20	59.30	55.00	0.00	0.00	0.00	TAGTGGAAAACCAGCCTCCC
RIGHT PRIMER	1288	20	60.04	55.00	0.00	0.00	0.00	TCCTCCTCGTCGCAGTAGAA

SEQUENCE SIZE: 4515
INCLUDED REGION SIZE: 4515

PRODUCT SIZE: 112, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00
TARGETS (start len

```
    1 GGAGTTTATTCATAACGCGCTCTCCAAGTATACGTGGCAATGCGTTGCTGGGTTATTTTA

   61 ATCATTCTAGGCATCGTTTTCCTCCTTATGCCTCTATCATTCCTCCCTATCTACACTAAC

  121 ATCCCACGCTCTGAACGCGCGCCCATTAATACCCTTCTTTCCTCCACTCTCCCTGGGACT

  181 CTTGATCAAAGCGCGGCCCTTTCCCCAGCCTTAGCGAGGCGCCCTGCAGCCTGGTACGCG

  241 CGTGGCGTGGCGGTGGGCGCGCAGTGCGTTCTCGGTGTGGAGGGCAGCTGTTCCGCCTGC

  301 GATGATTTATACTCACAGGACAAGGATGCGGTTTGTCAAACAGTACTGCTACGGAGGAGC

  361 AGCAGAGAAAGGGAGAGGGTTTGAGAGGGAGCAAAACAAAATGGTAGGCGCGCGTAGTTA

  421 ATTCATGCGGCTCTCTTACTCTGTTTACATCCTAGAGCTAGAGTGCTCGGCTGCCCGGCT

  481 GAGTCTCCTCCCCACCTTCCCCACCCTCCCCACCCTCCCCATAAGCGCCCCTCCCGGGTT

  541 CCCAAAGCAGAGGGCGTGGGGGAAAAGAAAAAAGATCCTCTCTCGCTAATCTCCGCCCAC

  601 CGGCCCTTTATAATGCGAGGGTCTGGACGGCTGAGGACCCCCGAGCTGTGCTGCTCGCGG

  661 CCGCCACCGCCGGGCCCCGGCCGTCCCTGGCTCCCCTCCTGCCTCGAGAAGGGCAGGGCT

  721 TCTCAGAGGCTTGGCGGGAAAAAGAACGGAGGGAGGGATCGCGCTGAGTATAAAAGCCGG

  781 TTTTCGGGGCTTTATCTAACTCGCTGTAGTAATTCCAGCGAGAGGCAGAGGGAGCGAGCG
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 841 GGCGGCCGGCTAGGGTGGAAGAGCCGGGCGAGCAGAGCTGCGCTGCGGGCGTCCTGGGAA

 901 GGGAGATCCGGAGCGAATAGGGGGCTTCGCCTCTGGCCCAGCCCTCCCGCTGATCCCCCA

 961 GCCAGCGGTCCGCAACCCTTGCCGCATCCACGAAACTTTGCCCATAGCAGCGGGCGGGCA

1021 CTTTGCACTGGAACTTACAACACCCGAGCAAGGACGCGACTCTCCCGACGCGGGGAGGCT

1081 ATTCTGCCCATTTGGGGACACTTCCCCGCCGCTGCCAGGACCCGCTTCTCTGAAAGGCTC

1141 TCCTTGCAGCTGCTTAGACGCTGGATTTTTTTCGGGTAGTGGAAAACCAGCCTCCCGCGA
                                          >>>>>>>>>>>>>>>>>>>>

1201 CGATGCCCCTCAACGTTAGCTTCACCAACAGGAACTATGACCTCGACTACGACTCGGTGC
       **********

1261 AGCCGTATTTCTACTGCGACGAGGAGGAGAACTTCTACCAGCAGCAGCAGCAGAGCGAGC
               <<<<<<<<<<<<<<<<<<<

1321 TGCAGCCCCCGGCGCCCAGCGAGGATATCTGGAAGAAATTCGAGCTGCTGCCCACCCCGC

1381 CCCTGTCCCCTAGCCGCCGCTCCGGGCTCTGCTCGCCCTCCTACGTTGCGGTCACACCCT

1441 TCTCCCTTCGGGGAGACAACGACGGCGGTGGCGGGAGCTTCTCCACGGCCGACCAGCTGG

1501 AGATGGTGACCGAGCTGCTGGGAGGAGACATGGTGAACCAGAGTTTCATCTGCGACCCGG

1561 ACGACGAGACCTTCATCAAAAACATCATCATCCAGGACTGTATGTGGAGCGGCTTCTCGG

1621 CCGCCGCCAAGCTCGTCTCAGAGAAGCTGGCCTCCTACCAGGCTGCGCGCAAAGACAGCG

1681 GCAGCCCGAACCCCGCCCGCGGCCACAGCGTCTGCTCCACCTCCAGCTTGTACCTGCAGG

1741 ATCTGAGCGCCGCCGCCTCAGAGTGCATCGACCCCTCGGTGGTCTTCCCCTACCCTCTCA

1801 ACGACAGCAGCTCGCCCAAGTCCTGCGCCTCGCAAGACTCCAGCGCCTTCTCTCCGTCCT

1861 CGGATTCTCTGCTCTCCTCGACGGAGTCCTCCCCGCAGGGCAGCCCCGAGCCCCTGGTGC

1921 TCCATGAGGAGACACCGCCCACCACCAGCAGCGACTCTGAGGAGGAACAAGAAGATGAGG

1981 AAGAAATCGATGTTGTTTCTGTGGAAAAGAGGCAGGCTCCTGGCAAAAGGTCAGAGTCTG
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2041 GATCACCTTCTGCTGGAGGCCACAGCAAACCTCCTCACAGCCCACTGGTCCTCAAGAGGT

2101 GCCACGTCTCCACACATCAGCACAACTACGCAGCGCCTCCCTCCACTCGGAAGGACTATC

2161 CTGCTGCCAAGAGGGTCAAGTTGGACAGTGTCAGAGTCCTGAGACAGATCAGCAACAACC

2221 GAAAATGCACCAGCCCCAGGTCCTCGGACACCGAGGAGAATGTCAAGAGGCGAACACACA

2281 ACGTCTTGGAGCGCCAGAGGAGGAACGAGCTAAAACGGAGCTTTTTTGCCCTGCGTGACC

2341 AGATCCCGGAGTTGGAAAACAATGAAAAGGCCCCCAAGGTAGTTATCCTTAAAAAAGCCA

2401 CAGCATACATCCTGTCCGTCCAAGCAGAGGAGCAAAAGCTCATTTCTGAAGAGGACTTGT

2461 TGCGGAAACGACGAGAACAGTTGAAACACAAACTTGAACAGCTACGGAACTCTTGTGCGT

2521 AAGGAAAAGTAAGGAAAACGATTCCTTCTAACAGAAATGTCCTGAGCAATCACCTATGAA

2581 CTTGTTTCAAATGCATGATCAAATGCAACCTCACAACCTTGGCTGAGTCTTGAGACTGAA

2641 AGATTTAGCCATAATGTAAACTGCCTCAAATTGGACTTTGGGCATAAAAGAACTTTTTTA

2701 TGCTTACCATCTTTTTTTTTCTTTAACAGATTTGTATTTAAGAATTGTTTTTAAAAAAT

2761 TTTAAGATTTACACAATGTTTCTCTGTAAATATTGCCATTAAATGTAAATAACTTTAATA

2821 AAACGTTTATAGCAGTTACACAGAATTTCAATCCTAGTATATAGTACCTAGTATTATAGG

2881 TACTATAAACCCTAATTTTTTTTATTTAAGTACATTTTGCTTTTTAAAGTTGATTTTTTT

2941 CTATTGTTTTTAGAAAAAATAAAATAACTGGCAAATATATCATTGAGCCAAATCTTAAGT

3001 TGTGAATGTTTTGTTTCGTTTCTTCCCCCTCCCAACCACCACCATCCCTGTTTGTTTTCA

3061 TCAATTGCCCCTTCAGAGGGTGGTCTTAAGAAAGGCAAGAGTTTTCCTCTGTTGAAATGG

3121 GTCTGGGGGCCTTAAGGTCTTTAAGTTCTTGGAGGTTCTAAGATGCTTCCTGGAGACTAT

3181 GATAACAGCCAGAGTTGACAGTTAGAAGGAATGGCAGAAGGCAGGTGAGAAGGTGAGAGG

3241 TAGGCAAAGGAGATACAAGAGGTCAAAGGTAGCAGTTAAGTACACAAAGAGGCATAAGGA

3301 CTGGGGAGTTGGGAGGAAGGTGAGGAAGAAACTCCTGTTACTTTAGTTAACCAGTGCCAG

3361 TCCCCTGCTCACTCCAAACCCAGGAATTCTGCCCAGTTGATGGGGACACGGTGGGAACCA

3421 GCTTCTGCTGCCTTCACAACCAGGCGCCAGTCCTGTCCATGGGTTATCTCGCAAACCCCA

3481 GAGGATCTCTGGGAGGAATGCTACTATTAACCCTATTTCACAAACAAGGAAATAGAAGAG

3541 CTCAAAGAGGTTATGTAACTTATCTGTAGCCACGCAGATAATACAAAGCAGCAATCTGGA

3601 CCCATTCTGTTCAAAACACTTAACCCTTCGCTATCATGCCTTGGTTCATCTGGGTCTAAT

3661 GTGCTGAGATCAAGAAGGTTTAGGACCTAATGGACAGACTCAAGTCATAACAATGCTAAG

3721 CTCTATTTGTGTCCCAAGCACTCCTAAGCATTTTATCCCTAACTCTACATCAACCCCATG

3781 AAGGAGATACTGTTGATTTCCCCATATTAGAAGTAGAGAGGGAAGCTGAGGCACACAAAG

3841 ACTCATCCACATGCCCAAGATTCACTGATAGGGAAAAGTGGAAGCGAGATTTGAACCCAG

3901 GCTGTTTACTCCTAACCTGTCCAAGCCACCTCTCAGACGACGGTAGGAATCAGCTGGCTG

3961 CTTGTGAGTACAGGAGTTACAGTCCAGTGGGTTATGTTTTTTAAGTCTCAACATCTAAGC

4021 CTGGTCAGGCATCAGTTCCCCTTTTTTTGTGATTTATTTTGTTTTTATTTTGTTGTTCAT

4081 TGTTTAATTTTTCCTTTTACAATGAGAAGGTCACCATCTTGACTCCTACCTTAGCCATTT

4141 GTTGAATCAGACTCATGACGGCTCCTGGGAAGAAGCCAGTTCAGATCATAAAATAAAACA

4201 TATTTATTCTTTGTCATGGGAGTCATTATTTTAGAAACTACAAACTCTCCTTGCTTCCAT

4261 CCTTTTTTACATACTCATGACACATGCTCATCCTGAGTCCTTGAAAAGGTATTTTTGAAC

4321 ATGTGTATTAATTATAAGCCTCTGAAAACCTATGGCCCAAACCAGAAATGATGTTGATTA

4381 TATAGGTAAATGAAGGATGCTATTGCTGTTCTAATTACCTCATTGTCTCAGTCTCAAAGT

