

Pu Promoter Primers from PU for Real

Forward Primer

5' GGTGCTGCAGACTAGTATTGAAGGGTCACCACTATTTTTATTTTA 3'

Reverse Primer

5' ATGAATTCTCTAGACCTGCTGGAGGGCGTGAAC

Amplification Summary

Upper Primer:	45-mer	5' GGTGCTGCAGACTAGTATTGAAGGGTCACCACTATTTTTATTTTA	3'
Lower Primer:	33-mer	5' ATGAATTCTCTAGACCTGCTGGAGGGCGTGAAC	3'
DNA 250 pM, Salt 50 mM		Upper Primer	Lower Primer
Primer Tm		69.9 °C	69.5 °C
Primer Overall Stability		-79.1 kc/m	-62.5 kc/m
Primer Location		1..45	370..338
Product Tm - Primer Tm		7.1 °C	
Primers Tm Difference		0.4 °C	
Optimal Annealing Temperature		59.5 °C	
Product Length		370 bp	
Product Tm (%GC Method)		76.6 °C	
Product GC Content		45.1%	
Product Tm at 6xSSC		98.2 °C	

Product Melting Temperature (%GC Method)

Salt			Formamide			
mM	xSSC	xSSPE	0%	10%	20%	50%
1	0.005	0.006	48.4	41.9	35.4	15.9
10	0.051	0.062	65.0	58.5	52.0	32.5
50	0.256	0.312	76.6	70.1	63.6	44.1
165	0.846	1.031	85.2	78.7	72.2	52.7
330	1.692	2.062	90.2	83.7	77.2	57.7
500	2.564	3.125	93.2	86.7	80.2	60.7
1000	5.128	6.250	98.2	91.7	85.2	65.7
195	1.000	1.219	+ 0.0	%formamide = Tm 86.4 °C		

Tm of Pu promoter analogous regions only

Forward primer: 53 C

Reverse: 58 C

From NCBI P. putida pWW0 complete sequence

75381-75720

Note: primer pair dimer -4.9 dG