Reaction Conditions - GT271

				Cocktail Volume (uL)	1130	Use PCI	R Cycles F	PROPDNR	
Reagent	[]	[] Units	Desired []	Des'd [] Units	Amount to add	5 min	@	94	
Buffer	10	X	1	X	113	30 sec	@	94	
dNTP's	2	mM Each	0.2	mM Each	113	1 min	@	58	35x
MgCl2	50	mM	1.5	mM	33.9	1 min	@	72	
RW34-F-6FAM	20	uM	0.5	uM	28.25	45 min	@	60	
RW34-R	20	uM	0.5	uM	28.25				1
GT023-F-HEX		uM		uM	16.95	Is a 10u	I reaction	volume	
GT023-R		uM	0.3	uM	16.95				
GT271-F-NED		uM	0.1	uM	5.65				
GT271-R	20	uM	0.1	uM	5.65				
TR2G5-F-NED	10		0.05		5.65				
TR2G5-R		uM	0.05	uM	5.65				
IRZG5-R				U/uL	11.3				
Taq	5	U/uL	0.05	U/UL					
		ng/uL		ng/rxn (2 uL/rxn)	226				
Taq					226 519.8				
Taq DNA	5		10						
Taq DNA Autoclaved ddH2O	5		10		519.8	Use PCI	R Cycles F	PROPDNR	
Taq DNA Autoclaved ddH2O Don't Dilute Prior to	5		10	ng/rxn (2 uL/rxn)	519.8 1130	Use PCI	R Cycles F	PROPDNR 94	
Taq DNA Autoclaved ddH2O Don't Dilute Prior to RXN #2 Reagent Buffer	Mixing	ng/uL	10 to 1130 uL	ng/rxn (2 uL/rxn) Cocktail Volume (uL) Des'd [] Units X	519.8 1130 1130 Amount to add	5 min 30 sec	@	94 94	
Taq DNA Autoclaved ddH2O Don't Dilute Prior to RXN #2 Reagent Buffer dNTP's	Mixing [] 10 2	ng/uL [] Units X mM Each	10 to 1130 uL Desired [] 1 0.2	ng/rxn (2 uL/rxn) Cocktail Volume (uL) Des'd [] Units X mM Each	519.8 1130 1130 Amount to add 113 113	5 min 30 sec 1 min	@	94 94	30 X
Taq DNA Autoclaved ddH2O Don't Dilute Prior to RXN #2 Reagent Buffer dhTP's MgCl2	Mixing [] 10 2 50	ng/uL [] Units X mM Each	Desired [] 10 10 1130 uL	ng/rxn (2 uL/rxn) Cocktail Volume (uL) Des'd [] Units X mM Each mM	519.8 1130 Amount to add 113 113 33.9	5 min 30 sec	@	94 94	30 X
Taq DNA Autoclaved ddH2O Don't Dilute Prior to RXN #2 Reagent Buffer dNTP's MgCl2 TR3G6-F-HEX	Mixing [] 10 2 50 20	I J Units X MM Each mM	Desired [] 10 10 130 uL	ng/rxn (2 uL/rxn) Cocktail Volume (uL) Des'd [] Units X mM Each mM	519.8 1130 1130 Amount to add 113 113 33.9 4.52	5 min 30 sec 1 min	@	94 94 54	30 X
Taq DNA Autoclaved ddH2O Don't Dilute Prior to RXN #2 Reagent Buffer dNTP's MgCl2 TR3G6-F-HEX TR3G6-F-R	Mixing [] 10 2 50 20 20 20	[] Units X mM Each mM uM	Desired [] 0.2 1.5 0.08 0.08	ng/rxn (2 uL/rxn) Cocktail Volume (uL) Des'd [] Units X mM Each mM	1130 Amount to add 113 113 33.9 4.52 4.52	5 min 30 sec 1 min 1 min 45 min	@ @ @	94 94 54 72 60	30 X
