

**FIG. S1.** Growth kinetics of *Chlamydia muridarum* Nigg and plaque-purified isolates in L929 cells.

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TC_0447      1 -----MTSPL-----STAASSYLRT--LQRAFPLGGGGYPT--N-----PNSAQTALRV---
TC_0434      1 -----MTAPL-----ITTTSPYLRL--LQKRFLGGSRLGPY--SLLFYET-----PAHRILTT-----TPSQEGMTSTKKIQG
TC_0435      1 -----MVSP-----ITTTSPYLLT--LQAR-----LLF-----TLTTS-----CSSQNQKCAKKIQE
TC_0436      1 MDPIRGFHQKSSKKIRSF--MRINETHLFV--LQERFLSWGGA--SSLFSWI-----PARRTLLTP-----SPYQQKEIFTEKVQK
TC_0440      1 -----MCSFPCPRHLFPVIVTDCPTCTSP--LRKRPLSSTTAAAVVVTQLFKYHRRKDLSHRVIYDYDHGKNKTPRKIQCSSNRHSLATSSKTSKSSSRKILR
TC_0433      1 ---MRAEFNIVKTKQKYMCSFCSRHFIPVDCPTCTSP--LRKRSLSTTAAALVATKVKFYPRKDLSHRVIYDYDHGKNKPARRIQFGSDRHSLETSSKTSRSSSRKILK
TC_0432
TC_0357      1 -----MKKTKQLISKI-----TFSLISLFIGG-----YLLKAPPNPQSSDTFQTFIE--
NucT (H. pylori)

TC_0447      41 --QTAATSLAIVPYRPLSSEPVSF--SKHCKNDAQSIIENAILSASSVFLKIFLSSEPIVQDLICKSEESIPVTIHYQH--IPEPIVKNLEEAGAEIRCHRNR
TC_0434      61 LTQTRIQSLTILVPSYPLSPPVCF--SRHCRYNALEIIEHAILSATTSIVLNTFNLSSERLIQALVLKAQQGVSVTVQYHH--MLSNTWFKLCKPNIEIIPLG-SKR
TC_0435      43 L--TRPSSLALVPYSRPLSSETVCF--SKHCGYNALDLIEKVLSATSSIVLKNVVPSSKSLAQALILKALQGVVPSIHCPH--KTLKALGDLSKQTN--VTLY-SSH
TC_0436      78 LPTP--SSLALIPYWNSSPSESICF--SKHCGHDAINVIETAILSAKTSIFLKFISISSKKITRALAKSIQGVSVSMLYHH--ISTKTYSDLSKTTIELIQFQENSH
TC_0440      94 ARRTGSS--VP--RPIAQQLITFISAQKTSNSNPKSICKFIDRTHQNLIHQVCFNHPTIIQHHLVKAAREVRVSVQYRDGTELEEACKD-STICLQKQS----GR
TC_0433      107 ARRTES--VP--RPVAQQLITFVSAQQTNSNPLSAICKLMLNLSKRLFIQVYRFTHTPIIQHALNVAAKNVRTVLFVFRDGDDELVEASQG-SPIILEQQP----QR
TC_0432      1 -----MSVICKLMLNQA--SLFIQVYRFTHTPIIQHALNVAAKNVRTVLFVFRDGDDELISAAQN-SSIILQQP----GR
TC_0357      43 -----SNEPIIF--SKQCGDNITQVLCDAINSAKKDI--FLSIYDLSSAII--SELKKQEE--SQIPLCI---HYQRISKSTNFSPPQLTVEHPPVK
NucT (H. pylori)

TC_0447      144 SLLHRKTMILDEKQVITGSANFTRNSINNDVNLARVNVNHIANLMOQKQKGAIIENSTKGGKQIVCYLFI-N-HKKCGN-ER-QIVKA--INNATSSIQIGMCIILTHRG
TC_0434      165 SLFHKKTLIIDKQVITGTGNFTIASLHRDVLNMMRVNSPELADLMEKNQKGVKCV----GEQTCYRPM-N-NRLDGN-EG-KILKE-IQKASSIQLGMCILTNEV
TC_0435      143 ASSDKQTLIVDEHQVVTGARNFTTSLHREANLMMRISLDELANLIEKNQKGEVCL----GQKVCYCFI-SKNKKG-N-EK-EILKE-IQKATSSIQLGTSCLTSQSI
TC_0436      181 ALLHNKTLIIDKQIITGNFTSASLCEVNLMMRINNMHLASLMEKNQAGRACI----KKQKRYLFPV-NR-----KG-EIVKE-IQKASSIQLGMYIILTNEAI
TC_0440      193 SLFHKSMVGSQRVLA--SSGFTPDSTEQDINLSLINDSELARHIETNQVTRQI----GNQFLVYIFIFRNRK-DFRLAV-RTIKKCAQAKKIVVVVYIILTHPVI
TC_0433      206 ALFHRKSLVVDHRMLLVSTGNFTINSTEQDINLSIVFHHSKLAARVEHSQPFSGVV----GNQPVSYLPIFRNRKSSQIGG-QFIQSFIEEAKSSILIAMYILSHPGI
TC_0432      69 SLFHRKSLVVDNKKLFISTGNFTINSTEQDINLSAIFKDALATRVAC--SFTGII----GSQVLSYLFYRRSQKSSQIGI-HLVQDLIQNAKSNILIAMYILSHPGI
TC_0357      128 KLMHOKTMAIDGKTAWIASANLFTSLEKSANLVIGLKSPELCHFIKTQTSIGICCI----NNQPIEYFSL----NEGNSAALEKVLQHIRSAKESIQIGMFALTLPQI
NucT (H. pylori)

