

1 **Supplemental Fig. 1. Sequence alignment of Cu-containing nitrite reductases.** Amino acid
2 sequence of the enzyme from *N. oceani* (Nco) is aligned with those from *A. xylooxidans* (Axy), *B.*
3 *japonicum* (Bja), *A. cycloclastes* (Acy), *A. faecalis* (Afa), *N. gonorrhoeae* (Ngo), *B. pseudomallei*
4 (Bps), *F. oxysporum* (Fox), *A. oryzae* (Aor), *H. marismortui* (Hma), *H. volcanii* (Hvo), *N. europaea*
5 (Neu), and *N. maritimus* (Nma). Amino acids are numbered at the right margin. Putative Cu-binding
6 residues for type 1 and type 2 Cu are indicated by I and II, respectively. Insertions and deletions of
7 the sequence that are common in the class 1 or class 2 enzymes are boxed. Accession numbers of
8 the sequences are indicated in Figure 1.

9

		II I I	
Noc	LRFKADKAGTYVYHCAPGGL-MTPYHVVSGMYGAIMIFPKKGLRDQNGKKVYTDKAYYVGEQGWYIPKDK		218
Axy	LRFKADRSGLTFVYHCAPEG--MVPWHVVGMSGTLMLVLRDGLKDPQGKPLRYDRAYTIGFEDLYIPKDA		207
Bja	LRWKATKTGVFVYHCAPGGP-MIPWHVVSQMGAVMVLPRDGLNDGKGHALKYDKVYVYVGEQDMYVPRDE		211
Acy	LRFKATKPGVFVYHCAPEG--MVPWHVVSQMGAIMVLRDGLKDEKQPLTYDKIYVYVGEQDFYVPKDE		226
Afa	LRFKATKPGVFVYHCAPPG--MVPWHVVSQMGAIMVLRDGLHDKGKALTYDKIYVYVGEQDFYVPRDE		225
Ngo	FSFKALQPLGIYIYHCAVAP---VGMHIANGMYGLILVEPK-----EGLPKVDKEFYIVQGDFYTK---		217
Bps	FTFKALNEGLFVYHCAVAP---VGMHVANGMYGLILVEPP-----EGLPKVDREYVVMQGFYTN---		209
Fox	GRFKLLYPGLVYVYHCAAAP---VPVHIANGMYGLMYVQPEG-----NDLPPVDKEYVVMQSEFYHEPPE		340
Aor	ARFRLQNPGLYIYHCAVGP---VGVHIANGMYGLLYVQPE-----QDLPPVDKEYVVMQSEFYHEPPE		297
Hma	FRFKATYPGAFIYHCAVFN---LDMHISSGMFGMILVEPK-----EGLPEVDHEFYFGQHELTYT---		216
Hvo	LRFKATYPGAYIYHCAVFN---MDMHISSGMFGMILVEPP-----EGMPEVDHEIYLQHELTYT---		210
Neu	YTFTADNPGVFFYHCGSDP---MICHIAARGMYGVIVDPKD-----ANALPKADREYVLIQAEHYEN---		167
Nma	YCYIAESAGMFKYHCSGVKLIQMDCHVLSQMYGITIVDPANG--YKLMVEKTSVSGELDRKFDADALE		226
Noc	NGKYKRYANAIEPYGDTLEVMRGLVPTHTVYNGSKGALTG-----		258
Axy	NGKYKDYPTLAESYGDVAVMRTLTPSHIVFNGKVGALTG-----		247
Bja	KGNFKSYDSPGEAFTDTEEMMKLIPSHVFNKVGALTG-----		251
Acy	AGNYKKYETPGEAYEDAVKAMRTLTPTHIVFNGAVGALTG-----		266
Afa	NGKYKYEAPGDAYEDTVKMRTLTPTHVFNKVGALTG-----		265
Ngo	-----GKKGAQGLQPFDMDKAVAEQPEYVVFNGHVGAIAAG-----		252
Bps	-----GKYREKGLQSFDMDKAIDERPTYVFNKVGALTG-----		244