```
4441 AGGTCTTCAGCTCCCTGTACTTTGGGATTTTAATCTACCACCACCCATAAATCAATAAAT


4501 AATTACTTTCTTTGA


KEYS (in order of precedence):
****** target
>>>>>> left primer
<<<<<< right primer

ADDITIONAL OLIGOS
                  start  len      tm      gc%  any_th  3'_th hairpin seq

 1 LEFT PRIMER       1149   20   58.63   50.00    0.00   0.00    0.00 GCTGCTTAGACGCTGGATTT
   RIGHT PRIMER      1291   20   60.04   55.00    0.00   0.00    0.00 TTCTCCTCCTCGTCGCAGTA
   PRODUCT SIZE: 143, PAIR ANY_TH COMPL: 18.08, PAIR 3'_TH COMPL: 3.80

 2 LEFT PRIMER       1122   20   59.75   55.00    0.00   0.00    0.00 CCGCTTCTCTGAAAGGCTCT
   RIGHT PRIMER      1263   19   60.51   63.16   12.31   0.00    0.00 GCTGCACCGAGTCGTAGTC
   PRODUCT SIZE: 142, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00
```

IDT OligoAnalyzer

	Forward	Reverse
Hairpin ΔG (kcal.mole-1)	-0.02	1.04
Self-dimer ΔG (kcal.mole-1)	-5.02	-3.61
Hetero-dimer ΔG (kcal.mole-1)	-4.64	

NCBI BLAST- PASSED

PAK1

PRIMER PICKING RESULTS FOR

Template masking selected
Using library: homo_sapiens with failure rate: 0.1
No mispriming library specified
Using 1-based sequence positions

OLIGO	start	len	tm	gc%	any_th	3'_th	hairpin	seq
LEFT PRIMER	1248	20	59.96	55.00	0.00	0.00	0.00	CACCACTCCACCAGATGCTT
RIGHT PRIMER	1354	22	58.15	45.45	0.00	0.00	0.00	CCACACTCACTATGCTTCGTAA

SEQUENCE SIZE: 3508
INCLUDED REGION SIZE: 3508

PRODUCT SIZE: 107, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00

```
    1 GTACAATAGCGCGGCTGTGGGCGGGGGAGGCTGCCCTCCCGCGGCTGCAGCCGGAGCCGA

   61 AGGTGGTGGCTGCACAGTAGACGCCCCCTCACGGCTTCCCCCACACGCTCCCGCCCCCTC

  121 GCTCGCCCATCGCGCTTCCCTCACAGGCTCTGCAGTCCTCCCCCACAGACGCCTTCCCCC

  181 TTGGACTCTCATTCCCTTTTCCACGGAGCCCCGCGCTTTCGTGAGCCCCCTCGAGGAACC

  241 TGGTCTCCGCATCCAGTTACCACCTCCTGCCTCAGAGGCCATCTGAGCCCTTCGCACCTC

  301 GCCCCTCAGTCCCCCCTTCCCCCCCCGCCCGCGTCGCCTCGCTCCCTCCCGCCCCCCCAT

  361 CATCCCTTCCCTCGCAGTTCCCCTGTCCTGAGGGGAGCCCCGCCACGGGCAGCGCGGCGG

  421 CGGCGGCAGGAGGGAGAAAGTGAAGCGGTAGCTCGCGCACACTCGCGCCCTCACTCCCGG

  481 CTAGGCGGCACCCACCGCCGGGAGGAGGAGGAGGAGCCGAGAGGAGCTGAGCGAGCGCGG

  541 AAGTAGCTGCTGCTGGTGGTGACAATGTCAAATAACGGCCTAGACATTCAAGACAAACCC

  601 CCAGCCCCTCCGATGAGAAATACCAGCACTATGATTGGAGCCGGCAGCAAAGATGCTGGA

  661 ACCCTAAACCATGGTTCTAAACCTCTGCCTCCAAACCCAGAGGAGAAGAAAAAGAAGGAC

  721 CGATTTTACCGATCCATTTTACCTGGAGATAAAACAAATAAAAAGAAAGAGAAAGAGCGG

  781 CCAGAGATTTCTCTCCCTTCAGATTTTGAACACACAATTCATGTCGGTTTTGATGCTGTC
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 841 ACAGGGGAGTTTACGGGAATGCCAGAGCAGTGGGCCCGCTTGCTTCAGACATCAAATATC

