

PRIMER PICKING RESULTS FOR *Puccinia_recondita*_consensus_consensus_group_1

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Template masking not selected
No mispriming library specified
No internal oligo mishyb library specified
Using 1-based sequence positions

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OLIGO	<u>start</u>	<u>len</u>	<u>tm</u>	<u>gc%</u>	<u>any th</u>	<u>3' th</u>	<u>hairpin</u>	<u>seq</u>
LEFT PRIMER	76	20	59.39	50.00	17.19	0.00	0.00	aaggagctcacatacgccaa
RIGHT PRIMER	237	20	59.08	50.00	0.00	0.00	0.00	aagagtgtgggtggttgaa
INTERNAL OLIGO	165	20	60.03	50.00	13.59	0.00	38.28	cattctgcgaagggtcaagt

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SEQUENCE SIZE: 547
INCLUDED REGION SIZE: 547
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PRODUCT SIZE: 162, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00

1 tcccgccccgacatccaagacgaactcctccaagaacaaaaggatatgctcggNgtgaac

61 gccgacggcagtatcaaggagctcacatacgccaacctctcgaaactNaccctcctcaat
 >>>>>>>>>>>>>>>>>>

121 caagtcgtcaaagaaacccttcgtattcacgNtccaNtccactccattctgcgaagggtc

[illegible]

