

Appendix A PCR primers and characterization of SNP loci

Primer number, Contig and SNP	Primer sequence (5'-3')	<i>T_m</i> (°C)	<i>T_a</i> (°C)	Amplicon size (bp)
5, T2-31299, C/T	FI: AAAGGCGGCGGGTCCCTT	61.9	62	T 211
	RI: GGGGTATGCGGGATATTCGCG	63.9		C 241
	FO: CCCC GTTGTACGACGAGAATCGTA	63.7		Outer 413
	RO: GGTGAAGTCTCTCCTCCGGCCTTT	65.4		
9, T3-30695, G/A	FI: TCAACAAGTACAATAGGAAACCG	56.6	54	G 197
	RI: AACAGGATCCTGGGTATAGCAT	58.2		A 293
	FO: ATGTGACTCAGCCTCGTTATGTA	58.4		Outer 445
	RO: CTGGTAAAGAGACTGACAAGCTTAA	58.7		
11, T2-32906, C/G	FI: GTCGGGGAAATCAAAGAAATGC	58.0	61	C 215
	RI: CAGACTCCA ACTGCGATTGAGTAC	62.0		G 252
	FO: CTGATGGTGATGAGTCGAGGATAG	62.0		Outer 422
	RO: ATCAGACGTGTCCTCAGTACCCTT	62.0		
15, T1-26224, C/T	FI: ACTACAAGGTCAAAAACTAGTCTCGTC	60.6	56	C 171
	RI: AGAAGAAAAGCAGAAAGCCGA	56.1		T 248
	FO: TTA ACTATGATCTCCCTACTCAACCC	60.4		Outer 370
	RO: CAGTTACAAACCTCAAATGCAAAAT	55.4		
21, T2-26968, G/A	FI: ACCGCTCATGAACCTGCTGAAG	63.7	63	G 180
	RI: GAACCCGA ACTGGGTGTGGGTT	63.8		A 267
	FO: GCGTTTGAGCTTTCAGAGCGGATT	62.0		Outer 402
	RO: GGTGTACCCACCGACATTCTGATC	65.3		
25, T3-31578, T/C	FI: GACTCTTTTGTGTCATCCTCTCCT	60.3	60	T 151
	RI: CACTTGCTGCAAGTTTATCACG	58.2		C 178
	FO: CTCAATTGGGGATGTAATACTTGAG	58.7		Outer 283
	RO: AGAACTGGAGTTAGGTGAGAGAGGT	62.0		
26, T2-30620, T/C	FI: CAGGACCCCGATGATAAGGTT	60.0	57	T 210
	RI: TGCTTGATATTTCACTGCAAACG	57.1		C 261
	FO: ATCCATATGGAAGCCAAAACCTAGT	58.7		Outer 425
	RO: AACAGTCGGATCTATTCAACTCGAC	60.3		
27, T3-35667, C/T	FI: ACCGGTCATGAATGATTGTGT	56.1	60	T 213
	RI: GTGGTTCTTCTTAGTCTCCAGGG	62.0		C 249
	FO: ACTTTGTTTCATCAAATCTGTGCTCT	57.1		Outer 418
	RO: GGTACTATAAAGGGGAGGAGGACA	62.0		
30, T3-29141, C/T	FI: TGGA ACTCAGACTCCAAATGTC	58.2	57	C 208
	RI: GAACCAAGCCTTGAAAGGACA	58.0		T 242
	FO: GGAAGCCCAGGTAGTCGTTTAATAT	60.3		Outer 407
	RO: ATATCTTCCCAGTCAATTCAAATG	57.1		