 901 ACTAAGTCGGAGCAGAAGAAAAACCCGCAGGCTGTTCTGGATGTGTTGGAGTTTTACAAC

 961 TCGAAGAAGACATCCAACAGCCAGAAATACATGAGCTTTACAGATAAGTCAGCTGAGGAT

1021 TACAATTCTTCTAATGCCTTGAATGTGAAGGCTGTGTCTGAGACTCCTGCAGTGCCACCA

1081 GTTTCAGAAGATGAGGATGATGATGATGATGATGCTACCCCACCACCAGTGATTGCTCCA

1141 CGCCCAGAGCACACAAAATCTGTATACACACGGTCTGTGATTGAACCACTTCCTGTCACT

1201 CCAACTCGGGACGTGGCTACATCTCCCATTTCACCTACTGAAAATAACACCACTCCACCA
                                                    >>>>>>>>>>>>>

1261 GATGCTTTGACCCGGAATACTGAGAAGCAGAAGAAGAAGCCTAAAATGTCTGATGAGGAG
     >>>>>>>

1321 ATCTTGGAGAAATTACGAAGCATAGTGAGTGTGGGCGATCCTAAGAAGAAATATACACGG
                  <<<<<<<<<<<<<<<<<<<<

1381 TTTGAGAAGATTGGACAAGGTGCTTCAGGCACCGTGTACACAGCAATGGATGTGGCCACA

1441 GGACAGGAGGTGGCCATTAAGCAGATGAATCTTCAGCAGCAGCCCAAGAAAGAGCTGATT

1501 ATTAATGAGATCCTGGTCATGAGGGAAAACAAGAACCCAAACATTGTGAATTACTTGGAC

1561 AGTTACCTCGTGGGAGATGAGCTGTGGGTTGTTATGGAATACTTGGCTGGAGGCTCCTTG

1621 ACAGATGTGGTGACAGAAACTTGCATGGATGAAGGCCAAATTGCAGCTGTGTGCCGTGAG

1681 TGTCTGCAGGCTCTGGAGTTCTTGCATTCGAACCAGGTCATTCACAGAGACATCAAGAGT

1741 GACAATATTCTGTTGGGAATGGATGGCTCTGTCAAGCTAACTGACTTTGGATTCTGTGCA

1801 CAGATAACCCCAGAGCAGAGCAAACGGAGCACCATGGTAGGAACCCCATACTGGATGGCA

1861 CCAGAGGTTGTGACACGAAAGGCCTATGGGCCCAAGGTTGACATCTGGTCCCTGGGCATC

1921 ATGGCCATCGAAATGATTGAAGGGGAGCCTCCATACCTCAATGAAAACCCTCTGAGAGCC

1981 TTGTACCTCATTGCCACCAATGGGACCCCAGAACTTCAGAACCCAGAGAAGCTGTCAGCT
```

2041 ATCTTCCGGGACTTTCTGAACCGCTGTCTCGAGATGGATGTGGAGAAGAGAGGTTCAGCT

2101 AAAGAGCTGCTACAGGTGAGAAAACTGAGGTTTCAAGTGTTTAGTAACTTTTCCATGATA

2161 GCTGCATCAATTCCTGAAGATTGCCAAGCCCCTCTCCAGCCTCACTCCACTGATTGCTGC

2221 AGCTAAGGAGGCAACAAAGAACAATCACTAAAACCACACTCACCCCAGCCTCATTGTGCC

2281 AAGCCTTCTGTGAGATAAATGCACATTTCAGAAATTCCAACTCCTGATGCCCTCTTCTCC

2341 TTGCCTTGCTTCTCCCATTTCCTGATCTAGCACTCCTCAAGACTTTGATCCTTGGAAACC

2401 GTGTGTCCAGCATTGAAGAGAACTGCAACTGAATGACTAATCAGATGATGGCCATTTCTA

2461 AATAAGGAATTTCCTCCCAATTCATGGATATGAGGGTGGTTTATGATTAAGGGTTTATAT

2521 AAATAAATGTTTCTAGTCTTCCGTGTGTCAAAATCCTCACCTCCTTCATAACCATCTCCC

2581 ACAATTAATTCTTGACTATATAAATTTATGGTTTGATAATATTATCAATTTGTAATCAAT

2641 TGAGATTTCTTTAGTGCTTGCTTTTCTGTGACTCAACTGCCCAGACACCTCATTGTACTT

2701 GAAAACTGGAACAGCTTGGGAATGCCATGGGGTTTGATAATCTGCCAGGGACATGAAGAG

2761 GCTCAGCTTCCTGGACCATGACTTTGGCTCAGCTGATCCTGACATGGGAGAACAACCACA

2821 TTTTTCTTTGTGTGTGCTTCTAGCAGCTGTTCGGGAGGACCTTGACCCAATAGTGTTCCC

2881 ATGCTGTTTCTTGTGAAATGCTCTCGGCTATGTAGCAGCTTTTGATTCCCTGCATACCCT

2941 AGGCTGCTGCCCCTATCCTGTCCCTTGTTTATAACATTGAGAGGTTTTCTAGGGCACATA

3001 CTGAGTGAGAGCAGTGTTGAGAAGTCGGGGAAAATGGTGACTACTTTTAGAGCAAGGCTG

3061 GGCATCAGCACCTGTCCAGCTCTACTTGTGTGATGTTTCAGGAACTCAGCCCCTTTTTCT

3121 GCCTAGGATAAGGAGCTGAAAGATTAACTTGGATCTTCTAATGGTCCAAATCTTTTGGTC

3181 ACAATAAAGAGTCTCCAAATTAGAGACTGCATGTTAGTTCTGGATGGATTTGGTGGCCTG