TC_0447      249 LQALNEAATQRSVLVTIIID----SLESQOTIDILKALG---SKLRVVRGT-GDR----IHCACILDHNTAIIIG-SANWAASGLKANKEDIIVNPLTERQREDLSIWW
TC_0434      265 IQAINEAA-KRSVLVTIIID----PHKKAQTLLEMLKSLN---SQITLRIVTLFPY--MHCKVCIIDHKTIVIG-SSNWSIRGLNANKEEVLIIINPLTESQKQDLNTWW
TC_0435      244 ILALDEAV-TRSVIVTVIID----SNKQYTLLEKLSL---TKINIRVGTSS--SNCQVCIVDHTTAIIIGCSSNRPTRRQNANQEAVALIISPLTECQKQDLCTWW
TC_0436      276 IKALDEAASQRSVLVTIIID----SITKQTLGLTKALN---SKIRVRAGTLASC---IHCKVCIIIDHKTIVIG-SANWSNRGLNNTKEDLLIISPLTEGQKQDLCTWW
TC_0440      296 LKSL-QAAAARGVKVEVAVD---TRESEQTQRTLERLQ---LSLPLRVRKPGAPR---VHVKMCYIDNKILICG-SANWSSAGLTRNREDFVIRGLTETQKQSLSEIW
TC_0433      310 LQSI-QAAAARGVKVQIAID---TRESKQTQRTLERLQ---LSLPLRVRKPGSPP---LHVKMCIDGKTLIFG-SANWSLVGLARNVEDLFIVRNLTALQYQSLSEIW
TC_0432      173 LQAL-QAAAARGVKVEVAVD---TRESKQTQRTLERLQ---LSLPLRVRKPGSPP---LHVKMCIDNKILIIIG-SSNWSFVGLARNTEDLFIVRGLTETQKQSLSEIW
TC_0357      228 IKELNNA-QNRGVDVVLVD---KGFKSLTLKIQQLIQLIHPSLSIYEKV-TPYQ----LHKKFGIFDKKTLITG-SVNWSEKGFLLNTEDMIVIENTERQKQCKIAIW
NucT (H. pylori)

TC_0447      346 RYLCDNS-AI-----LPED-----
TC_0434      362 HFLCENSTAI-----TYEEAENA--DSSSEQELS-----
TC_0435      341 RSLCDSSTAI-----TYEEAENA--DSSSE-----
TC_0436      374 HFLCENSTVL-----A--EETKTPKILD-----
TC_0440      394 QSVEEKTEPLTAQSLKRPREEEDDPGEGTSSGISSAGASAKKAKTQ
TC_0433      408 KAVEENTQP----LKRSREEEEDPLEGTSSTQEDLSPPTKKARTQ

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**Fig. S2.** Sequence alignment of the outer membrane-associated nuclease NucT of *Helicobacter pylori* G27 (1) with phosphatidyl choline-hydrolysing phospholipase D proteins of *C. muridarum*. Alignment generated using CLUSTAL(2). Blue highlighting indicates amino acids conserved in  $\geq 60\%$  of aligned proteins, red stars denote functional residues important for NucT function (3).