Continued

Primer number, Contig and SNP	Primer sequence (5'-3')	T _m (°C)	T _a (°C)	Amplicon size (bp)
34, T1-26603, T/C	FI: CAGACAGTTGAAATAACAATGCACCTC	60.5	59	C 226
	RI: TTGTTTCAACCCGAGGAACGA	58.0		T 279
	FO: GTTGTTAATGATTCCATAATCGCGG	58.7		Outer 457
	RO: TGGAAATGTTGAACGTGATGAGTCT	58.7		
38, T2-28199, G/A	FI: AACTACTACCCAAAATGCCCTCAGACG	63.5	60	G 215
	RI: TCCACGCCGAGAGCTAGCGCT	65.8		A 164
	FO: CTGAGGGCATTAAACCACAACGGTGT	63.6		Outer 331
	RO: TGCTCTTGTACTIONGCCGTTGCTGAG	63.6		
41, T3-32657, T/C	FI: CCACTGTACGAGAGGGACAACGTC	65.4	57	C 190
	RI: TTCCATTCCCCTGCAGGATTTT	58.6		T 272
	FO: TCGGAAGAGACCAAGCTATCTGAGC	63.6		Outer 414
	RO: CAAGTGCAGATATGGATGATGAGC	62.0		
49, T3-23002, C/T	FI: ATGTCTTCTTGGTCCTGGCAT	58.0	55	T 167
	RI: GGAGAACAACCTACCAGAATATGGAGTAG	62.0		C 117
	FO: ATAGATTTTGGGACTGTCACATGAT	57.1		Outer 235
	RO: GATTCTTGAAAAATGGGAGGTATTA	57.1		
55, T3-30010, G/A	FI: GTGCAGGAGGTGAAAAACGCA	60.0	58	A 186
	RI: GTTCAACAAAATCCCCTTCTGGAAAC	60.4		G 213
	FO: TACCAAATGTTGAAGCTAGCAGCCA	60.3		Outer 352
	RO: CCCGCCAAAAGAAATCTTGAACCTAG	60.3		
62, T1-25257, G/A	FI: AAAAATCAGTAGTGATAATAATTTGTCACG	56.6	59	G 215
	RI: GCAGCATACGAGGACAACCAT	60.0		A 284
	FO: GATATAATACTGCAAAAGCACCAAGC	58.8		Outer 448
	RO: ATTGGAGTTCAAGATCCAGTAATGC	58.7		
67, T1-29128, G/C	FI: GCAATCACACCCGCACTAGAG	61.9	54	G 197
	RI: GCTTTAATTTAGGGGTATGCGG	60.2		C 261
	FO: GGGTCCTCACTACATGAACAAGCTT	62.0		Outer 414
	RO: TCATGTTTGGTGCATTGGATAATTT	55.4		
89, T2-31924, T/A	FI: GCTAAAATTATCGGAAGCATAAGCT	57.1	55	T 198
	RI: AAATAATGATTCTCTCATCACTGTTCT	57.6		A 236
	FO: AATCTTCAGGTCAAACACCTTGTA	57.1		Outer 381
	RO: TTATTGAAATAGTTGCTGCATGTGA	55.4		
93, T3-35889, T/C	FI: AAGGTACCAAAAAAAAAATCTGTGTC	55.4	56	C 188
	RI: AAATCTTCAATCACAGGCAAAA	52.6		T 222
	FO: TTCATTGGACCTAATTAAGAGTTCA	55.4		Outer 363
	RO: AAGTACATGATGTCACCTAAGCATGT	57.3		

