

Supporting Information

Hobbie et al. 10.1073/pnas.0811258106

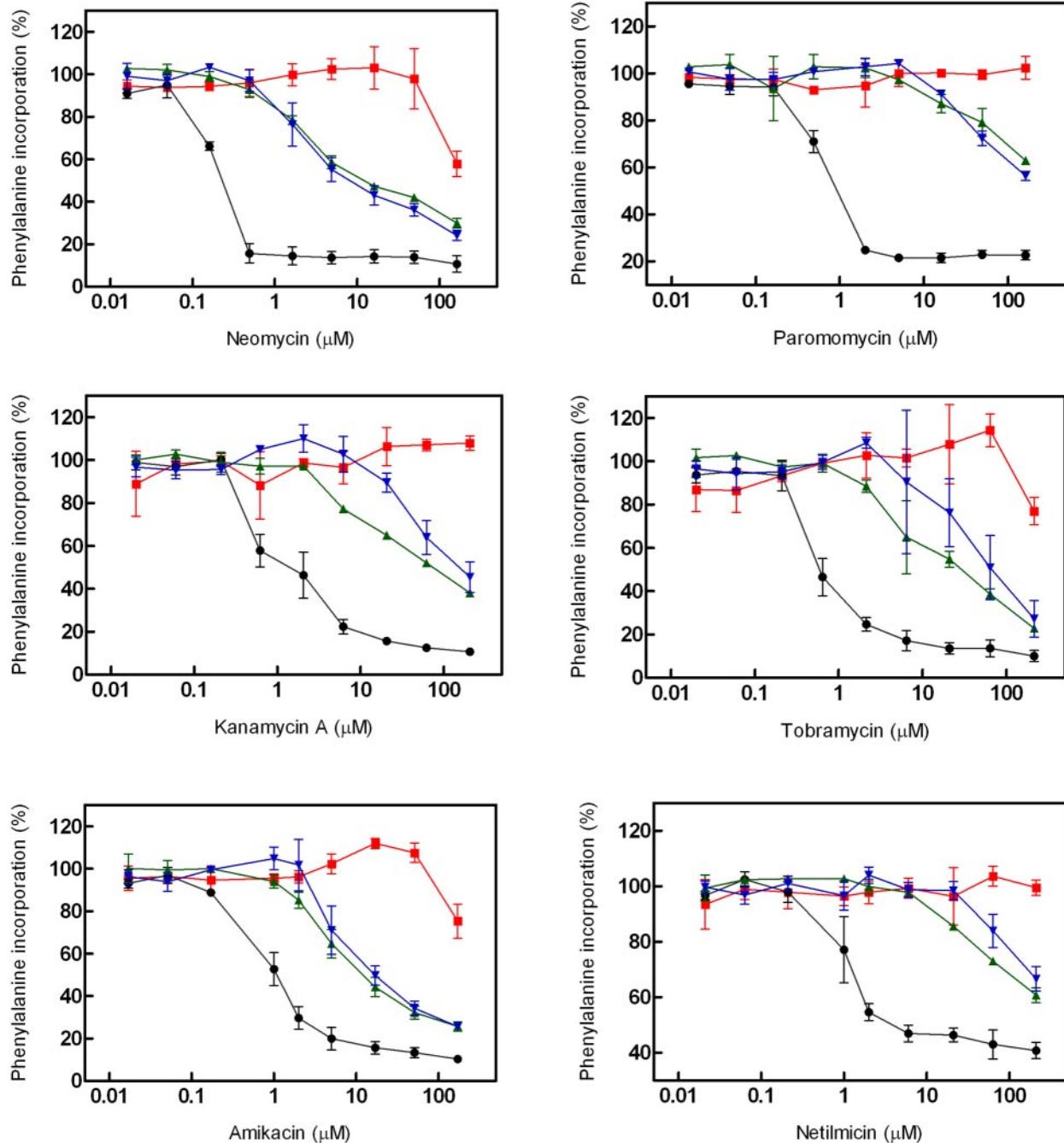


Fig. S1. Aminoglycoside susceptibility of bacterial hybrid ribosomes in AUG(UUU)₁₂-directed polypeptide synthesis. Dose-response curves of bacterial (black circles), mitochondrial (red squares), mutant A1555G (green triangles), and mutant C1494T (blue inverted triangles) hybrid ribosomes. Corresponding IC₅₀ values are given in Table 2.

