Primer3 Output

```
PRIMER PICKING RESULTS FOR
Template masking not selected
No mispriming library specified
No internal oligo mishyb library specified
Using 1-based sequence positions
0LIG0
                 start len
                                           any th 3' th hairpin seq
                                tm
                                       qc%
                   38
                         20
                              58.83
                                     55.00
                                              0.00
                                                     0.00
LEFT PRIMER
                                                             0.00 gttggtgaaggacatgaggc
RIGHT PRIMER
                   219
                        20
                              59.17
                                     50.00
                                              0.00
                                                     0.00
                                                             0.00 tcagcacttgactcacacca
INTERNAL OLIGO
                   125
                        20
                             59.89
                                     50.00
                                              0.00
                                                     0.00
                                                             0.00 catcacatcgggtttgcgat
SEQUENCE SIZE: 301
INCLUDED REGION SIZE: 301
PRODUCT SIZE: 182, PAIR ANY TH COMPL: 0.00, PAIR 3' TH COMPL: 2.93
   1 ctttgatggcgcaggttttccgtttgaagttccaacagttggtgaaggacatgaggcagt
                                          >>>>>>>>>
  61 acctccaccqatqcgtggagactggtaaggacttcaacattgcgctcgcggtcaagacaa
  121 atatcatcacatcgggtttgcgatactgcctggcgactggaaattggggtgaccaaaaga
          ^^^^^^
  181 aggccgcaagcgcaaaagctggtgtgagtcaagtgctgaacagatacacctacgcctcga
                         <<<<<<<<
  241 ccttgagtcatttgcggcgaacgaacacccccatcggtcgtgacggcaagattgcgaagc
  301 c
KEYS (in order of precedence):
>>>>> left primer
<<<<< right primer
^^^^^ internal oligo
ADDITIONAL OLIGOS
                   start len _____tm ___gc% _any th _3' th hairpin seq
 1 LEFT PRIMER
                      71
                           20
                                59.46
                                        55.00
                                                 0.00
                                                        0.00
                                                                0.00 atgcgtggagactggtaagg
   RIGHT PRIMER
                     239
                           20
                                58.98
                                        55.00
                                                 0.00
                                                        0.00
                                                                0.00 cgaggcgtaggtgtatctgt
                     200
   INTERNAL OLIGO
                           20
                                60.10
                                        50.00
                                                 0.00
                                                        0.00
                                                                0.00 tggtgtgagtcaagtgctga
  PRODUCT SIZE: 169, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00
 2 LEFT PRIMER
                       88
                                58.84
                           20
                                        50.00
                                                 9.73
                                                        0.00
                                                                0.00 aggacttcaacattgcgctc
```