241 gccgctccgggcacaacgagccgcatggacgagcactttcccgactgcctccattgggag

301 ccgcatcgatgggacgagagcccggtccgagaaatacaagcacctgtccccNacgactgcc

361 ctaggNagcatcgccgaggagaaagaagacNNNgNcNNNggcctNgtaagcaagggcgcg

421 gcgtcgccatacNtaccctttggtgcgggacgacacagatgtatcggcgagcaattcgcg

481 tacgtgcaattgcagaccattacagcgacgatggttcgcgatttcaagttttacaatgtg

541 gatNgca

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

ADDITIONAL OLIGOS

	<u>start</u>	<u>len</u>	<u>tm</u>	<u>gc%</u>	<u>any_th</u>	<u>3' th</u>	<u>hairpin</u>	<u>seq</u>
1 LEFT PRIMER	316	20	58.97	55.00	0.00	0.00	0.00	gagagcccggtccgagaaata
RIGHT PRIMER	509	20	58.56	50.00	0.00	0.00	0.00	gtcgctgtaatggtctgcaa
INTERNAL OLIGO	371	20	60.38	55.00	0.00	0.00	0.00	tcgccgaggagaaagaagac
PRODUCT SIZE: 194,	PAIR	ANY_TH	COMPL: 0.00,	PAIR	3'_TH	COMPL: 0.00		
2 LEFT PRIMER	174	20	58.54	55.00	0.00	0.00	0.00	caaggtcaagtctcccatgc
RIGHT PRIMER	335	20	58.97	55.00	0.00	0.00	0.00	tatttctcggacgggctctc
INTERNAL OLIGO	218	20	60.02	50.00	0.00	0.00	0.00	ttccaaccacccacactctt
PRODUCT SIZE: 162,	PAIR	ANY_TH	COMPL: 0.00,	PAIR	3'_TH	COMPL: 0.00		
3 LEFT PRIMER	214	20	59.04	55.00	0.00	0.00	0.00	gtcattccaaccacccacac
RIGHT PRIMER	390	20	59.47	55.00	10.14	10.14	0.00	gtcttctttctcctcggcga
INTERNAL OLIGO	319	20	60.03	50.00	0.00	0.00	0.00	agcccgtccgagaaatacaa
PRODUCT SIZE: 177,	PAIR	ANY_TH	COMPL: 0.00,	PAIR	3'_TH	COMPL: 0.00		
4 LEFT PRIMER	179	20	59.45	55.00	0.00	0.00	0.00	tcaagtctcccatgcccac
RIGHT PRIMER	338	20	59.10	50.00	0.00	0.00	0.00	ttgtatttctcggacgggct
INTERNAL OLIGO	218	20	60.02	50.00	0.00	0.00	0.00	ttccaaccacccacactctt
PRODUCT SIZE: 160,	PAIR	ANY_TH	COMPL: 2.78,	PAIR	3'_TH	COMPL: 0.00		
5 LEFT PRIMER	65	20	59.54	55.00	4.71	4.71	0.00	acggcagtatcaaggagctc
RIGHT PRIMER	233	20	59.04	55.00	0.00	0.00	0.00	gtgtgggtggttggaatgac
INTERNAL OLIGO	85	20	60.03	50.00	0.00	0.00	0.00	acatacgccaacctctcgaa
PRODUCT SIZE: 169,	PAIR	ANY_TH	COMPL: 0.00,	PAIR	3'_TH	COMPL: 0.00		
6 LEFT PRIMER	9	20	58.37	55.00	0.00	0.00	0.00	cgacatccaagacgaactcc
RIGHT PRIMER	185	20	58.84	50.00	11.31	0.00	0.00	gacttgaccttgcgcagaat
INTERNAL OLIGO	85	20	60.03	50.00	0.00	0.00	0.00	acatacgccaacctctcgaa
PRODUCT SIZE: 177,	PAIR	ANY_TH	COMPL: 0.00,	PAIR	3'_TH	COMPL: 0.00		
7 LEFT PRIMER	14	20	58.29	50.00	0.00	0.00	0.00	tccaagacgaactcctcaa
RIGHT PRIMER	212	20	59.32	55.00	20.44	12.14	0.00	tatgccgtaccttcgatggg
INTERNAL OLIGO	85	20	60.03	50.00	0.00	0.00	0.00	acatacgccaacctctcgaa
PRODUCT SIZE: 199,	PAIR	ANY_TH	COMPL: 0.00,	PAIR	3'_TH	COMPL: 0.00		
8 LEFT PRIMER	88	20	59.04	50.00	0.00	0.00	0.00	tacgccaacctctcgaaact
RIGHT PRIMER	281	19	59.20	57.89	6.66	0.00	0.00	ggaaagtgtcgtccatgc
INTERNAL OLIGO	218	20	60.02	50.00	0.00	0.00	0.00	ttccaaccacccacactctt
PRODUCT SIZE: 194,	PAIR	ANY_TH	COMPL: 0.00,	PAIR	3'_TH	COMPL: 0.00		
9 LEFT PRIMER	62	19	59.19	57.89	0.00	0.00	0.00	ccgacggcagtatcaagga
RIGHT PRIMER	230	20	58.94	50.00	0.00	0.00	0.00	tgggtggttggaatgacgta
INTERNAL OLIGO	85	20	60.03	50.00	0.00	0.00	0.00	acatacgccaacctctcgaa
PRODUCT SIZE: 169,	PAIR	ANY_TH	COMPL: 0.00,	PAIR	3'_TH	COMPL: 0.00		
10 LEFT PRIMER	319	20	59.10	50.00	0.00	0.00	0.00	agcccgtccgagaaatacaa