```
3241 ACATGATACCCTGCCAGCTGTGAGGGGACCCCGTTTTTAAGATGCATGGCCAAGCTCTCT

3301 GCAAATGGAAATGCTTACACTGGGTGTTGGGGATGTTTGCTACCTCCTGCTATTTTTGTG

3361 GTTTTGGTTCTCCCACTATGGTAGGACCCCTGGCCAGCATTGTGGCTTGTCATGTCAGCC

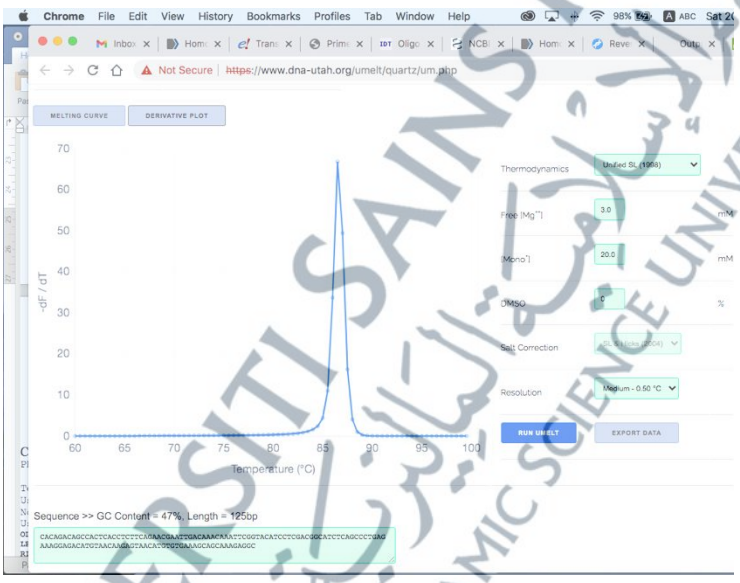
3421 CCATTGACTACCTTCTCATGCTCTGAGGTACTACTGCCTCTGCAGCACAAATTTCTATTT

3481 CTGTCAATAAAAGGAGATGAAAATATTC
```

```
KEYS (in order of precedence):
>>>>>> left primer
<<<<<< right primer
```

IDT OligoAnalyzer

	Forward	Reverse
Hairpin ΔG (kcal.mole-1)	-0.05	1.01
Self-dimer ΔG (kcal.mole-1)	-3.14	-3.61
Hetero-dimer ΔG (kcal.mole-1)	-3.14	



NCBI BLAST-PASSED

TERT

Forward primer is at exon 5 & 6, reverse primer at exon 6

```
PRIMER PICKING RESULTS FOR

Template masking selected
Using library: homo_sapiens with failure rate: 0.1
No mispriming library specified
Using 1-based sequence positions
OLIGO            start  len      tm     gc%  any_th  3'_th hairpin
seq
LEFT PRIMER       2205   19   59.32   57.89    0.00   0.00    0.00
TCAAGGTGGATGTGACGGG
RIGHT PRIMER      2277   20   60.25   55.00    0.00   0.00    0.00
ATGATGCTGGCGATGACCTC
SEQUENCE SIZE: 4039
INCLUDED REGION SIZE: 4039

PRODUCT SIZE: 73, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00
TARGETS (start len

