

Reaction Conditions - SAM25

RXN#1			Cocktail Volume (uL)		1130
Reagent	[]	[] Units	Desired []	Des'd [] Units	Amount to add
Buffer	10	X	1	X	113
dNTP's	2	mM Each	0.2	mM Each	113
MgCl2	50	mM	1.5	mM	33.9
SAM25-F-HEX	40	uM	1	uM	28.25
SAM25-R	40	uM	1	uM	28.25
RW31-F-HEX	40	uM	2.5	uM	70.625
RW31-R	40	uM	2.5	uM	70.625
EV1Pm-F-NED	40	uM	0.3	uM	8.475
EV1Pm-R	40	uM	0.3	uM	8.475
Taq	5	U/uL	0.05	U/uL	11.3
DNA	5	ng/uL	10	ng/rxn (2 uL/rxn)	226
Autoclaved ddH2O			to 1130 uL		418.1
					1130
Don't Dilute Prior To De-salting					
RXN#2			Cocktail Volume (uL)		1130
Reagent	[]	[] Units	Desired []	Des'd [] Units	Amount to add
Buffer	10	X	1	X	113
dNTP's	2	mM Each	0.2	mM Each	113
MgCl2	50	mM	1.5	mM	33.9
TR3A1-F-6FAM	40	uM	0.45	uM	12.7125
TR3A1-R	40	uM	0.45	uM	12.7125
TR2F3-F-NED	20	uM	0.3	uM	16.95
TR2F3-R	20	uM	0.3	uM	16.95
Taq	5	U/uL	0.05	U/uL	11.3
DNA	5	ng/uL	10	ng/rxn (2 uL/rxn)	226
Autoclaved ddH2O			to 1130 uL		573.475
					1130
Don't Dilute Prior To De-salting					

Use PCR Cycles PROPDNR		
5 min	@	94
30 sec	@	94
1 min	@	54.5
1 min	@	72
45 min	@	60

Is a 10ul reaction volume

Use PCR Cycles PROPDNR		
5 min	@	94
30 sec	@	94
1 min	@	54
1 min	@	72
45 min	@	60

Is a 10ul reaction volume

SAM25 Scoring Criteria

- Score the allele, although the +A is still there
- Allele bins are even