	RIGHT PRIMER	513	21	59.15	52.38	0.00	0.00	0.00	catcgctcgctgtaatggtctg
	INTERNAL OLIGO	371	20	60.38	55.00	0.00	0.00	0.00	tcgccgaggagaaagaagac
	PRODUCT SIZE: 195,	PAIR ANY_TH	COMPL: 0.00,	PAIR 3'_TH	COMPL: 0.00				
11	LEFT PRIMER	211	20	58.94	50.00	0.00	0.00	0.00	tacgtcattccaaccaccca
	RIGHT PRIMER	387	20	60.46	55.00	11.27	0.00	0.00	ttctttctcctcggcgatgc
	INTERNAL OLIGO	319	20	60.03	50.00	0.00	0.00	0.00	agcccgtccgagaaatacaa
	PRODUCT SIZE: 177,	PAIR ANY_TH	COMPL: 0.00,	PAIR 3'_TH	COMPL: 0.00				
12	LEFT PRIMER	276	20	60.04	55.00	0.00	0.00	0.00	ctttcccgactgcctccatt
	RIGHT PRIMER	474	20	59.54	50.00	0.00	0.00	0.00	ttgctcgccgatacatctgt
	INTERNAL OLIGO	319	20	60.03	50.00	0.00	0.00	0.00	agcccgtccgagaaatacaa
	PRODUCT SIZE: 199,	PAIR ANY_TH	COMPL: 0.00,	PAIR 3'_TH	COMPL: 0.00				
13	LEFT PRIMER	80	19	59.02	52.63	0.00	0.00	0.00	agctcacatacgccaacct
	RIGHT PRIMER	240	20	58.40	55.00	0.00	0.00	0.00	cagaagagtgtgggtggttg
	INTERNAL OLIGO	165	20	60.03	50.00	13.59	0.00	38.28	cattctgcgcaaggtaagt
	PRODUCT SIZE: 161,	PAIR ANY_TH	COMPL: 0.00,	PAIR 3'_TH	COMPL: 0.00				
14	LEFT PRIMER	109	20	58.35	50.00	0.00	0.00	0.00	accctcctcaatcaagtcgt
	RIGHT PRIMER	295	20	60.04	55.00	0.00	0.00	0.00	aatggaggcagtcgggaaag
	INTERNAL OLIGO	218	20	60.02	50.00	0.00	0.00	0.00	ttccaaccacccacactctt
	PRODUCT SIZE: 187,	PAIR ANY_TH	COMPL: 1.96,	PAIR 3'_TH	COMPL: 0.00				
15	LEFT PRIMER	18	21	58.68	47.62	0.00	0.00	0.00	agacgaactcctccaagaaca
	RIGHT PRIMER	198	20	59.45	55.00	0.00	0.00	0.00	gatgggcatgggagacttga
	INTERNAL OLIGO	85	20	60.03	50.00	0.00	0.00	0.00	acatacgccaacctctcgaa
	PRODUCT SIZE: 181,	PAIR ANY_TH	COMPL: 0.00,	PAIR 3'_TH	COMPL: 0.00				
16	LEFT PRIMER	6	19	59.41	57.89	0.00	0.00	0.00	ccccgacatccaagacgaa
	RIGHT PRIMER	192	20	58.54	55.00	0.00	0.00	0.00	catgggagacttgaccttgc
	INTERNAL OLIGO	85	20	60.03	50.00	0.00	0.00	0.00	acatacgccaacctctcgaa
	PRODUCT SIZE: 187,	PAIR ANY_TH	COMPL: 0.00,	PAIR 3'_TH	COMPL: 0.00				
17	LEFT PRIMER	187	19	59.16	57.89	5.87	1.83	0.00	cccatgcccatacgaaggta
	RIGHT PRIMER	350	21	58.91	52.38	0.00	0.00	0.00	ggggacaggtgcttgatttc
	INTERNAL OLIGO	218	20	60.02	50.00	0.00	0.00	0.00	ttccaaccacccacactctt
	PRODUCT SIZE: 164,	PAIR ANY_TH	COMPL: 0.00,	PAIR 3'_TH	COMPL: 0.00				
18	LEFT PRIMER	85	20	59.11	50.00	0.00	0.00	0.00	acatacgccaacctctcgaa
	RIGHT PRIMER	247	18	59.34	61.11	2.24	0.00	0.00	gagcgccagaagagtgt
	INTERNAL OLIGO	165	20	60.03	50.00	13.59	0.00	38.28	cattctgcgcaaggtaagt
	PRODUCT SIZE: 163,	PAIR ANY_TH	COMPL: 0.00,	PAIR 3'_TH	COMPL: 0.00				
19	LEFT PRIMER	184	19	59.17	57.89	0.00	0.00	0.00	tctcccatgcccatacgaag
	RIGHT PRIMER	343	21	59.34	52.38	0.00	0.00	0.00	ggtgcttgattttctcgacg
	INTERNAL OLIGO	218	20	60.02	50.00	0.00	0.00	0.00	ttccaaccacccacactctt
	PRODUCT SIZE: 160,	PAIR ANY_TH	COMPL: 0.00,	PAIR 3'_TH	COMPL: 0.00				
20	LEFT PRIMER	322	20	58.00	50.00	0.00	0.00	0.00	ccgtccgagaaatacaagca
	RIGHT PRIMER	507	21	58.21	47.62	0.00	0.00	0.00	cgctgtaatggtctgcaattg
	INTERNAL OLIGO	371	20	60.38	55.00	0.00	0.00	0.00	tcgccgaggagaaagaagac

