

27/04/2016

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Additive: DMSO		0	%			
30 uL	1X_65ul 1.5ul Mg+	1ul	x	Master Mix (no primers)		2 x
Amount	Compound			Amount	Compound	
20.7 uL	ddH ₂ O	0.69		41.4 uL	ddH ₂ O	41.4
0 uL	DMSO 100%	0		0 uL	DMSO 100%	0
3 uL	10x PCR buffer	0.1		6 uL	10x PCR buffer	6
2.4 uL	dNTPs (2.5mM each)	0.08		4.8 uL	dNTPs (2.5mM each)	4.8
1.8 uL	MgCl ₂	0.06		3.6 uL	MgCl ₂	3.6
0.6 uL	Pf, 5µM	0.02		1.2 uL	Pf, 5µM	55.8
0.6 uL	Pr, 5 µM	0.02		1.2 uL	Pr, 5 µM	
0.6 uL	DNA template	0.02		1.2 uL	DNA template	
0.3 uL	Polymerase	0.01		0.6 uL	Polymerase	
30 uL	Total	1		60 uL	Total mm per25 ul:	27.9 30

original protocol with 5x enhancer

Notes:		Genomic DNA BL2 tested in 16S PCR reaction, 2 types with and with no DMSO			
		94 C - 5 min			
		94 C, 45 s: 48 C, 1 min: 72 C, 2 min: 33x			
		72 C, 5 min			
		4 C, inf			
primers done from stock: 9f 400, 1492r 200: by dilution to 50					
ul	stock: uM	h2o ul	resulting uM	ul resulting	
9f	20	400	140 50	160	
1492r	40	200	120 50	160	

Prepare Mmix Taq		0	%			
500 uL	1X_65ul 1.5ul Mg+	1ul	x	Master Mix (no primers)		1 x
Amount	Compound			Amount	Compound	
345 uL	ddH ₂ O	0.69		345 uL	ddH ₂ O	345
0 uL	DMSO 100%	0		0 uL	DMSO 100%	0
50 uL	10x PCR buffer	0.1		50 uL	10x PCR buffer	50
40 uL	dNTPs (2.5mM each)	0.08		40 uL	dNTPs (2.5mM each)	40
30 uL	MgCl ₂	0.06		30 uL	MgCl ₂	30
10 uL	Pf, 5µM	0.02		10 uL	Pf, 5µM	465
10 uL	Pr, 5 µM	0.02		10 uL	Pr, 5 µM	
10 uL	DNA template	0.02		10 uL	DNA template	
5 uL	Polymerase	0.01		5 uL	Polymerase	
500 uL	Total	1		500 uL	Total mm per25 ul:	465 500