

NuoMEcoli ---MLLPWLILIPFIGGF<sup>L</sup>LCWQTER----FGVKVPRWIALITMGLT<sup>L</sup>LALS<sup>L</sup>QLWLQGGYS 53  
 Nqo13Thermus ----M<sup>V</sup>VLA<sup>V</sup>LLP<sup>V</sup>VVFGALLLLG-----LPRALG<sup>V</sup>L<sup>G</sup>AGLS<sup>F</sup>LLNLYLFLT---- 42  
 Nqo13Paracoccus -MTNLLSIITFLPIVAAILMALFLRGQDEAAARNAKWLALLTTTATFVISL<sup>F</sup>VLF<sup>R</sup>---F 56  
 NuoMRhodobacter -MENLLSIITFIPLVAALIMALFLRGDDAAAQRNAKWLALLATSATFLVSL<sup>F</sup>LLAG---F 56  
 NuoMPseudomonas ---MILPWLILIPFIGGF<sup>L</sup>LCWIAEH----SSKTLPRWIALLSMTLV<sup>L</sup>ILSLWIWATGDFQ 53  
 NuoMStretomyces MSFP<sup>L</sup>LLTVTAALPAVGAIATAAVPA----AKRTAAKWLALLVSLATLGLAIAVLV<sup>R</sup>---- 52  
 ND5Yarrowia -MFLTSILLSSLYL<sup>F</sup>NRILAWQGNV-----KHFYLFASNLL<sup>L</sup>LFIVVLYINFNTFSN 51  
 ND4Bovine ---MLKYIPTIMLMP--LTWLSKN-----NMIWVNSTAHSLLISFTSLLLMNQFG- 46  
 ND4Human ---MLKLIVPTIMLLP--LTWLSKK-----HMIWINTTTHSLIISIIP<sup>L</sup>LLFFNQIN- 46

NuoMEcoli LTQSAG-IPQWQSEFDMPWI<sup>P</sup>RFGISIH<sup>L</sup>AIDGLSLLMVVL<sup>T</sup>GLLGVLAVLCSWKEIEKY 112  
 Nqo13Thermus --HPGG----VAHAFQAPLLPGAGVYWAFGLDGLSALFFLTIALTVFLGALVARVEG--- 93  
 Nqo13Paracoccus DPANTG----FQFVEDHAWIM--GVCYKMGVDGISVLFVLLTTFMMP<sup>L</sup>TILSTWQVQDKV 110  
 NuoMRhodobacter DAQDTG----FQFVESYDWIA--GLQYKMGVDGISILFVMLTTFMPLTIASAWEVETRV 110  
 NuoMPseudomonas LAPAPGGEPEWTLQFKVLWIERLGISVHLAMDGLSLLMVALTGLLGVLSVLC<sup>S</sup>WNEIQRR 113  
 NuoMStretomyces FDPDGD--RYQLTESHSWIADFGVRYELGVDGIAVALIALTALLIPFIILAGWHDADPL 109  
 ND5Yarrowia SFQFNF-ELFNSLNPFGLSNSDI<sup>S</sup>NGLLFGIDGLSLTFILLTVLLIPLTLLGNWYNIN-- 108  
 ND4Bovine -----DNSLNFSLLFFSDSLSTPLLILTMWLLPLMLMASQHHSKE 87  
 ND4Human -----NNLFS<sup>C</sup>SPTFSSDPLTTPLLMLTTWLLPLTIMASQRHLSSE 87

NuoMEcoli QG-----FFHLNLMWILGGVIGVFLAIDMFLFFFWEMMLVPMYFLIALWG----- 158  
 Nqo13Thermus -----RFLGLALLMEGLLLGLFAARDLLV<sup>F</sup>YVFFEAALIPALLMLLYG----- 137  
 Nqo13Paracoccus KE-----YMIAFLVLEGLMIGVFTALDLVLFYLF<sup>F</sup>EAGLIPMFLIIGIWG----- 155  
 NuoMRhodobacter KE-----YMIAFLILETLMLGVFCALDLVLFYLF<sup>F</sup>EAGLIPMFLIIGIWG----- 155  
 NuoMPseudomonas IG-----FFHLNLLWILGGVIGVFLAIDLFLFFFWEMMLVPMYFLIALWG----- 159  
 NuoMStretomyces ETGSSRWRPTQGF<sup>F</sup>FALILAVEAMV<sup>I</sup>ISFEATDVFLFYIFFEAMLIPLYFLIGGF<sup>G</sup>DRAHE 169  
 ND5Yarrowia -----FNSNLYYTLVLAIGLVILLNFWALDYISFYILFEATLPLLFI<sup>L</sup>IHIYGS---- 157  
 ND4Bovine NL-----TRKKL<sup>F</sup>ITMLISLQLFLIMTFTAMELILFYILFEATLVPTLIIITRWGNQ--- 139  
 ND4Human PL-----SRKKLYLSMLISLQISLIMTFTATELIMFYIF<sup>F</sup>ETT<sup>L</sup>IPTLAIITRWGNQ--- 139