    1 CTCTCCTCGCGGCGCGAGTTTCAGGCAGCGCTGCGTCCTGCTGCGCACGTGGGAAGCCCT

   61 GGCCCCGGCCACCCCCGCGATGCCGCGCGCTCCCCGCTGCCGAGCCGTGCGCTCCCTGCT

  121 GCGCAGCCACTACCGCGAGGTGCTGCCGCTGGCCACGTTCGTGCGGCGCCTGGGGCCCCA

  181 GGGCTGGCGGCTGGTGCAGCGCGGGGACCCGGCGGCTTTCCGCGCGCTGGTGGCCCAGTG

  241 CCTGGTGTGCGTGCCCTGGGACGCACGGCCGCCCCCCGCCGCCCCCTCCTTCCGCCAGGT

  301 GTCCTGCCTGAAGGAGCTGGTGGCCCGAGTGCTGCAGAGGCTGTGCGAGCGCGGCGCGAA

  361 GAACGTGCTGGCCTTCGGCTTCGCGCTGCTGGACGGGGCCCGCGGGGGCCCCCCCCGAGGC

  421 CTTCACCACCAGCGTGCGCAGCTACCTGCCCAACACGGTGACCGACGCACTGCGGGGGAG

  481 CGGGGCGTGGGGGCTGCTGCTGCGCCGCGTGGGCGACGACGTGCTGGTTCACCTGCTGGC

  541 ACGCTGCGCGCTCTTTGTGCTGGTGGCTCCCAGCTGCGCCTACCAGGTGTGCGGGCCGCC

  601 GCTGTACCAGCTCGGCGCTGCCACTCAGGCCCGGCCCCCGCCACACGCTAGTGGACCCCG

  661 AAGGCGTCTGGGATGCGAACGGGCCTGGAACCATAGCGTCAGGGAGGCCGGGGTCCCCCT

  721 GGGCCTGCCAGCCCCGGGTGCGAGGAGGCGCGGGGGCAGTGCCAGCCGAAGTCTGCCGTT

  781 GCCCAAGAGGCCCAGGCGTGGCGCTGCCCCTGAGCCGGAGCGGACGCCCGTTGGGCAGGG
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 841 GTCCTGGGCCCACCCGGGCAGGACGCGTGGACCGAGTGACCGTGGTTTCTGTGTGGTGTC
 901 ACCTGCCAGACCCGCCGAAGAAGCCACCTCTTTGGAGGGTGCGCTCTCTGGCACGCGCCA
 961 CTCCCACCCATCCGTGGGCCGCCAGCACCACGCGGGCCCCCCATCCACATCGCGGCCACC
1021 ACGTCCCTGGGACACGCCTTGTCCCCCGGTGTACGCCGAGACCAAGCACTTCCTCTACTC
1081 CTCAGGCGACAAGGAGCAGCTGCGGCCCTCCTTCCTACTCAGCTCTCTGAGGCCCAGCCT
1141 GACTGGCGCTCGGAGGCTCGTGGAGACCATCTTTCTGGGTTCCAGGCCCTGGATGCCAGG
1201 GACTCCCCGCAGGTTGCCCCGCCTGCCCCAGCGCTACTGGCAAATGCGGCCCCTGTTTCT
1261 GGAGCTGCTTGGGAACCACGCGCAGTGCCCCTACGGGGTGCTCCTCAAGACGCACTGCCC
1321 GCTGCGAGCTGCGGTCACCCCAGCAGCCGGTGTCTGTGCCCGGGAGAAGCCCCAGGGCTC
1381 TGTGGCGGCCCCCGAGGAGGAGGACACAGACCCCCGTCGCCTGGTGCAGCTGCTCCGCCA
1441 GCACAGCAGCCCCTGGCAGGTGTACGGCTTCGTGCGGGCCTGCCTGCGCCGGCTGGTGCC
1501 CCCAGGCCTCTGGGGCTCCAGGCACAACGAACGCCGCTTCCTCAGGAACACCAAGAAGTT
1561 CATCTCCCTGGGGAAGCATGCCAAGCTCTCGCTGCAGGAGCTGACGTGGAAGATGAGCGT
1621 GCGGGACTGCGCTTGGCTGCGCAGGAGCCCAGGGGTTGGCTGTGTTCCGGCCGCAGAGCA
1681 CCGTCTGCGTGAGGAGATCCTGGCCAAGTTCCTGCACTGGCTGATGAGTGTGTACGTCGT
1741 CGAGCTGCTCAGGTCTTTCTTTTATGTCACGGAGACCACGTTTCAAAAGAACAGGCTCTT
1801 TTTCTACCGGAAGAGTGTCTGGAGCAAGTTGCAAAGCATTGGAATCAGACAGCACTTGAA
1861 GAGGGTGCAGCTGCGGGAGCTGTCGGAAGCAGAGGTCAGGCAGCATCGGGAAGCCAGGCC
1921 CGCCCTGCTGACGTCCAGACTCCGCTTCATCCCCAAGCCTGACGGGCTGCGGCCGATTGT
1981 GAACATGGACTACGTCGTGGGAGCCAGAACGTTCCGCAGAGAAAAGAGGGCCGAGCGTCT
```

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2041 CACCTCGAGGGTGAAGGCACTGTTCAGCGTGCTCAACTACGAGCGGGCGCGGCGCCCCGG

2101 CCTCCTGGGCGCCTCTGTGCTGGGCCTGGACGATATCCACAGGGCCTGGCGCACCTTCGT

2161 GCTGCGTGTGCGGGCCCAGGACCCGCCGCCTGAGCTGTACTTTGTCAAGGTGGATGTGAC
                                                  >>>>>>>>>>>>>>>>

2221 GGGCGCGTACGACACCATCCCCCAGGACAGGCTCACGGAGGTCATCGCCAGCATCATCAA
     >>>         ********                  <<<<<<<<<<<<<<<<<<<<

2281 ACCCCAGAACACGTACTGCGTGCGTCGGTATGCCGTGGTCCAGAAGGCCGCCCATGGGCA

2341 CGTCCGCAAGGCCTTCAAGAGCCACGTCTCTACCTTGACAGACCTCCAGCCGTACATGCG

2401 ACAGTTCGTGGCTCACCTGCAGGAGACCAGCCCGCTGAGGATGCCGTCGTCATCGAGCA

2461 GAGCTCCTCCCTGAATGAGGCCAGCAGTGGCCTCTTCGACGTCTTCCTACGCTTCATGTG

2521 CCACCACGCCGTGCGCATCAGGGGCAAGTCCTACGTCCAGTGCCAGGGGATCCCGCAGGG

2581 CTCCATCCTCTCCACGCTGCTCTGCAGCCTGTGCTACGGCGACATGGAGAACAAGCTGTT

2641 TGCGGGGATTCGGCGGGACGGGCTGCTCCTGCGTTTGGTGGATGATTTCTTGTTGGTGAC

2701 ACCTCACCTCACCCACGCGAAAACCTTCCTCAGGACCCTGGTCCGAGGTGTCCCTGAGTA

2761 TGGCTGCGTGGTGAACTTGCGGAAGACAGTGGTGAACTTCCCTGTAGAAGACGAGGCCCT

2821 GGGTGGCACGGCTTTTGTTCAGATGCCGGCCCACGGCCTATTCCCCTGGTGCGGCCTGCT

2881 GCTGGATACCCGGACCCTGGAGGTGCAGAGCGACTACTCCAGCTATGCCCGGACCTCCAT

2941 CAGAGCCAGTCTCACCTTCAACCGCGGCTTCAAGGCTGGGAGGAACATGCGTCGCAAACT

3001 CTTTGGGGTCTTGCGGCTGAAGTGTCACAGCCTGTTTCTGGATTTGCAGGTGAACAGCCT

3061 CCAGACGGTGTGCACCAACATCTACAAGATCCTCCTGCTGCAGGCGTACAGGTTTCACGC

3121 ATGTGTGCTGCAGCTCCCATTTCATCAGCAAGTTTGGAAGAACCCCACATTTTTCCTGCG

3181 CGTCATCTCTGACACGGCCTCCCTCTGCTACTCCATCCTGAAAGCCAAGAACGCAGGGAT
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3241 GTCGCTGGGGGCCAAGGGCGCCGCCGGCCCTCTGCCCTCCGAGGCCGTGCAGTGGCTGTG

3301 CCACCAAGCATTCCTGCTCAAGCTGACTCGACACCGTGTCACCTACGTGCCACTCCTGGG

3361 GTCACTCAGGACAGCCCAGACGCAGCTGAGTCGGAAGCTCCCGGGGACGACGCTGACTGC

3421 CCTGGAGGCCGCAGCCAACCCGGCACTGCCCTCAGACTTCAAGACCATCCTGGACTGATG

3481 GCCACCCGCCCACAGCCAGGCCGAGAGCAGACACCAGCAGCCCTGTCACGCCGGGCTCTA

3541 CGTCCCAGGGAGGGAGGGGCGGCCCACACCCAGGCCCGCACCGCTGGGAGTCTGAGGCCT

3601 GAGTGAGTGTTTGGCCGAGGCCTGCATGTCCGGCTGAAGGCTGAGTGTCCGGCTGAGGCC

3661 TGAGCGAGTGTCCAGCCAAGGGCTGAGTGTCCAGCACACCTGCCGTCTTCACTTCCCCAC

3721 AGGCTGGCGCTCGGCTCCACCCCAGGGCCAGCTTTTCCTCACCAGGAGCCCGGCTTCCAC

3781 TCCCCACATAGGAATAGTCCATCCCCAGATTCGCCATTGTTCACCCCTCGCCCTGCCCTC

3841 CTTTGCCTTCCACCCCCACCATCCAGGTGGAGACCCTGAGAAGGACCCTGGGAGCTCTGG

3901 GAATTTGGAGTGACCAAAGGTGTGCCCTGTACACAGGCGAGGACCCTGCACCTGGATGGG

3961 GGTCCCTGTGGGTCAAATTGGGGGGAGGTGCTGTGGGAGTAAAATACTGAATATATGAGT

4021 TTTTCAGTTTTGAAAAAAA
```

KEYS (in order of precedence):
****** target
>>>>>> left primer
<<<<<< right primer

