

Reaction Conditions - IGF

RXN#1				Cocktail Volume (uL)	1130	Use PCR Cycles PROPDNR	
Reagent	[]	[] Units	Desired []	Des'd [] Units	Amount to add		
Buffer	10	X	1	X	113	5 min @	94
dNTP's	2	mM Each	0.2	mM Each	113	30 sec @	94
MgCl2	50	mM	1.5	mM	33.9	1 min @	52 30 X
GATA028-F-NED	40	uM	1	uM	28.25	1 min @	72
GATA028-R	40	uM	1	uM	28.25	45 min @	60
TR3G11-F-6FAM	20	uM	0.52	uM	29.38	Is a 10ul reaction volume	
TR3G11-R	20	uM	0.52	uM	29.38		
TV14-F-NED	20	uM	0.1	uM	5.65		
TV14-R	20	uM	0.1	uM	5.65		
IGF-F-HEX	40	uM	0.675	uM	19.06875		
IGF-R	40	uM	0.675	uM	19.06875		
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		468.1025		
					1130		
Don't Dilute Prior To De-salting							
RXN#2				Cocktail Volume (uL)	1130	Use PCR Cycles PROPDNR	
Reagent	[]	[] Units	Desired []	Des'd [] Units	Amount to add		
Buffer	10	X	1	X	113	5 min @	94
dNTP's	2	mM Each	0.2	mM Each	113	30 sec @	94
MgCl2	50	mM	1.5	mM	33.9	1 min @	60 30 X
TR3G13-F-NED	20	uM	0.4	uM	22.6	1 min @	72
TR3G13-R	20	uM	0.4	uM	22.6	45 min @	60
TR3G10-F-6FAM	40	uM	0.76	uM	21.47	Is a 10ul reaction volume	
TR3G10-R	40	uM	0.76	uM	21.47		
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		544.66		
					1130		
Don't Dilute Prior To De-salting							

IGF Scoring Criteria

- Score the +A
- Allele bins are even
- Beware of pull-up from GATA028 in size range!!

