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NuoIEco -----
Nqo9Tth -----
TYKYHsa -----MRCLTTPMLLRAL---AQA--ARAGPPGGRSLHSSAVAATYKYVNMQDP 44
TYKYBta -----MRCLTMPMLLRAL---AQAQAARAGHASVRGLHSSAVAATYKYVNLREP 46
TYKYX1a -----MLGARLIYNAA---KRP---GIPPVLSRSLSLASQMHSYKYVNAREE 41
Nqo9Pde -----MAFDFA----- 6
NuoIRca -----MAFDYV----- 6
TYKYPpa MFKQLITAVPRQPIVGQRFISNASRLYKPAATAAEYLTTGGSTQSVWPKGFRAPKPKTW 60
EchFMba -----

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NuoIEco -----MTLKELLVGFQTQVRSIWMIGLHAFAKRETRMYPEEPVYLP PPRYRGRIVLTRD 53
Nqo9Tth -----MTLKALAQSLGITLK-----YLFKPVTVPYPDAPVALKPRFHGRHVLRH 46
TYKYHsa EMDMKSVTDRAARTLLWTELEFRGLGMTLSYLFREPATINYPFEKGPLSPRFRGEHALRRY 104
TYKYBta SMDMKSVTDRAAQTLWTELEIRGLGMTLSYLFREPATINYPFEKGPLSPRFRGEHALRRY 106
TYKYX1a ATDLKSVTDRAAQTLWTELEFRGLGMTLSYMFREPATINYPFEKGPLSPRFRGEHALRRY 101
Nqo9Pde -----RATKYFLMWDFIKGFGLGMRYFVSPKPTLNYPHEKGPLSPRFRGEHALRRY 57
NuoIRca -----RAKYFVLWDFIKGFALGMKYFVAPKPTLNYPHEKGPLSPRFRGEHALRRY 57
TYKYPpa EESSESALS KATKFFFLSEIAR GMYICMEMYFRSPYTIYYPFEKGPI SPRFRGEHALRRY 120
EchFMba -----MGMLNLVLTNISRKPATRLYPFEIREFPFKEFKGRIVFD-- 38

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NuoIEco PDGEERC VACNLCAVA CPVGCISLQKAETK-----DGRWYPEFFRINFSRCIFCGLCEE 107
Nqo9Tth PNGLEKCI GCSLCAAACPAYAIYVEPAENDPENPVSA GERYAKVYEINMLRCIFCGLCEE 106
TYKYHsa PSGEERC IACKLCEAICPAQAITIEAEPR-----DGSRRTRYDIDMTKCIYCGFCQE 158
TYKYBta PSGEERC IACKLCEAVCPAQAITIEAEPR-----DGSRRTRYDIDMTKCIYCGFCQE 160
TYKYX1a PSGEERC IACKLCEAACPAQAITIEAEPR-----DGSRRTRYDIDMTKCIYCGFCQE 155
Nqo9Pde PNGEERC IACKLCEAVCPAQAITIDAEPRE-----DGSRRTRYDIDMTKCIYCGFCQE 111
NuoIRca PSGEERC IACKLCEAICPAQAITIDAEPRE-----DGSRRTRYDIDMTKCIYCGY CQE 111
TYKYPpa PSGEERC IACKLCEAICPAQAITIEAEERI-----DGSRRTYKYDIDMTKCIYCGY CQE 174
EchFMba ---PEN CILCGLCQKKCPDAITVTKADKT-----WELNLFRCIMCTECVN 81

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NuoIEco	ACPTTAIQLTLPDFEMGEYKRQDLVYEKEDLLISGPGKYPEYNFYRMAGMAIDG-----KD	162
Nqo9Tth	ACPTGAIVLGYDFEMADYEYSDLVYGKEDMLVDVVGTKPQRREAKRTGKPVKVGIVVPPYV	166
TYKYHsa	ACPVDIVVEGPNFEFSTETHEELLYNKEKLLNNGDKWEAEIAANIQADYLYR-----	210
TYKYBta	ACPVDIVVEGPNFEFSTETHEELLYNKEKLLNNGDKWEAEIAANIQADYLYR-----	212
TYKYXla	ACPVDIVVEGPNFEFSTETHEELLYNKEKLLNNGDKWEVEIGANIQADFLYR-----	207
Nqo9Pde	ACPVDIVVEGPNFEYATETREELFYDKOKLLANGERWEAEIARNLQLDAPYR-----	163
NuoIRca	ACPVDIVVEGPNFEYATETREELFYTKKLLENGARWEAEIARNIEMDAPYR-----	163
TYKYPpa	SCPVDIVVESPNVEYSTATREELLYNKEKLLENGDKWEQELQYCIDADAPYR-----	226
EchFMba	GCPKGCLSISN--ERAKTGAEVVIKIAVPIVDKPKAPKAAP-----	120

NuoIEco	KGEAENEAKPIDVKSLLP	180
Nqo9Tth	RPELEGFKAPTEGGKR--	182
TYKYHsa	-----	
TYKYBta	-----	
TYKYXla	-----	
Nqo9Pde	-----	
NuoIRca	-----	
TYKYPpa	-----	
EchFMba	-----	

Supplemental Figure 2. Similarity of the deduced amino acid sequences among the *E. coli* NuoI subunit and its homologues from various organisms. The alignment was carried out using the ClustalW program. Red boxes with white letters show perfectly conserved residues among the listed organisms. Blue boxes with white letters illustrate conserved residues among at least five listed organisms. Amino acids mutated in this study are marked by arrows. Sequence sources and their Swiss-Prot accession numbers are: NuoIEco, *E. coli* K-12 NuoI subunit [P0AFD6]; Nqo9Tth, *T. thermophilus* HB-8 Nqo9 [Q72GD6]; TYKYHsa, *Homo sapiens* TYKY subunit [O00217]; TYKYBta, *Bos taurus* TYKY subunit [P42028]; TYKYXla, *X. laevis* TYKY subunit [Q66KT2]; Nqo9Pde, *P. denitrificans* Nqo9 subunit [A1B486]; NuoIRca, *R. capsulatus* NuoI subunit [P42031]; TYKYPpa, *Pichia pastoris* TYKY subunit [A3LRW9]; EchFMba, *Methanosarcina barkeri* EchF subunit [O59657].