ADDITIONAL OLIGOS

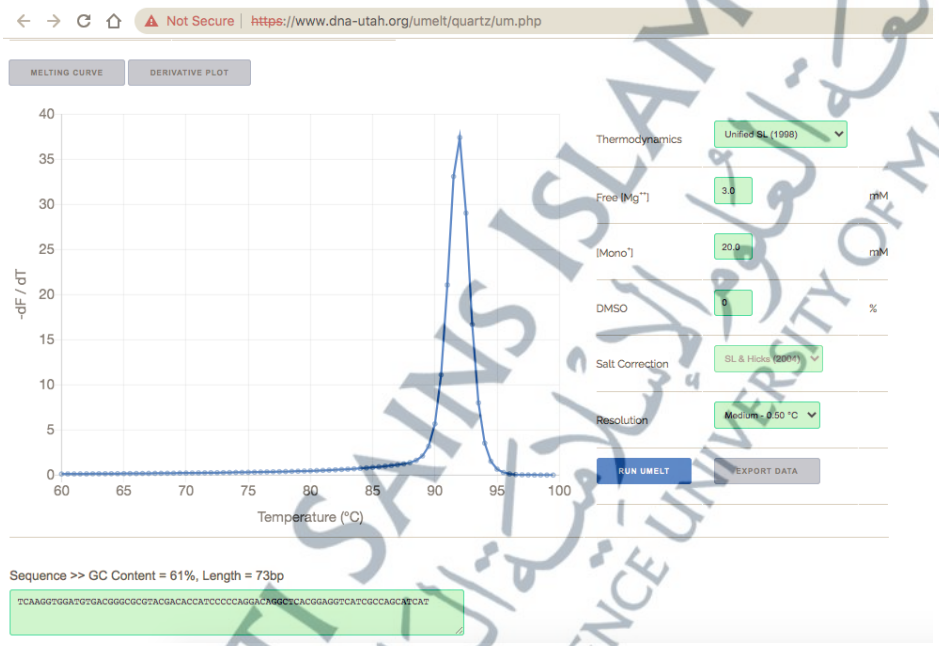
	start	len	tm	gc%	any th	3' th	hairpin	seq
1 LEFT PRIMER	2119	19	60.00	63.16	12.70	0.00	0.00	GCTGGGCCTGGACGATATC
RIGHT PRIMER	2266	19	59.86	63.16	0.00	0.00	0.00	GATGACCTCCGTGAGCCTG
PRODUCT SIZE: 148, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 2.93								
2 LEFT PRIMER	2213	18	61.25	66.67	29.59	24.02	0.00	GATGTGACGGGCGCGTAC
RIGHT PRIMER	2326	20	60.46	60.00	0.00	0.00	0.00	CTTCTGGACCACGGCATACC
PRODUCT SIZE: 114, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00								

```
 3 LEFT PRIMER        2129   19   58.56   57.89    0.00   0.00    0.00
GACGATATCCACAGGGCCT
   RIGHT PRIMER       2269   18   59.89   66.67    0.00   0.00    0.00
GGCGATGACCTCCGTGAG
   PRODUCT SIZE: 141, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00

 4 LEFT PRIMER        2113   19   62.62   68.42   24.79  24.79    0.00
CTCTGTGCTGGGCCTGGAC
   RIGHT PRIMER       2260   18   59.42   66.67    0.00   0.00    0.00
CTCCGTGAGCCTGTCCTG
   PRODUCT SIZE: 148, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00
```

IDT OligoAnalyzer

	Forward	Reverse
Hairpin ΔG (kcal.mole-1)	0.82	-0.12
Self-dimer ΔG (kcal.mole-1)	-3.61	-3.61
Hetero-dimer ΔG (kcal.mole-1)	-6.01	



NCBI BLAST- PASSED

TERT
Primer3 Output

PRIMER PICKING RESULTS FOR NM_198253.3 Homo sapiens telomerase reverse transcriptase (TERT), transcript variant 1, mRNA

Template masking selected
Using library: homo_sapiens with failure rate: 0.1
Using mispriming library humrep_and_simple.txt
Using 1-based sequence positions

OLIGO	start	len	tm	gc%	any_th	3'_th	hairpin	rep	seq
LEFT PRIMER	2058	20	60.04	55.00	1.33	1.33	0.00	10.00	CACTGTTCAGCGTGCTCAAC
RIGHT PRIMER	2148	20	59.97	60.00	1.39	0.00	0.00	10.00	CAGGCCCTGTGGATATCGTC

SEQUENCE SIZE: 4039
INCLUDED REGION SIZE: 4039

PRODUCT SIZE: 91, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00
PRODUCT Tm: 90.7012, PRODUCT Tm - min(OLIGO Tm): 30.7353

```
   1 CTCTCCTCGCGGCGCGAGTTTCAGGCAGCGCTGCGTCCTGCTGCGCACGTGGGAAGCCCT

  61 GGCCCCGGCCACCCCCGCGATGCCGCGCGCTCCCCGCTGCCGAGCCGTGCGCTCCCTGCT

 121 GCGCAGCCACTACCGCGAGGTGCTGCCGCTGGCCACGTTCGTGCGGCGCCTGGGGCCCCA

 181 GGGCTGGCGGCTGGTGCAGCGCGGGGACCCGGCGGCTTTCCGCGCGCTGGTGGCCCAGTG

 241 CCTGGTGTGCGTGCCCTGGGACGCACGGCCGCCCCCCGCCGCCCCCTCCTTCCGCCAGGT

 301 GTCCTGCCTGAAGGAGCTGGTGGCCCGAGTGCTGCAGAGGCTGTGCGAGCGCGGCGCGAA

 361 GAACGTGCTGGCCTTCGGCTTCGCGCTGCTGGACGGGGCCCGCGGGGGCCCCCCCGAGGC

 421 CTTCACCACCAGCGTGCGCAGCTACCTGCCCAACACGGTGACCGACGCACTGCGGGGGAG

 481 CGGGGCGTGGGGGCTGCTGCTGCGCCGCGTGGGCGACGACGTGCTGGTTCACCTGCTGGC

 541 ACGCTGCGCGCTCTTTGTGCTGGTGGCTCCCAGCTGCGCCTACCAGGTGTGCGGGCCGCC

 601 GCTGTACCAGCTCGGCGCTGCCACTCAGGCCCGGCCCCCGCCACACGCTAGTGGACCCCG

 661 AAGGCGTCTGGGATGCGAACGGGCCTGGAACCATAGCGTCAGGGAGGCCGGGGTCCCCCT

 721 GGGCCTGCCAGCCCCGGGTGCGAGGAGGCGCGGGGGCAGTGCCAGCCGAAGTCTGCCGTT
```