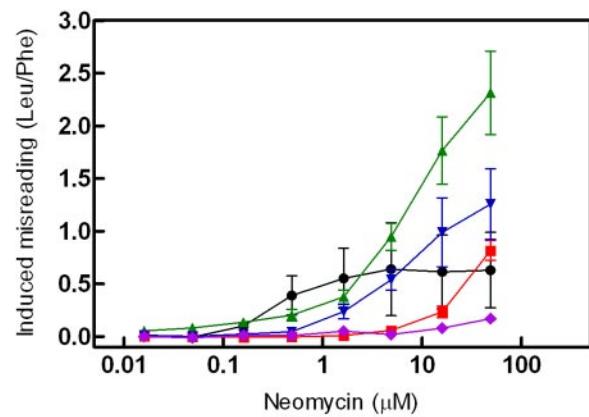
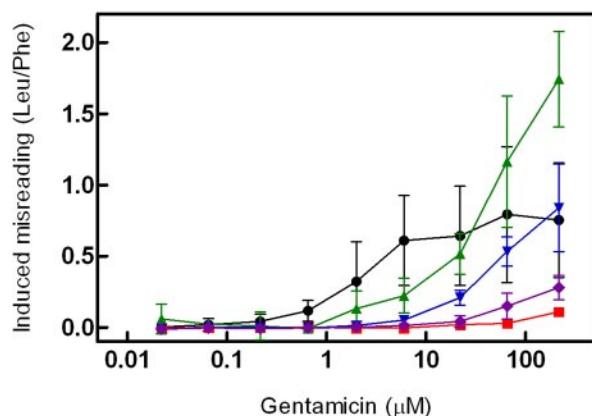


Fig. S2. Aminoglycoside-induced miscoding relative to a drug-free control in a *M. smegmatis* G1491C mutant in comparison to mutant mitochondrial decoding sites. Dose-response curves of *M. smegmatis* G1491C (purple diamonds), mitochondrial (red squares), mutant A1555G (green triangles), and mutant C1494T (blue inverted triangles) hybrid ribosomes. Bacterial wild-type ribosomes (black circles) are included for comparison.