Continued

Primer number, Contig and SNP	Primer sequence (5'-3')	T _m (°C)	T _a (°C)	Amplicon size (bp)
101, T2-30832, A/G	FI: ACGGGCCTTCCCACTCCCCCA	67.8	64	A 200
	RI: TTTACCAATCCTTGCATGATGGTTGAGTCC	63.4		G 289
	FO: CGTGACCTTTATCGTTGCCCCGAATC	65.1		Outer 438
	RO: CCCCCAAGGATCCTCTGGTGAGGAA	66.9		
110, T3-34224, T/C	FI: CCATCAAGCCCAAGAACACTAC	60.1	54	C 220
	RI: TTATCTTCATCTACATCTTCATCTGCA	57.4		T 298
	FO: AGGAGAAATGGGAGAATATAACAAGT	57.4		Outer 469
	RO: TGCAAAACAGCTTCTTAACACCTAT	57.1		
112, T1-26622, G/T	FI: AGTTACCTTGACTTCATCAAGGACTATATG	60.7	58	G 161
	RI: CTTCCAAGGCCAATCCCCGCA	65.7		T 207
	FO: GAATGGTTGAAGCAGCCTGTTTTCA	60.3		Outer 316
	RO: TTTCGTTGCGAAAAGTGTAAGGGTG	60.3		
113, T1-28028, G/C	FI: GAACCCCATCGTCGGATTCGTC	63.8	57	C 234
	RI: CCAGCAAGGGTAAATTGTTCTGTCCC	63.6		G 188
	FO: CTTGAGAAACGTGCACAACGCGTTAT	62.0		Outer 374
	RO: CGCTCTCGGAGACAACAATGGGTAT	63.6		
116, T2-24666, T/G	FI: ACCACCTCGCATCTTACGCCTT	61.9	64	T 176
	RI: CAATTCAGTAATCAAAGGTGTGGACCC	62.0		G 219
	FO: GTGCTTCCGCTTAGACATGTGCTTT	62.0		Outer 346
	RO: ACCGTCTGAAACTGTTACAGCATGAA	60.3		
123, T1-28812, T/A	FI: TTTTTTTTGTTCAGTGAGAGTCCCT	57.1	58	T 206
	RI: CCATTGCAGTGAACCTTACATACTCT	60.4		A 287
	FO: GAACAAACCGGAAAATAAAATCAAA	53.8		Outer 442
	RO: GATCATGCATAACAAAAACAACAGC	57.1		
125, T2-26567, A/C	FI: AACGGATTATTTGCTCACATGCC	60.3	64	C 202
	RI: GCTCGGACCTGCTCCTTCGTT	63.9		A 274
	FO: TGCATGAATTCGGGGAGATGATAAA	58.7		Outer 431
	RO: ATGGTACTCCAGACCCTTCCACTGG	65.3		
126, T2-33341, A/G	FI: ACTGAACGAAATAATGAGAATATGCCATA	57.8	60	A 236
	RI: TGGGAGTGACAGAGGGGCTAC	63.9		G 291
	FO: CTGCTCACACTCCAAGAAAATGAAC	60.3		Outer 477
	RO: AGATCCAAAAGGCATCATCTTGAAC	58.7		
128, T3-28938, A/C	FI: AATAAAACATATTATGCTTCCTCCTTTTA	55.0	54	A 213
	RI: AACAACAAATATATCTCTAAATTCTCTTCG	5.6		C 279
	FO: ACTGAATTCTTTACCGAGTATGGAG	58.7		Outer 433
	RO: TATTTTTTGGTTTTAAATCTCGGTG	53.8		

Continued

Primer number, Contig and SNP	Primer sequence (5'-3')	T _m (°C)	T _a (°C)	Amplicon size (bp)
131, T1-22617, G/A	FI: CTTAATATCCCAGGGTGATGTGTAG	60.4	60	G 171
	RI: TACGAGTCGAGCTTCTTCGCT	60.0		A 254
	FO: ATATACATAACTGAGAGGAGCCCC	62.0		Outer 378
	RO: ACCATTGATATCCCCTACTTCCTTC	60.3		
140, T3-32052, C/G	FI: AATGAACACAGCCATGTCAAAGTATC	58.8	60	C 167
	RI: CACATCAGAGTAAAAAAAAACACGAATC	57.6		G 227
	FO: CGGATTCATGAAAGTGTATGCAG	58.6		Outer 340
	RO: AATAATCTATAACGCCCGTTCAAA	57.1		
147, T1-27404, A/G	FI: TTCGTATTTTATTAGACATTGTTAG	52.1	53	G 183
	RI: CGGATAGTGAGTTCTCTAGGT	58.0		A 271
	FO: TAAGAGAGAAGCTAGCAGATTC	56.3		Outer 408
	RO: CTTGAGTACTCTGCTGAATAGAT	56.6		
148, T1-27856, G/C	FI: AGGTTCTTGCACAGCTTGTC	58.0	56	C 199
	RI: TTCGGAGAGGAGTGAACAGTAAC	60.2		G 278
	FO: CATTATCATCGTCTTCTTCATCATC	57.1		Outer433
	RO: GAAGTAATTGTTCCAGCTCTAAGGA	58.7		
152, T3-23789, T/C	FI: GTTGTTGTGATGGGGAGGAC	61.9	57	C 211
	RI: CAACAGTTCTCTATTGAATCATAAACTGCA	59.3		T 266
	FO: CCTGCTCTCCAACCAAGTTAATAG	62.0		Outer 426
	RO: GATTATGAACTGCTTCCAGACTCCC	62.0		