Fox	---VDDGRRSEIVEFSYPNGLREEPQVAVFNKSESALTR-----		377
Aor	---PDDNGQMSSTVEFSWPHALREAADVVFNKSEAAALT-----		333
Hma	-----GDTGEGHHDFFDEAMAAEPTVYVLMNGEKYAITPDRH-----		254
Hvo	-----KDAGEEGQHAFDYEAMRNEEPTVYVLMNGEKYAWTPNGR-----		248
Neu	-----PDDKTAMMKNKWSNVVFNKGGVFKYDPVHDS-----EA		199
Nma	FQLQYVQLYLTPEGNYDAGAMFKHONTATVFNKMGQFQVYVPMMAHLLVNGDVNKNIFVAQPWNGLEHKQY		296
Noc	DNAMKAKVGDVSLVFIHS--QANEDSRIHLIGGHGDLVWPGGSFKNTMPVMDKETWHVNGGESVAALYTFKQ		326
Axy	ANALTAKVGETVLLIHS--QANRDTRPHLIGGHGDVWVETGKFNPPQKDLQETWIFIRGGSAGAALYTFKQ		315
Bja	KNALTANVGENVLIVHS--QANRDSRPHLIGGHGDYVWETGKFGNAPEVGLQETWIFIRGGSAGAAKYFMQ		319
Acy	DHALTAAVGERVLVHS--QANRDTRPHLIGGHGDYVWATGKFRNPPDLQETWILIPGGTAGAAFYTFRQ		334
Afa	DKAMTAAVGERVLIVHS--QANRDTRPHLIGGHGDYVWATGKFNTPDQETWIFIPGGTAGAAFYTFQQ		333
Ngo	DNALKAKAGETVRMYVGNNGPMLVSSSFHVIGEIFDKVYVEGGKLINEN--VQSTIVPAGGSAIVEFKVDI		320
Bps	ERAMRARTDETURLFVGNNGPMLVSSSFHVIGAVFDKVRADGSNVTQND--VQTTIPAGGAATIEFHTRV		312
Fox	DHPLKAHVGDVDRIFFGNAGPNLTSSSFHIGTHFKNVYRDGGVTSNPSKGIQTVSVPCGGSTIVDLKMAV		447
Aor	EKPLKATLDDTVRIFFGNAGPNLTSSSFHIGTCFNKYRDSVLSPPGQCQTVSVPPGGSTIVDMKVVV		403
Hma	G-SPSMQVGETARVYFVTGGPNLSSSFHPIGSVWDEVWQQGSIAGPPNRYVQTTIPVKPGSCAIATLHAEV		323
Hvo	GPAATVGLDETVRVYFVDGGPNLSSSFHPIGSVWETLYPEGSLTTEPQTHIQTRQVPPGSTTIATMSSPV		318
Neu	TSWLQAKPGERVRIYFVNAGPNELSSLHPIAGIWRVYPSGN-PKNVQYALQSYLIGAGDAATLDLISPV		268
Nma	QSQLLFVENDQHURLFVENQNEPVFFHIVGEILDRVTQGNRVQSAAT---ETWLLGGSQGMIVDLVDFE		363
		II	
Noc	PGLYAYLHNLIAQAFMLGGAHVKVDGE-WNNDLMEQVVKPSPINAD.		372
Axy	PGVYAYLHNLIEAFELGAAGHIKVEGK-WNDDLKMKQIKAPGPIPR.		360
Bja	PGIYAYVHNLIEAADLGATAHFKVEGK-WNDDLMTQVKAPAEIPANTN.		367
Acy	PGVYAYVHNLIEAFELGAAGHFVKTGE-WNDDLMTSVVKPASM.		377
Afa	PGIYAYVHNLIEAFELGAAGHFVKTGE-WNDDLMTSVLAPSGT.		376
Ngo	PGNYTLVDHIFRAFNKALGQLKVEGA-ENPEIMTQKLSDTAYAGSAAAS(+22aa).		392
Bps	PGNYTFVDHIFRAFNKALAILKVDGP-ENKAIYSGKELDAPYSG-DAVT(+159aa).		520
Fox	PGTYTLVDHIFR-LDKGAVGFLNVSGP-QNPGVYQSSQPPRCVCGCKLHS.		496
Aor	PGTYTIVDHAIFR-LEKGAKGFLNVSGE-PRPMLYYSTLPPQCEGFNLKY(+107aa).		559
Hma	PGPIKLVHSLSRVARKGTMAIINREGA-ANPDVFEPEA.		361
Hvo	PGDFKLVHSLSRVVRKGCMAVVRAEGA-EDPEIFDPPDPQ.		359
Neu	EGANAIVDHSRHAHSGAIAVIMFTNDA-DPEARGENILIR.		309
Nma	PGAYAAVNHEDYAAIYTGAAVTFVAGDPFGLNPLVLEKGVIPAPVASAYAL(+56aa).		470