781 GCCCAAGAGGCCCAGGCGTGGCGCTGCCCCTGAGCCGGAGCGGACGCCCGTTGGGCAGGG

841 GTCCTGGGCCCACCCGGGCAGGACGCGTGGACCGAGTGACCGTGGTTTCTGTGTGGTGTC

901 ACCTGCCAGACCCGCCGAAGAAGCCACCTCTTTGGAGGGTGCGCTCTCTGGCACGCGCCA

961 CTCCCACCCATCCGTGGGCCGCCAGCACCACGCGGGCCCCCCATCCACATCGCGGCCACC

1021 ACGTCCCTGGGACACGCCTTGTCCCCCGGTGTACGCCGAGACCAAGCACTTCCTCTACTC

1081 CTCAGGCGACAAGGAGCAGCTGCGGCCCTCCTTCCTACTCAGCTCTCTGAGGCCCAGCCT

1141 GACTGGCGCTCGGAGGCTCGTGGAGACCATCTTTCTGGGTTCCAGGCCCTGGATGCCAGG

1201 GACTCCCCGCAGGTTGCCCCGCCTGCCCCAGCGCTACTGGCAAATGCGGCCCCTGTTTCT

1261 GGAGCTGCTTGGGAACCACGCGCAGTGCCCCTACGGGGTGCTCCTCAAGACGCACTGCCC

1321 GCTGCGAGCTGCGGTCACCCCAGCAGCCGGTGTCTGTGCCCGGGAGAAGCCCCAGGGCTC

1381 TGTGGCGGCCCCCGAGGAGGAGGACACAGACCCCCGTCGCCTGGTGCAGCTGCTCCGCCA

1441 GCACAGCAGCCCCTGGCAGGTGTACGGCTTCGTGCGGGCCTGCCTGCGCCGGCTGGTGCC

1501 CCCAGGCCTCTGGGGCTCCAGGCACAACGAACGCCGCTTCCTCAGGAACACCAAGAAGTT

1561 CATCTCCCTGGGGAAGCATGCCAAGCTCTCGCTGCAGGAGCTGACGTGGAAGATGAGCGT

1621 GCGGGACTGCGCTTGGCTGCGCAGGAGCCCAGGGGTTGGCTGTGTTCCGGCCGCAGAGCA

1681 CCGTCTGCGTGAGGAGATCCTGGCCAAGTTCCTGCACTGGCTGATGAGTGTGTACGTCGT

1741 CGAGCTGCTCAGGTCTTTCTTTTATGTCACGGAGACCACGTTTCAAAAGAACAGGCTCTT

1801 TTTCTACCGGAAGAGTGTCTGGAGCAAGTTGCAAAGCATTGGAATCAGACAGCACTTGAA

1861 GAGGGTGCAGCTGCGGGAGCTGTCGGAAGCAGAGGTCAGGCAGCATCGGGAAGCCAGGCC

1921 CGCCCTGCTGACGTCCAGACTCCGCTTCATCCCCAAGCCTGACGGGCTGCGGCCGATTGT

```
1981 GAACATGGACTACGTCGTGGGAGCCAGAACGTTCCGCAGAGAAAAGAGGGCCGAGCGTCT

2041 CACCTCGAGGGTGAAGGCACTGTTCAGCGTGCTCAACTACGAGCGGGCGCGGCGCCCCGG
                     >>>>>>>>>>>>>>>>>>>>

2101 CCTCCTGGGCGCCTCTGTGCTGGGCCTGGACGATATCCACAGGGCCTGGCGCACCTTCGT
                                  <<<<<<<<<<<<<<<<<<<<

2161 GCTGCGTGTGCGGGCCCAGGACCCGCCGCCTGAGCTGTACTTTGTCAAGGTGGATGTGAC

2221 GGGCGCGTACGACACCATCCCCCAGGACAGGCTCACGGAGGTCATCGCCAGCATCATCAA

2281 ACCCCAGAACACGTACTGCGTGCGTCGGTATGCCGTGGTCCAGAAGGCCGCCCATGGGCA

2341 CGTCCGCAAGGCCTTCAAGAGCCACGTCTCTACCTTGACAGACCTCCAGCCGTACATGCG

2401 ACAGTTCGTGGCTCACCTGCAGGAGACCAGCCCGCTGAGGGATGCCGTCGTCATCGAGCA

2461 GAGCTCCTCCCTGAATGAGGCCAGCAGTGGCCTCTTCGACGTCTTCCTACGCTTCATGTG

2521 CCACCACGCCGTGCGCATCAGGGGCAAGTCCTACGTCCAGTGCCAGGGGATCCCGCAGGG

2581 CTCCATCCTCTCCACGCTGCTCTGCAGCCTGTGCTACGGCGACATGGAGAACAAGCTGTT

2641 TGCGGGGATTCGGCGGGACGGGCTGCTCCTGCGTTTGGTGGATGATTTCTTGTTGGTGAC

2701 ACCTCACCTCACCCACGCGAAAACCTTCCTCAGGACCCTGGTCCGAGGTGTCCCTGAGTA

2761 TGGCTGCGTGGTGAACTTGCGGAAGACAGTGGTGAACTTCCCTGTAGAAGACGAGGCCCT

2821 GGGTGGCACGGCTTTTGTTCAGATGCCGGCCCACGGCCTATTCCCCTGGTGCGGCCTGCT

2881 GCTGGATACCCGGACCCTGGAGGTGCAGAGCGACTACTCCAGCTATGCCCGGACCTCCAT

2941 CAGAGCCAGTCTCACCTTCAACCGCGGCTTCAAGGCTGGGAGGAACATGCGTCGCAAACT

3001 CTTTGGGGTCTTGCGGCTGAAGTGTCACAGCCTGTTTCTGGATTTGCAGGTGAACAGCCT

3061 CCAGACGGTGTGCACCAACATCTACAAGATCCTCCTGCTGCAGGCGTACAGGTTTCACGC

3121 ATGTGTGCTGCAGCTCCCATTTCATCAGCAAGTTTGGAAGAACCCCACATTTTTCCTGCG
```

3181 CGTCATCTCTGACACGGCCTCCCTCTGCTACTCCATCCTGAAAGCCAAGAACGCAGGGAT

3241 GTCGCTGGGGGCCAAGGGCGCCGCCGGCCCTCTGCCCTCCGAGGCCGTGCAGTGGCTGTG

3301 CCACCAAGCATTCCTGCTCAAGCTGACTCGACACCGTGTCACCTACGTGCCACTCCTGGG

3361 GTCACTCAGGACAGCCCAGACGCAGCTGAGTCGGAAGCTCCCGGGGACGACGCTGACTGC

3421 CCTGGAGGCCGCAGCCAACCCGGCACTGCCCTCAGACTTCAAGACCATCCTGGACTGATG

3481 GCCACCCGCCCACAGCCAGGCCGAGAGCAGACACCAGCAGCCCTGTCACGCCGGGCTCTA

3541 CGTCCCAGGGAGGGAGGGGCGGCCCACACCCAGGCCCGCACCGCTGGGAGTCTGAGGCCT

3601 GAGTGAGTGTTTGGCCGAGGCCTGCATGTCCGGCTGAAGGCTGAGTGTCCGGCTGAGGCC

3661 TGAGCGAGTGTCCAGCCAAGGGCTGAGTGTCCAGCACACCTGCCGTCTTCACTTCCCCAC

3721 AGGCTGGCGCTCGGCTCCACCCCAGGGCCAGCTTTTCCTCACCAGGAGCCCGGCTTCCAC

3781 TCCCCACATAGGAATAGTCCATCCCCAGATTCGCCATTGTTCACCCCTCGCCCTGCCCTC

3841 CTTTGCCTTCCACCCCCACCATCCAGGTGGAGACCCTGAGAAGGACCCTGGGAGCTCTGG

3901 GAATTTGGAGTGACCAAAGGTGTGCCCTGTACACAGGCGAGGACCCTGCACCTGGATGGG

3961 GGTCCCTGTGGGTCAAATTGGGGGGAGGTGCTGTGGGAGTAAAATACTGAATATATGAGT

4021 TTTTCAGTTTTGAAAAAAA

KEYS (in order of precedence):
>>>>>> left primer
<<<<<< right primer

ADDITIONAL OLIGOS

	start	len	tm	gc%	any_th	3'_th	hairpin	rep	seq
1 LEFT PRIMER	2759	20	59.96	50.00	0.00	0.00	0.00	10.00	TATGGCTGCGTGGTGAACTT
RIGHT PRIMER	2851	20	60.11	55.00	30.85	0.00	0.00	11.00	GGCCGGCATCTGAACAAAAG

PRODUCT SIZE: 93, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00
PRODUCT Tm: 85.8143, PRODUCT Tm - min(OLIGO Tm): 25.8505