	PRODUCT SIZE:	186,	PAIR ANY_TH COMPL:	0.00,	PAIR 3'_TH COMPL:	0.00	
21	LEFT PRIMER	371	20	59.47	55.00	0.00	0.00
	RIGHT PRIMER	530	21	57.53	42.86	15.24	9.83
	INTERNAL OLIGO	455	20	60.45	50.00	17.70	3.41
	PRODUCT SIZE:	160,	PAIR ANY_TH COMPL:	0.00,	PAIR 3'_TH COMPL:	0.00	
22	LEFT PRIMER	330	21	58.91	52.38	0.00	0.00
	RIGHT PRIMER	517	20	57.00	50.00	0.00	0.00
	INTERNAL OLIGO	371	20	60.38	55.00	0.00	0.00
	PRODUCT SIZE:	188,	PAIR ANY_TH COMPL:	0.00,	PAIR 3'_TH COMPL:	0.00	
23	LEFT PRIMER	68	20	57.32	50.00	17.19	0.00
	RIGHT PRIMER	228	21	59.60	52.38	0.00	0.00
	INTERNAL OLIGO	165	20	60.03	50.00	13.59	0.00
	PRODUCT SIZE:	161,	PAIR ANY_TH COMPL:	0.00,	PAIR 3'_TH COMPL:	0.00	
24	LEFT PRIMER	158	19	59.63	52.63	13.05	0.00
	RIGHT PRIMER	346	21	58.30	47.62	0.00	0.00
	INTERNAL OLIGO	218	20	60.02	50.00	0.00	0.00
	PRODUCT SIZE:	189,	PAIR ANY_TH COMPL:	0.00,	PAIR 3'_TH COMPL:	0.00	
25	LEFT PRIMER	20	22	58.12	40.91	0.00	0.00
	RIGHT PRIMER	194	19	59.01	57.89	0.00	0.00
	INTERNAL OLIGO	85	20	60.03	50.00	0.00	0.00
	PRODUCT SIZE:	175,	PAIR ANY_TH COMPL:	0.00,	PAIR 3'_TH COMPL:	0.00	
26	LEFT PRIMER	263	19	59.20	57.89	0.00	0.00
	RIGHT PRIMER	461	18	59.74	61.11	0.00	0.00
	INTERNAL OLIGO	319	20	60.03	50.00	0.00	0.00
	PRODUCT SIZE:	199,	PAIR ANY_TH COMPL:	23.28,	PAIR 3'_TH COMPL:	24.73	
27	LEFT PRIMER	111	21	57.68	47.62	0.00	0.00
	RIGHT PRIMER	286	19	59.71	57.89	0.00	0.00
	INTERNAL OLIGO	218	20	60.02	50.00	0.00	0.00
	PRODUCT SIZE:	176,	PAIR ANY_TH COMPL:	0.00,	PAIR 3'_TH COMPL:	0.00	
28	LEFT PRIMER	293	18	59.17	55.56	0.00	0.00
	RIGHT PRIMER	477	20	57.14	50.00	0.00	0.00
	INTERNAL OLIGO	319	20	60.03	50.00	0.00	0.00
	PRODUCT SIZE:	185,	PAIR ANY_TH COMPL:	4.29,	PAIR 3'_TH COMPL:	0.00	
29	LEFT PRIMER	120	21	57.45	42.86	0.00	0.00
	RIGHT PRIMER	296	18	59.01	61.11	0.00	0.00
	INTERNAL OLIGO	218	20	60.02	50.00	0.00	0.00
	PRODUCT SIZE:	177,	PAIR ANY_TH COMPL:	0.00,	PAIR 3'_TH COMPL:	0.00	
30	LEFT PRIMER	4	18	60.51	66.67	0.00	0.00
	RIGHT PRIMER	187	19	58.84	57.89	11.31	1.78
	INTERNAL OLIGO	85	20	60.03	50.00	0.00	0.00
	PRODUCT SIZE:	184,	PAIR ANY_TH COMPL:	0.00,	PAIR 3'_TH COMPL:	0.00	