NuoMEcoli HKASD GK-TRIT AATKFFIY TQASGLVML IAILALVFVHY NATGVWTFNYEE-----LL 211  
 Nqo13Thermus -----GEGRTR ALYTFVLF TLVGSLPMLA AVLGARLLSG----SPTFLLED-----LL 181  
 Nqo13Paracoccus -----GKDRIY ASFKFFLY TFLGSLVMLV AMIAMYRMA GTTDIPTLLTFDFPSENFRLI 209  
 NuoMRhodobacter -----GKDRIY AAFKFFLY TFLGSLVML IAMIAMY YDAGTTDIP TLLSHEFSSGTMTLA 209  
 NuoMPseudomonas HSSDDGKK TRIYAATKFFIFT QASGLVMLVA IILGLVFVNFNATGVI TFDYAT-----LL 213  
 NuoMStretomyces HGEKTAATQRSYAAVKFLLYNLAGGLIMLAAVIGLYV VVAGNFSLTEIAEARA-----NG 223  
 ND5Yarrowia -----SDSER ASFYVLMFTL SGSLFMLLSIVVISIVLNTTNF INHNLFVLSLD----- 205  
 ND4Bovine -----TERLNAGLYFLFYTLGAGSLPLLVALIYIQNTV GSLNFLMLQYVWQPVHN---- 188  
 ND4Human -----PERLNAGTYFLFYTLVGS LPLLIALIYTHNTL GSLNILLTLTAQELSN---- 188

NuoMEcoli NTPMSSGVEYLLMLGFFIAFAVKMPV VPLHGWL PDAHSQAPTAGSV DLAGILLKTAAYGL 271  
 Nqo13Thermus AHPLQEEAAFWVFLGFALAFAIK TPLFPLHAWLP PFHQENHPSGLADALG TLYKVGVF 241  
 Nqo13Paracoccus GMTVVGGMQMLLFLAFFASFAVKMP MWPVHTWLP DAHVQAPTAGSVLLAAVLLKMGYGF 269  
 NuoMRhodobacter GFQIVGGLQTLFLAFFASFAVKMP MWPVHTWLP DAHVQAPTAGSVVLAAILKMGYGF 269  
 NuoMPseudomonas KTQLSPHVEWLLMLGFFVAFAVKMP VVH SWLP DAHAQAPTAGSV DLAGILLKTAAYGL 273  
 NuoMStretomyces SLDMATSTERWFLFLGFFAFAVKAPL FPLHTWLP NAMGESTAPVAVLITAVVDK VGT 283  
 ND5Yarrowia -----LQTI IWLGLFIAIMVK TPLFPIHVWLP VVHSE SPLAGSMILAGLILKLALYAI 258  
 ND4Bovine -----SWSNVFMWLA CMMAFMVKMPLYGLHLWLP KAHVEAPIAGSMVLA AVLLKLG YGM 243  
 ND4Human -----SWANNLMWLA YTMAFMVKMPLYGLHLWLP KAHVEAPIAGSMVLA AVLLKLG YGM 243