2 LEFT PRIMER	2614	20	59.83	55.00	16.62	0.00	0.00	11.00	CTACGGCGACATGGAGAACA
RIGHT PRIMER	2778	20	59.96	50.00	0.00	0.00	0.00	10.00	AAGTTCACCACGCAGCCATA

```
   PRODUCT SIZE: 165, PAIR ANY_TH COMPL: 20.59, PAIR 3'_TH COMPL: 14.67
PRODUCT Tm: 88.6776, PRODUCT Tm - min(OLIGO Tm): 28.8502

 3 LEFT PRIMER       1599   20   60.11   55.00    0.00   0.00    0.00 11.00
AGCTGACGTGGAAGATGAGC
   RIGHT PRIMER      1712   20   60.11   60.00   23.57  18.73    0.00 10.00
GGAACTTGGCCAGGATCTCC
   PRODUCT SIZE: 114, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00
PRODUCT Tm: 90.1700, PRODUCT Tm - min(OLIGO Tm): 30.0630

 4 LEFT PRIMER       3595   20   59.89   55.00    3.11   0.00    0.00 10.00
AGGCCTGAGTGAGTGTTTGG
   RIGHT PRIMER      3791   20   59.82   60.00    0.00   0.00    0.00 11.00
CTATGTGGGGAGTGGAAGCC
   PRODUCT SIZE: 197, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00
PRODUCT Tm: 92.0534, PRODUCT Tm - min(OLIGO Tm): 32.2347
```

IDT OligoAnalyzer

	Forward	Reverse
Hairpin ΔG (kcal.mole-1)	0.37	0.91
Self-dimer ΔG (kcal.mole-1)	-5.38	-3.61
Hetero-dimer ΔG (kcal.mole-1)	-6.75	

Primer from (Y. Zhang et al., 2012)

air 1

	Sequence (5'->3')	Length	Tm	GC%	Self complementarity	Self 3' complementarity
Forward primer	ATGCGACAGTTCGTGGCTCA	20	62.43	55.00	3.00	2.00
Reverse primer	ATCCCCTGGCACTGGACGTA	20	62.82	60.00	5.00	2.00

Products on target templates

>NM_001193376.3 Homo sapiens telomerase reverse transcriptase (TERT), transcript variant 2, mRNA

```
product length = 176
Forward primer  1     ATGCGACAGTTCGTGGCTCA  20
Template        2396  ....................  2415

Reverse primer  1     ATCCCCTGGCACTGGACGTA  20
Template        2571  ....................  2552
```

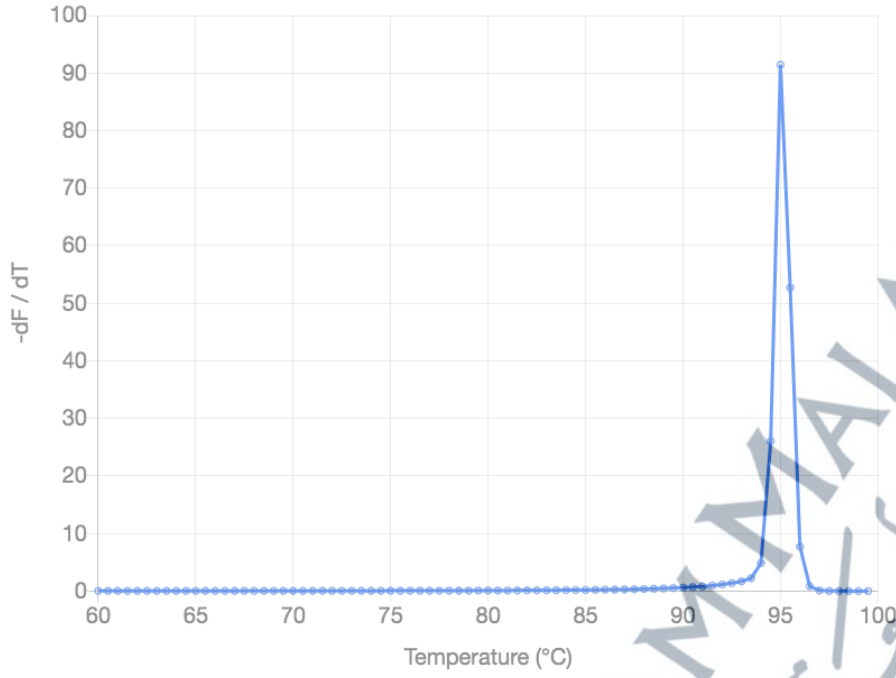
>NM_198253.3 Homo sapiens telomerase reverse transcriptase (TERT), transcript variant 1, mRNA

```
product length = 176
Forward primer  1     ATGCGACAGTTCGTGGCTCA  20
Template        2396  ....................  2415

Reverse primer  1     ATCCCCTGGCACTGGACGTA  20
Template        2571  ....................  2552
```

IDT OligoAnalyzer

	Forward	Reverse
Hairpin ΔG (kcal.mole-1)	-1.56	0.01
Self-dimer ΔG (kcal.mole-1)	-5.19	-6.3
Hetero-dimer ΔG (kcal.mole-1)	-5.09	



Sequence >> GC Content = 62%, Length = 176bp

ATGCGACAGTTCGTGGCTCACCTGCAGGAGACCAGCCCGCTGAGGGATGC
CGTCGTCATCGAGCAGAGCTCCTCCCTGAATGAGGCCAGCAGTGGCCTCTT
CGACGTCTTCCTACGCTTCATGTGCCACCACGCCGTGCGCATCAGGGGCAA
GTCCTACGTCCAGTGCCAGGGGAT

Appendix 4: Presentations and Publications

1. Syarifah Faezah Syed Mohamad and Marjanu Hikmah Elias. 2020. MicroRNA as a Potential Chronic Myeloid Leukaemia Treatment: An In-Silico Analysis. 4th USIM International Health E-Conference 2020 (IHEC 2020) In Conjunction with the 3rd International Conference on Medicine And Health Sciences (ICMHS). 16-17 December 2020. Oral presenter.

2. Syarifah Faezah Syed Mohamad and Marjanu Hikmah Elias. Effect of hsa-miR-3131 transfection upon cell viability in imatinib sensitive and imatinib resistance chronic myeloid leukaemia cells. 4th International Conference on Molecular Biology & Biotechnology 2021 (ICMBB2021). 1-3 June 2021. Poster presenter.

3. Syarifah Faezah Syed Mohamad and Saida Farhanah Sarkam. Bibliometric Analysis of MicroRNAs Associated with Chronic Myeloid Leukemia. 6th International Conference on Molecular Biology & Biotechnology (ICMBB2023). 7-9 June 2023. Poster presenter.

4. Mohamad, S. F. S., & Elias, M. H. (2021). MicroRNA as a potential chronic myeloid leukaemia treatment: an in-silico analysis. *In Malaysian Journal of Medicine and Health Sciences* (Vol. 17). Universiti Putra Malaysia. https://myjurnal.mohe.gov.my/public/article-view.php?id=175451

5. Mohamad, S. F. S., & Elias, M. H. (2021). *Potential treatment for chronic myeloid leukemia using microRNA: in silico comparison between plants and human microRNAs in targeting BCR-ABL1 gene*. Egyptian Journal of Medical Human Genetics, 22, 1-8.

6. Mohamad, S. F. S., & Elias, M. H. (2023). Bibliometric analysis of microRNAs associated with chronic myeloid leukemia. *In Asia Pacific Journal of Molecular Biology and Biotechnology* Vol. 31(2) (Suppl.) https://www.msmbb.my/images/publication/volume_31/issue_2/APJMBB-Volume-312-Supplementary.pdf

7. Elias, M. H., Syed Mohamad, S. F., & Abdul Hamid, N. (2022). *A systematic review of candidate miRNAs, its targeted genes and pathways in chronic myeloid leukemia–an integrated bioinformatical analysis*. Frontiers in Oncology, 12, 848199.