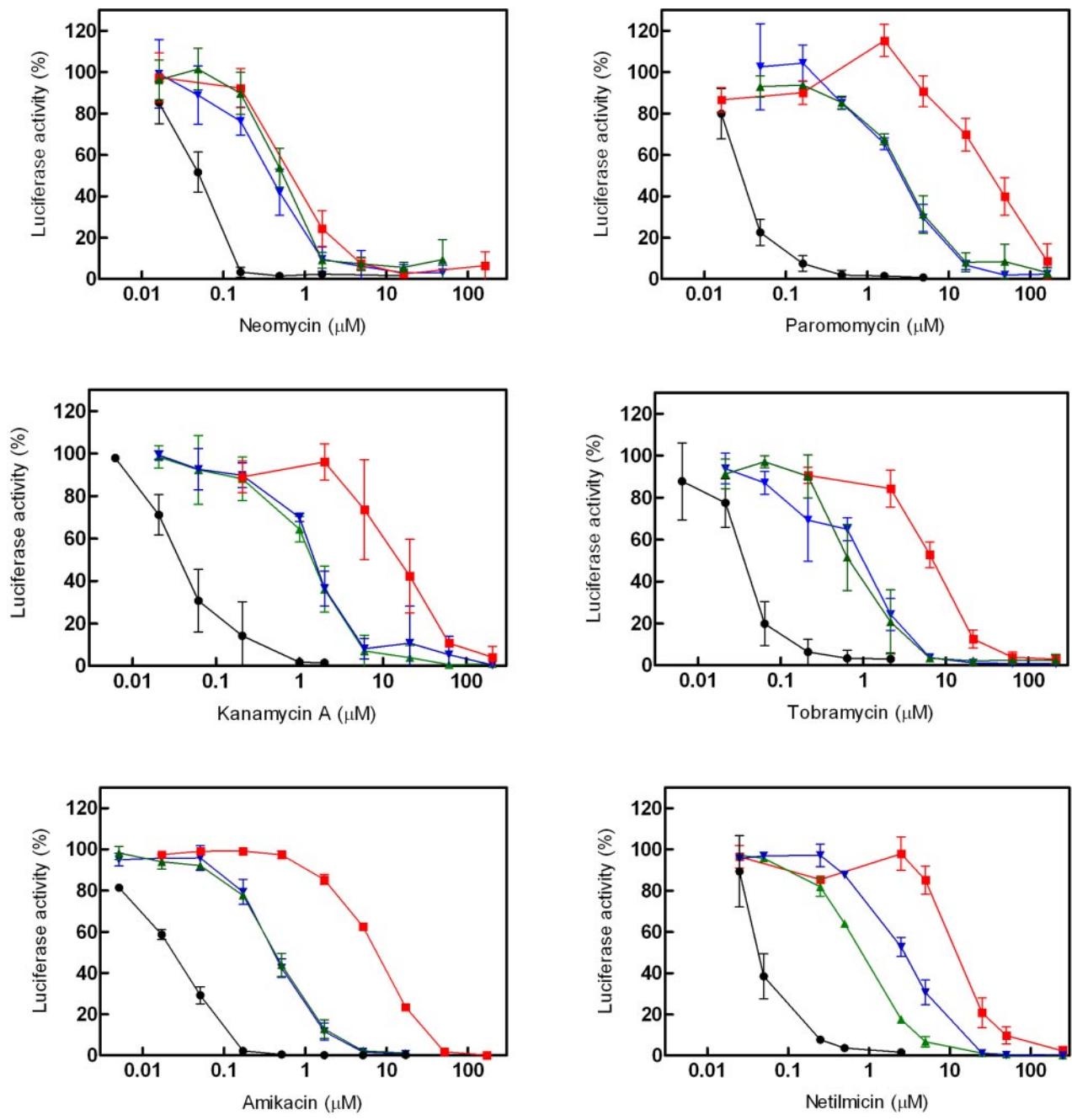


Fig. S3. Aminoglycoside-induced inhibition of luciferase synthesis. Dose-response curves of bacterial (black circles), mitochondrial (red squares), mutant A1555G (green triangles), and mutant C1494T (blue inverted triangles) hybrid ribosomes. Corresponding IC₅₀ values are given in Table 3.