Taq DNA Autoclaved ddH2O Don't Dilute Prior to RXN #2 Reagent Buffer Buffer Buffer MTP's MgCl2 TR3G6-F-HEX TR3G6-R EV37Mn-F-HEX	Mixing [] 10 2 50 20 20 20	[] Units X mM Each mM uM uM	Desired [] 10 10 10 1130 uL	ng/rxn (2 uL/rxn) Cocktail Volume (uL) Des'd [] Units X mM Each mM uM	519.8 1130 Amount to add 113 113 33.9 4.52 4.52 11.3	5 min 30 sec 1 min 1 min 45 min	@ @ @	94 94 54 72 60	30 X
Taq DNA Autoclaved ddH2O Don't Dilute Prior to RXN #2 Reagent Buffer dNTP's MgCl2 TR3G6-F-HEX TR3G6-F-R	Mixing [] 10 2 50 20 20 20 20	ng/uL [] Units X mM Each mM uM uM uM uM	Desired [] 10 10 130 uL 10 10 10 10 10 10 10 10 10 10 10 10 10	ng/rxn (2 uL/rxn) Cocktail Volume (uL) Des'd [] Units X mM Each mM uM uM uM	519.8 1130 Amount to add 113 313 4.52 4.52 11.3 11.3	5 min 30 sec 1 min 1 min 45 min	@ @ @	94 94 54 72 60	30 X
Taq DNA Autoclaved ddH2O Don't Dilute Prior to RXN #2 Reagent Buffer dMTP's MgCl2 TR3G6-F-HEX TR3G6-R EV37Mn-F-HEX EV37Mn-R Taq	Mixing [] 10 2 2 50 20 20 20 50	Ing/uL [] Units X Indicate the second of	Desired [] 0.2 1.5 0.08 0.08 0.2 0.2 0.05	ng/rxn (2 uL/rxn) Cocktail Volume (uL) Des'd [] Units X mM Each mM uM uM uM UM U/UL	1130 Amount to add 113 33.9 4.52 4.52 11.3 11.3 11.3	5 min 30 sec 1 min 1 min 45 min	@ @ @	94 94 54 72 60	30 X
Taq DNA Autoclaved ddH2O Don't Dilute Prior to RXN #2 Reagent Buffer ddffr/S MgCi2 TR3G6-F-HEX TR3G6-R EV37Mn-F-HEX EV37Mn-R	Mixing [] 10 2 2 50 20 20 20 50	ng/uL [] Units X mM Each mM uM uM uM uM	Desired [] 0.2 1.5 0.08 0.08 0.2 0.2 0.05	ng/rxn (2 uL/rxn) Cocktail Volume (uL) Des'd [] Units X mM Each mM uM uM uM	519.8 1130 Amount to add 113 313 4.52 4.52 11.3 11.3	5 min 30 sec 1 min 1 min 45 min	@ @ @	94 94 54 72 60	30 X
Taq DNA Autoclaved ddH2O Don't Dilute Prior to RXN #2 Reagent Buffer dMTP's MgCl2 TR3G6-F-HEX TR3G6-R EV37Mn-F-HEX EV37Mn-R Taq	Mixing [] 10 2 2 50 20 20 20 50	Ing/uL [] Units X Indicate the second of	Desired [] 0.2 1.5 0.08 0.08 0.2 0.2 0.05	ng/rxn (2 uL/rxn) Cocktail Volume (uL) Des'd [] Units X mM Each mM uM uM uM UM U/UL	1130 Amount to add 113 33.9 4.52 4.52 11.3 11.3 11.3	5 min 30 sec 1 min 1 min 45 min	@ @ @	94 94 54 72 60	30 X

GT271 Scoring Criteria

- Allele bins are even
- Score the +A
- Locus is monomorphic for 102 bp, so if you don't get that then your ladder is screwed up.
- Sometimes the allele (rather than the +A) is preferentially amplified