31	LEFT PRIMER	82	20	58.07	55.00	0.00	0.00	0.00	ctcacatacgccaacctctc
	RIGHT PRIMER	269	18	60.74	61.11	0.00	0.00	0.00	tccatgcggtcgtgtgtg
	INTERNAL OLIGO	218	20	60.02	50.00	0.00	0.00	0.00	ttccaaccacccacactctt
	PRODUCT SIZE: 188, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.70								
32	LEFT PRIMER	368	20	60.46	55.00	1.60	0.00	0.00	gcatcgccgaggagaaagaa
	RIGHT PRIMER	535	23	58.47	39.13	15.24	5.80	0.00	tgtaaaacttgaaatcggaacc
	INTERNAL OLIGO	455	20	60.45	50.00	17.70	3.41	0.00	acagatgtatcggcgagcaa
	PRODUCT SIZE: 168, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00								
33	LEFT PRIMER	268	19	59.71	57.89	0.00	0.00	0.00	gacgagcactttcccgact
	RIGHT PRIMER	455	18	60.28	61.11	0.00	0.00	0.00	tgtcgtcccgcaccaaag
	INTERNAL OLIGO	319	20	60.03	50.00	0.00	0.00	0.00	agcccgtccgagaaatacaa
	PRODUCT SIZE: 188, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00								
34	LEFT PRIMER	31	23	58.09	43.48	0.00	0.00	0.00	caagaacaaaaggatatgctcgg
	RIGHT PRIMER	205	19	59.16	57.89	20.44	0.54	0.00	taccttcgatgggcatggg
	INTERNAL OLIGO	85	20	60.03	50.00	0.00	0.00	0.00	acatacgccaacctctcgaa
	PRODUCT SIZE: 175, PAIR ANY_TH COMPL: 0.98, PAIR 3'_TH COMPL: 0.00								
35	LEFT PRIMER	27	23	57.75	43.48	0.00	0.00	0.00	cctccaagaacaaaaggatatgc
	RIGHT PRIMER	202	19	59.17	57.89	0.00	0.00	0.00	cttcgatgggcatgggaga
	INTERNAL OLIGO	85	20	60.03	50.00	0.00	0.00	0.00	acatacgccaacctctcgaa
	PRODUCT SIZE: 176, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00								
36	LEFT PRIMER	112	23	58.44	43.48	0.00	0.00	0.00	ctcctcaatcaagtcgtcaaaga
	RIGHT PRIMER	310	18	59.17	55.56	0.00	0.00	0.00	atcgatgcgggtcccaat
	INTERNAL OLIGO	218	20	60.02	50.00	0.00	0.00	0.00	ttccaaccacccacactctt
	PRODUCT SIZE: 199, PAIR ANY_TH COMPL: 0.00, PAIR 3'_TH COMPL: 0.00								

Statistics

	con sid ered	too many Ns	in tar get	in excl reg	not ok reg	bad GC% clamp	no GC clamp	tm too low	tm too high	high any_th compl	high 3'_th compl	high hair- pin	poly X	high end stab	ok
Left	1743	132	0	0	0	94	0	340	597	0	0	84	0	0	496
Right	1783	127	0	0	0	148	0	227	704	0	0	133	0	0	444
Intl	3726	198	0	0	0	3	0	383	2104	0	0	70	0	0	968

Pair Stats:
considered 28850, unacceptable product size 28808, primer in pair overlaps a primer in a better pair 53272, ok 42
libprimer3 release 2.4.0

(primer3_results.cgi release 4.1.0)