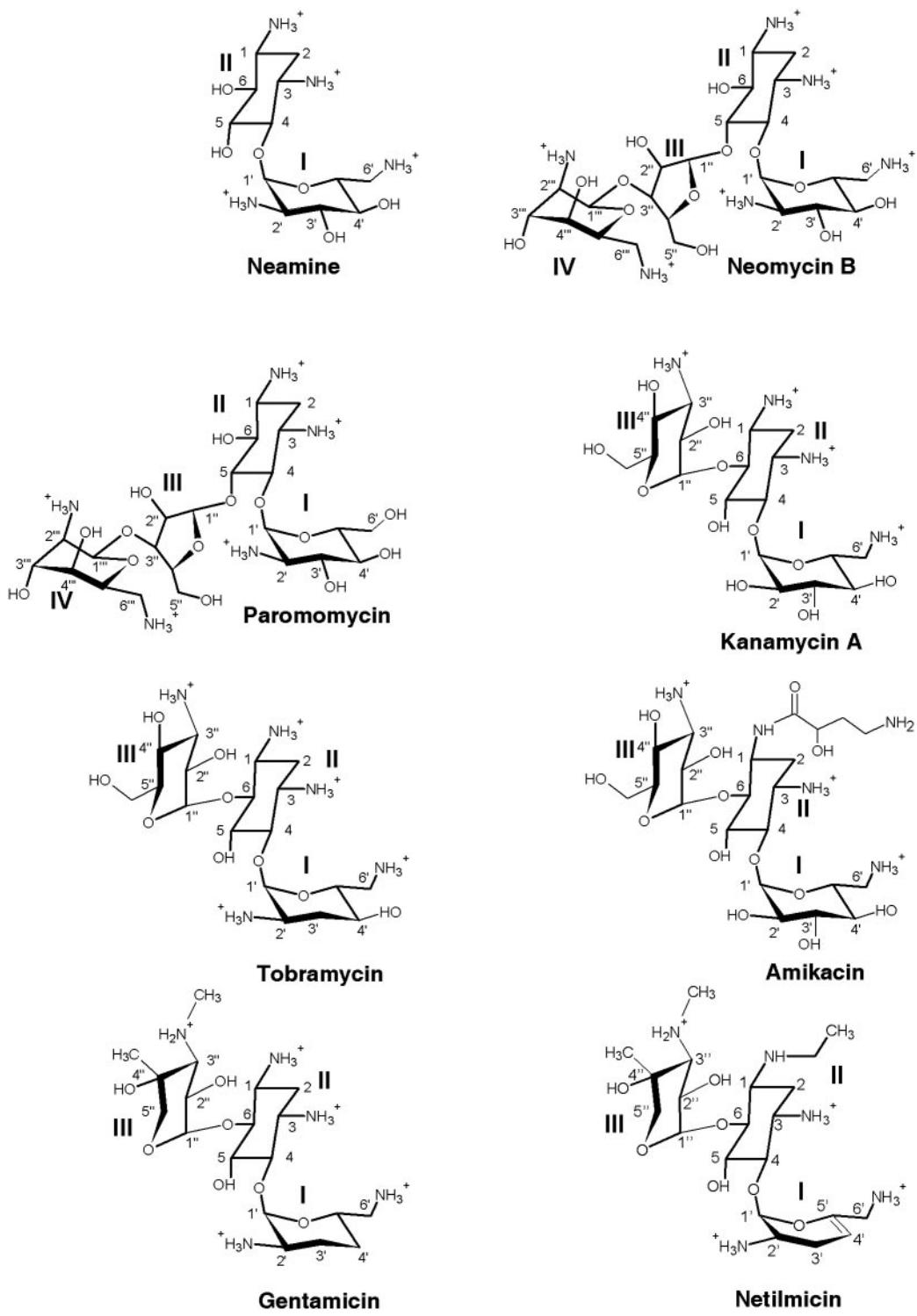


Fig. S4. Chemical structures of 2-deoxystreptamine antibiotics used in this study.

Table S1. Minimal inhibitory concentrations (μ g/ml) against *M. smegmatis* G1491C mutant in comparison to mutant mitochondrial hybrids

	<i>M. smegmatis</i>	A-site rRNA			
		Mitochondrial (mt) hybrid	mt A1555G hybrid	mt C1494T hybrid	<i>M. smegmatis</i> G1491C
Neomycin B	0.5	16–32	8	8	16
Paromomycin	1	> 1,024	256–512	256–512	512
Kanamycin A	1	256–512	16–32	16	16–32
Tobramycin	1	128	16	16	16
Amikacin	0.5	32–64	2–4	2–4	4
Gentamicin	1	64–128	16–32	16–32	16–32
Netilmicin	2	512–1,024	64	64–128	128

Table S2. Aminoglycoside susceptibility of the mitochondrial versus the cytosolic decoding site

	MIC ($\mu\text{g/ml}$)	
	Mitochondrial hybrid	Cytosolic hybrid
Neomycin B	16–32	> 1,024
Kanamycin A	256–512	> 1,024
Tobramycin	128	1,024
Amikacin	32–64	512–1,024
Gentamicin	64–128	> 1,024
Netilmicin	512–1,024	> 1,024