Reaction Conditions - TR3F2

RXN#1			5-100 may 1	Cocktail Volume (uL)	1130	PCR Cycles			
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	30 sec	@	68	5X
MgCl2	50	mM	1.5	mM	33.9	1 min	@	72	
TR3G1-6FAM	20	uM	0.255	uM	14.4075	30 sec	@	94	
TR3G1-R	20	uM	0.255	uM	14.4075	30 sec	@	64	5X
Taq	5	U/uL	0.05	U/uL	11.3	1 min	@	72	
DNA	5	ng/uL	10	ng/rxn (2 uL/rxn)	226	30 sec	@	94	
Autoclaved ddH2O	80 30	Š.	to 1130 uL		603.985	30 sec	@	61	5X
	F 10		¥3		1130	1 min	· @	72	
	9		P I		- N	30 sec	@	94	
	3 8					30 sec	@		5X
						1 min	@	72	
						30 sec	@	94	
Don't dilute prior to	de-saltin	a				30 sec	@	55	10X
			i i		7	1 min	@	72	
						10 min	<u> </u>	72	
			1		× ×				
	(i)					Is a 10ul rea	ction volume		
RXN#2				Cocktail Volume (uL)	1130				
Reagent	[]	[] Units	Desired []	Des'd [] Units	Amount to add				
Buffer	10	X	1	X	113	Use PCR Cy	cles PROPDNR		
	2	mM Each	0.2	mM Each	113		<u></u>	9.4	
dNTP's	2	mivi Each	0.2	mivi Each	113	5 min	(W	04	
MgCl2		mM Each		mM	33.9	30 sec	@	94	
MgCl2		mM					@ @		30 X
MgCl2 Q-Solution	50 5	mM		mM X	33.9	30 sec	@		30 X
MgCl2 Q-Solution TR3F2-F-HEX	50 5 40	mM X	1.5	mM X uM	33.9 226	30 sec 1 min	@	58	30 X
MgCl2 Q-Solution TR3F2-F-HEX TR3F2-R	50 5 40 40	mM X uM	1.5 1 0.255 0.255	mM X uM	33.9 226 7.20375	30 sec 1 min 1 min	@ @	58	30 X
MgCl2 Q-Solution TR3F2-F-HEX TR3F2-R Taq	50 5 40 40 5	mM X uM uM U/uL	1.5 1 0.255 0.255 0.05	mM X uM uM U/uL	33.9 226 7.20375 7.20375	30 sec 1 min 1 min 45 min	@ @	58	30 X
MgCl2 Q-Solution TR3F2-F-HEX TR3F2-R Taq DNA	50 5 40 40 5	mM X uM uM	1.5 1 0.255 0.255 0.05	mM X uM uM	33.9 226 7.20375 7.20375 11.3	30 sec 1 min 1 min 45 min	@ @	58	30 X
dNTP's MgCl2 Q-Solution TR3F2-F-HEX TR3F2-R Taq DNA Autoclaved ddH2O	50 5 40 40 5	mM X uM uM U/uL	1.5 1 0.255 0.255 0.05 10	mM X uM uM U/uL	33.9 226 7.20375 7.20375 11.3 226	30 sec 1 min 1 min 45 min	@ @	58	30 X
MgCl2 Q-Solution TR3F2-F-HEX TR3F2-R Taq DNA	50 5 40 40 5	mM X uM uM U/uL	1.5 1 0.255 0.255 0.05 10	mM X uM uM U/uL	33.9 226 7.20375 7.20375 11.3 226 392.3925	30 sec 1 min 1 min 45 min	@ @	58	30 X
MgCl2 Q-Solution TR3F2-F-HEX TR3F2-R Taq DNA	50 5 40 40 5 5	mM X uM uM U/uL ng/uL	1.5 1 0.255 0.255 0.05 10	mM X uM uM U/uL	33.9 226 7.20375 7.20375 11.3 226 392.3925	30 sec 1 min 1 min 45 min	@ @	58	30 X

TR3F2 Scoring Criteria

- Allele bins are even
- Score the +A, although you don't really see the allele