NuoMEcoli LRFSLPLFPNASAEFAPIAMWLG VIGIFYGAWMAFAQT DIKRLIAYTSVSHMGFVLIAY 331  
 Nqo13Thermus FRFAIPLAPEGFAQAQGLLLFLAALSALY GAWVFAAKDFK TLLAYAGLSHMGVAALGVF 301  
 Nqo13Paracoccus LRFSLPMFPV ASGVAQPYVFWLSAIAIVY TSLVALAQSDMKKVIAYSSVAHMGYV TMGVF 329  
 NuoMRhodobacter LRFSLPMFPIGSELLSPLVFWMSAIAIVY TSLVALAQSDMKKLIAYSSVAHMGYV TMGIF 329  
 NuoMPseudomonas IRFALPLFPNASAEFAPIAMWLG IIGIFYGALLSFAQT DIKRLVAYSSVSHMGFVMIGIY 333  
 NuoMStretomyces LRFCLQLFPEASKWATPVILVLAVIS IYIYGALLAVGQRDIKRLIAYASISHFGFIIMGIF 343  
 ND5Yarrowia LRLLLPLLCEAQILYTPMIYIISLLTI ILSLATLRQIDLKVI IAYSSISHMGIAILGVC 318  
 ND4Bovine LRITLILNPMTDFMAYPFIMLS-LWGMIM TSSICLRQTDL KSLIAYSSVSHMALVIVAIL 302  
 ND4Human MRLTLILNPLTKHMAYPFLVLS-LWGMIM TSSICLRQTDL KSLIAYSSVSHMALVVTAIL 302

NuoMEcoli TGSQLAYQGAVIQ<sup>▲</sup>MIAHGLSAAGLFILCG-QLYERIHTRDMRMMGGLWSKMKWLPALS<sup>▲</sup>LF 390  
 Nqo13Thermus SGTPEGAMGGLYL<sup>▲</sup>LAASGVYTGGLFLLAG-RLYERTGTLEIGRYRGLAQSAPGLAALALI 360  
 Nqo13Paracoccus AANQIGVDGAIFQ<sup>▲</sup>MLSHGFISGALFLCVG-VIYDRMH<sup>▲</sup>TREIDAYGGLVNRMPAYAAVFMF 388  
 NuoMRhodobacter AANQQGVDGAIFQ<sup>▲</sup>MLSHGFISGALFLCVG-VIYDRMH<sup>▲</sup>TREIDAYGGLVNRMPAYALIFMF 388  
 NuoMPseudomonas SGSQVALQGVVVQ<sup>▲</sup>MIAHGLSAAALFILCG-QLYERLHTRDMRKMGG<sup>▲</sup>LWSRMPYLPAISLF 392  
 NuoMStretomyces AMTSQ<sup>▲</sup>QSGATLYMVNHGISTAVLMLIAG-FLISR<sup>▲</sup>RGSR<sup>▲</sup>LIADFGGVQKVAPILAGTFLI 402  
 ND5Yarrowia SNTSLGIYGSIVLGV<sup>▲</sup>AHGFVSPALFLIVGGILYDRYHIRIVN<sup>▲</sup>YKGLTTYMPQLATYIII 378  
 ND4Bovine IQTPWSYMGATALMIAHGLTSSMLFCLAN-SNYER<sup>▲</sup>IHSRTMILARGLQ<sup>▲</sup>TLLPLMATWLL 361  
 ND4Human IQTPWSFTGAVILMIAHGLTSSLLFCLAN-SNYERT<sup>▲</sup>HSRIMILSQGLQ<sup>▲</sup>TLLPLMAFWLL 361

NuoMEcoli FAVATLGM<sup>▲</sup>PGTGNFVGEF<sup>▲</sup>MILFGSFQVVPVITVISTFGLV<sup>▲</sup>FASVYSLAMLHRAYFGKAKS 450  
 Nqo13Thermus LFLAMVGLPGLSGFPGEFLTLLGAYKASPWLAALAF<sup>▲</sup>LSVIASAA<sup>▲</sup>YALTA<sup>▲</sup>FQKTFWEEGGS 420  
 Nqo13Paracoccus FTMANVGLP<sup>▲</sup>GTSGFVGEFLTLMGVFRVDTWVALVATSGVILSAA<sup>▲</sup>YALWLYRRVTLGQLIK 448  
 NuoMRhodobacter FTMANVGLP<sup>▲</sup>GTSGFVGEFLTLVGIFQVNTWVAMVATSGVILSAA<sup>▲</sup>YGLWLYRRVVFGE<sup>▲</sup>LVK 448  
 NuoMPseudomonas FASASLGLP<sup>▲</sup>GTGNFVGEF<sup>▲</sup>LILIGAFKVV<sup>▲</sup>PVIIV<sup>▲</sup>IATFGLV<sup>▲</sup>FASVYSLIMIHRAYFGPSQS 452  
 NuoMStretomyces GGLATLSL<sup>▲</sup>PGLAPFVSEFLVLVGTFTRYPVIGI<sup>▲</sup>IATLGI<sup>▲</sup>VLAALYTLVLYQRTMTGPVKA 462  
 ND5Yarrowia LSFANIGT<sup>▲</sup>PLTGNFTGEFLSLQGGFIRNPIIGGISCISVLLAA<sup>▲</sup>IYQLKLTNKL<sup>▲</sup>TGGISSI 438  
 ND4Bovine ASLTNLALPPTINLIGELFVVMSTF<sup>▲</sup>SWSNITIIILMGVNMVITALYSLYMLIMTQRGKYTY 421  
 ND4Human ASLANLALPPTINLLGELSVLVTTFSWSNITLLLTGLNMLVTALYSLYMF<sup>▲</sup>TTTQWGLSLTH 421

