The 16s/23s ribosomal spacer region as a target for DNA probes to identify eubacteria

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Barany, F. 1991. The ligase chain reaction in a PCR world. *PCR Methods Applic.* **1**:5–16.

Table 1 of the above titled paper inadvertently listed an incorrect value for NAD in Method 1; the correct value is 1 mM, not 10 mM as stated. The correct version of Method 1 is reproduced on this page.

TABLE 1 Ligase Chain Reaction Methods

	Method 1[a]
Target DNA	β^A,β^S
Standard detection	1–10 attomoles (6×10^5 to 6×10^6 molecules)
Signal-to-noise no target under standard conditions	1700 to >2000[b]
single-base mismatch under standard conditions	75 to >500[b]
Lowest detection	200 molecules
Position of discriminating nucleotide	3′ base of both strands (single-base 3′ overhang)
T_m discrimination oligonucleotides	64°C–68°C (23- to 28-mers)
T_m adjacent oligonucleotides	70°C (22-mers)
Amount of each oligonucleotide	40 femtomoles (0.28 ng)
Volume	10 μl
Buffer conditions	20 mM Tris-HCl, pH 7.6[c] 100 mM or 150 mM KCl 10 mM $MgCl_2$ 10 mM DTT 1 mM NAD^+ 1 mM EDTA
Carrier DNA to suppress background	4 μg of salmon sperm DNA
Additional features for suppression of target independent background	5′ phosphate on adjacent oligonucleotides only; noncomplementary tails on outside of oligonucleotides; single-base 3′ overhang on discriminating oligonucleotides
Thermostable enzyme	15 nick-closing units[d]
Cycle conditions	94°C, 1 min 65°C, 4 min 20 or 30 cycles or 94°C, 0.5 min 65°C, 2 min 30 or 40 cycles

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Barry, T., G. Colleran, M. Glennon, L.K. Dunican, and F. Gannon. 1991. The 16s/23s ribosomal spacer region as a target for DNA probes to identify eubacteria. *PCR Methods Applic.* **1**:51–62.

Figure 4 has errors in the A1 and B1 primers. The correct primers are:
A1 5′-AGTCGTAACAAGGTAGCCG-3′
B1 5′-C T/C A/G T/C TGCCAAGGCAT CCACC-3′