	RIGHT PRIMER INTERNAL OLIGO PRODUCT SIZE: 174,	200 20 6	59.48 50.00 60.10 50.00 COMPL: 0.00,		0.00	ttcgccgcaaatgactcaag tggtgtgagtcaagtgctga
3	LEFT PRIMER RIGHT PRIMER INTERNAL OLIGO PRODUCT SIZE: 173,	173 20 5 125 20 5	58.30 50.00 58.83 55.00 59.89 50.00 COMPL: 0.00,	0.00 0.00	0.00	ctttgatggcgcaggttttc gtcaccccaatttccagtcg catcacatcgggtttgcgat
4	LEFT PRIMER RIGHT PRIMER INTERNAL OLIGO PRODUCT SIZE: 170,	294 19 5 200 20 6	58.98 50.00 59.21 57.89 50.10 50.00 COMPL: 0.00,	0.00 0.00 0.00 0.00	0.00	catcacatcgggtttgcgat caatcttgccgtcacgacc tggtgtgagtcaagtgctga
5	LEFT PRIMER RIGHT PRIMER INTERNAL OLIGO PRODUCT SIZE: 169,	242 20 5 200 20 6	58.45 55.00 59.69 60.00 50.10 50.00 COMPL: 0.00,	0.00 0.00 0.00 0.00	0.00	cgtggagactggtaaggact ggtcgaggcgtaggtgtatc tggtgtgagtcaagtgctga
6	LEFT PRIMER RIGHT PRIMER INTERNAL OLIGO PRODUCT SIZE: 175,	215 20 5 125 20 5	59.67 55.00 68.04 50.00 69.89 50.00 COMPL: 6.44,	0.00 0.00 0.00 0.00	0.00	ggtgaaggacatgaggcagt cacttgactcacaccagctt catcacatcgggtttgcgat
7	LEFT PRIMER RIGHT PRIMER INTERNAL OLIGO PRODUCT SIZE: 167,	235 21 5 125 20 5	59.19 57.89 58.81 52.38 59.89 50.00 COMPL: 0.00,	0.00 0.00 0.00 0.00	0.00	cgatgcgtggagactggta gcgtaggtgtatctgttcagc catcacatcgggtttgcgat
8	LEFT PRIMER RIGHT PRIMER INTERNAL OLIGO PRODUCT SIZE: 160,	263 18 5 200 20 6	58.64 50.00 59.14 55.56 50.10 50.00 COMPL: 5.44,	0.00 0.00 0.00 0.00	0.00	gctcgcggtcaagacaaata cgttcgccgcaaatgact tggtgtgagtcaagtgctga
9	LEFT PRIMER RIGHT PRIMER INTERNAL OLIGO PRODUCT SIZE: 171,	189 19 5 125 20 5	58.01 42.86 58.96 52.63 59.89 50.00 COMPL: 0.00,	0.00 0.00 0.00 0.00	0.00	tccgtttgaagttccaacagt ttgcggccttcttttggtc catcacatcgggtttgcgat
10	LEFT PRIMER RIGHT PRIMER INTERNAL OLIGO PRODUCT SIZE: 164,	292 20 6 200 20 6	58.35 50.00 51.95 55.00 50.10 50.00 COMPL: 0.00,	0.00 0.00 0.00 0.00	0.00	acatcgggtttgcgatactg atcttgccgtcacgaccgat tggtgtgagtcaagtgctga
11	LEFT PRIMER RIGHT PRIMER INTERNAL OLIGO PRODUCT SIZE: 173,	247 19 5 200 20 6	59.10 50.00 59.57 63.16 50.10 50.00 COMPL: 0.00,	0.00 0.00 0.00 0.00	0.00	gtggagactggtaaggacttca ctcaaggtcgaggcgtagg tggtgtgagtcaagtgctga
12	LEFT PRIMER RIGHT PRIMER INTERNAL OLIGO	244 19 6	69.11 61.11 60.61 57.89 60.10 50.00	0.00 0.00	0.00	gtacctccaccgatgcgt aaggtcgaggcgtaggtgt tggtgtgagtcaagtgctga

PRODUCT SIZE: 186, PAIR ANY TH COMPL: 8.19, PAIR 3' TH COMPL: 8.19 13 LEFT PRIMER 21 0.00 0.00 0.00 aacagttggtgaaggacatga 34 57.42 42.86 22 RIGHT PRIMER 214 59.32 45.45 0.00 0.00 acttgactcacaccagcttttg 0.00 20 59.89 INTERNAL OLIGO 125 50.00 0.00 0.00 0.00 catcacatcgggtttgcgat PRODUCT SIZE: 181, PAIR ANY_TH COMPL: 9.87, PAIR 3'_TH COMPL: 1.77 Statistics high high high too in in tm high con not no tm GC too any_th 3'_th hairsid many tar excl ok bad too poly end GC% clamp low high compl compl stab ered Ns get reg reg pin Χ ok Õ 852 276 254 214 Left 0 5 103 0 0 778 135 79 235 Right 0 0 0 10 0 300 0 19 0 2795 0 331 1584 22 858 Intl 0 0 0 0 0 0 0 0 0

Pair Stats:

considered 6612, unacceptable product size 6596, primer in pair overlaps a primer in a better pair 15289, ok 16 libprimer3 release 2.4.0

(primer3_results.cgi release 4.1.0)