NuoMEcoli QIASQELPGMSLRELF<sup>▲</sup>MILLLVLLLVLLGFYPQPILD<sup>▲</sup>TSHSAIGNIQQWFVNSVTTTRP- 509  
 Nqo13Thermus -----GVKDLAGA<sup>▲</sup>EWGFALLSVLALLLMGVFP<sup>▲</sup>GYFARGLHPLAEAFKLLGGGA----- 469  
 Nqo13Paracoccus -ESLKSITDMTP<sup>▲</sup>RE<sup>▲</sup>RWF<sup>▲</sup>FIPLIAMT<sup>▲</sup>LILGVY<sup>▲</sup>PRLVTDV<sup>▲</sup>TGP<sup>▲</sup>AVAALVQDYNQSQPAAPVA 507  
 NuoMRhodobacter -EALRTIRDM<sup>▲</sup>DREKA<sup>▲</sup>IFAPLVAMT<sup>▲</sup>LLLGVY<sup>▲</sup>PSLVTDI<sup>▲</sup>IGPSVARLTTDYQSATAALEAG 507  
 NuoMPseudomonas ---DEPILGLDARELSMVLGLAVLLVLLGVY<sup>▲</sup>QPVL<sup>▲</sup>DISAASM<sup>▲</sup>HGVQQLGAALSTLAGR 509  
 NuoMStretomyces --EVNGMPDLRVREL<sup>▲</sup>VV<sup>▲</sup>VAPLVAL<sup>▲</sup>LIFLGV<sup>▲</sup>FPK<sup>▲</sup>PVTDIVNPAVEQTMSDVHKTDPQPEVE 520  
 ND5Yarrowia --YMHRTNDVTIREK<sup>▲</sup>FIMN<sup>▲</sup>ILIIISTLIIGIC<sup>▲</sup>PQIMYNLLYWTVN<sup>▲</sup>NYIYII----- 486  
 ND4Bovine --HINNISPSTRENALMSLHILPL<sup>▲</sup>LLLT<sup>▲</sup>LNPKIILGPLY----- 459  
 ND4Human --HINNMKPSFTRENTLMFMHLSPI<sup>▲</sup>LLLSLN<sup>▲</sup>PDIITGFSS----- 459

NuoMEcoli	-----	
Nqo13Thermus	-----	
Nqo13Paracoccus	TAQASH	513
NuoMRhodobacter	TRLAME	513
NuoMPseudomonas	-----	
NuoMStreptomyces	AAK---	523
ND5Yarrowia	-----	
ND4Bovine	-----	
ND4Human	-----	

**Supplemental Figure 2. Comparison of the amino acid sequence of *E. coli* NuoM subunit with its homologs.** The totally conserved residues are shown in red letters, while blue letters indicate residues conserved in almost 60% of the cases. Amino acids that are conserved in almost 50% of the cases or the residues having similar characteristic in about 80% of the cases are shown in green letters. Yellow shadows indicate the predicted transmembrane helices. Potential transmembrane helices are underlined. Red arrows indicate residues mutated to introduce a STOP codon. Sequence sources and their accession numbers are: NuoMEcoli: *Escherichia coli* K-12 [P0AFE9], Nqo13Thermus: *Thermus thermophilus* [Q56228], Nqo13Paracoccus: *Paracoccus denitrificans* [P29925], NuoMRhodobacter: *Rhodobacter capsulatus* [P50974] NuoMPseudomonas: *Pseudomonas aeruginosa* [26990824], NuoMStreptomyces: *Streptomyces coelicolor* A3 [Q9XAR6], ND4Yarrowia: *Yarrowia lipolytica* [Q9B6D6], ND4Bovine: *Bovis taurus* [P03910], ND4Human: *Homo sapiens* [Q4F0G8].