	10	20	30	40	50	60	70	80	90	100	110	120	130	140	150	160	170	180	
CM001	MKKLLKSVLAF	AVLGSASSL	HALPVGNPAE	PSLMIDGIL	WEGFGGDP	CDPCTTWCDAI	SLRLGGY	GDFVDRVLKT	DVNVKQFEMGA	AAPTGDADLT	TAPTASREN	PAYGKHMQDA	EMFTNAAYMALNI	WDRFDV	FCTLGATS	GYLKGNSAA	AFNLVGLFGR	DETA	VAADDIPNVLSQAVV
CMUT3-5	MKKLLKSVLAF	AVLGSASSL	HALPVGNPAE	PSLMIDGIL	WEGFGGDP	CDPCTTWCDAI	SLRLGGY	GDFVDRVLKT	DVNVKQFEMGA	AAPTGDADLT	TAPTASREN	PAYGKHMQDA	EMFTNAAYMALNI	WDRFDV	FCTLGATS	GYLKGNSAA	AFNLVGLFGR	DETA	VAADDIPNVLSQAVV
G0.1.1	MKKLLKSVLAF	AVLGSASSL	HALPVGNPAE	PSLMIDGIL	WEGFGGDP	CDPCTTWCDAI	SLRLGGY	GDFVDRVLKT	DVNVKQFEMGA	AAPTGDADLT	TAPTASREN	PAYGKHMQDA	EMFTNAAYMALNI	WDRFDV	FCTLGATS	GYLKGNSAA	AFNLVGLFGR	DETA	VAADDIPNVLSQAVV
G28.38.1	MKKLLKSVLAF	AVLGSASSL	HALPVGNPAE	PSLMIDGIL	WEGFGGDP	CDPCTTWCDAI	SLRLGGY	GDFVDRVLKT	DVNVKQFEMGA	AAPTGDADLT	TAPTASREN	PAYGKHMQDA	EMFTNAAYMALNI	WDRFDV	FCTLGATS	GYLKGNSAA	AFNLVGLFGR	DETA	VAADDIPNVLSQAVV
Nigg3-28	MKKLLKSVLAF	AVLGSASSL	HALPVGNPAE	PSLMIDGIL	WEGFGGDP	CDPCTTWCDAI	SLRLGGY	GDFVDRVLKT	DVNVKQFEMGA	AAPTGDADLT	TAPTASREN	PAYGKHMQDA	EMFTNAAYMALNI	WDRFDV	FCTLGATS	GYLKGNSAA	AFNLVGLFGR	DETA	VAADDIPNVLSQAVV
CM006	MKKLLKSVLAF	AVLGSASSL	HALPVGNPAE	PSLMIDGIL	WEGFGGDP	CDPCTTWCDAI	SLRLGGY	GDFVDRVLKT	DVNVKQFEMGA	AAPTGDADLT	TAPTASREN	PAYGKHMQDA	EMFTNAAYMALNI	WDRFDV	FCTLGATS	GYLKGNSAA	AFNLVGLFGR	DETA	VAADDIPNVLSQAVV
Var003	MKKLLKSVLAF	AVLGSASSL	HALPVGNPAE	PSLMIDGIL	WEGFGGDP	CDPCTTWCDAI	SLRLGGY	GDFVDRVLKT	DVNVKQFEMGA	AAPTGDADLT	TAPTASREN	PAYGKHMQDA	EMFTNAAYMALNI	WDRFDV	FCTLGATS	GYLKGNSAA	AFNLVGLFGR	DETA	VAADDIPNVLSQAVV
Var004	MKKLLKSVLAF	AVLGSASSL	HALPVGNPAE	PSLMIDGIL	WEGFGGDP	CDPCTTWCDAI	SLRLGGY	GDFVDRVLKT	DVNVKQFEMGA	AAPTGDADLT	TAPTASREN	PAYGKHMQDA	EMFTNAAYMALNI	WDRFDV	FCTLGATS	GYLKGNSAA	AFNLVGLFGR	DETA	VAADDIPNVLSQAVV
Nigg	MKKLLKSVLAF	AVLGSASSL	HALPVGNPAE	PSLMIDGIL	WEGFGGDP	CDPCTTWCDAI	SLRLGGY	GDFVDRVLKT	DVNVKQFEMGA	AAPTGDADLT	TAPTASREN	PAYGKHMQDA	EMFTNAAYMALNI	WDRFDV	FCTLGATS	GYLKGNSAA	AFNLVGLFGR	DETA	VAADDIPNVLSQAVV
Nigg2MCR	MKKLLKSVLAF	AVLGSASSL	HALPVGNPAE	PSLMIDGIL	WEGFGGDP	CDPCTTWCDAI	SLRLGGY	GDFVDRVLKT	DVNVKQFEMGA	AAPTGDADLT	TAPTASREN	PAYGKHMQDA	EMFTNAAYMALNI	WDRFDV	FCTLGATS	GYLKGNSAA	AFNLVGLFGR	DETA	VAADDIPNVLSQAVV
Var001	MKKLLKSVLAF	AVLGSASSL	HALPVGNPAE	PSLMIDGIL	WEGFGGDP	CDPCTTWCDAI	SLRLGGY	GDFVDRVLKT	DVNVKQFEMGA	AAPTGDADLT	TAPTASREN	PAYGKHMQDA	EMFTNAAYMALNI	WDRFDV	FCTLGATS	GYLKGNSAA	AFNLVGLFGR	DETA	VAADDIPNVLSQAVV
Var001.1	MKKLLKSVLAF	AVLGSASSL	HALPVGNPAE	PSLMIDGIL	WEGFGGDP	CDPCTTWCDAI	SLRLGGY	GDFVDRVLKT	DVNVKQFEMGA	AAPTGDADLT	TAPTASREN	PAYGKHMQDA	EMFTNAAYMALNI	WDRFDV	FCTLGATS	GYLKGNSAA	AFNLVGLFGR	DETA	VAADDIPNVLSQAVV

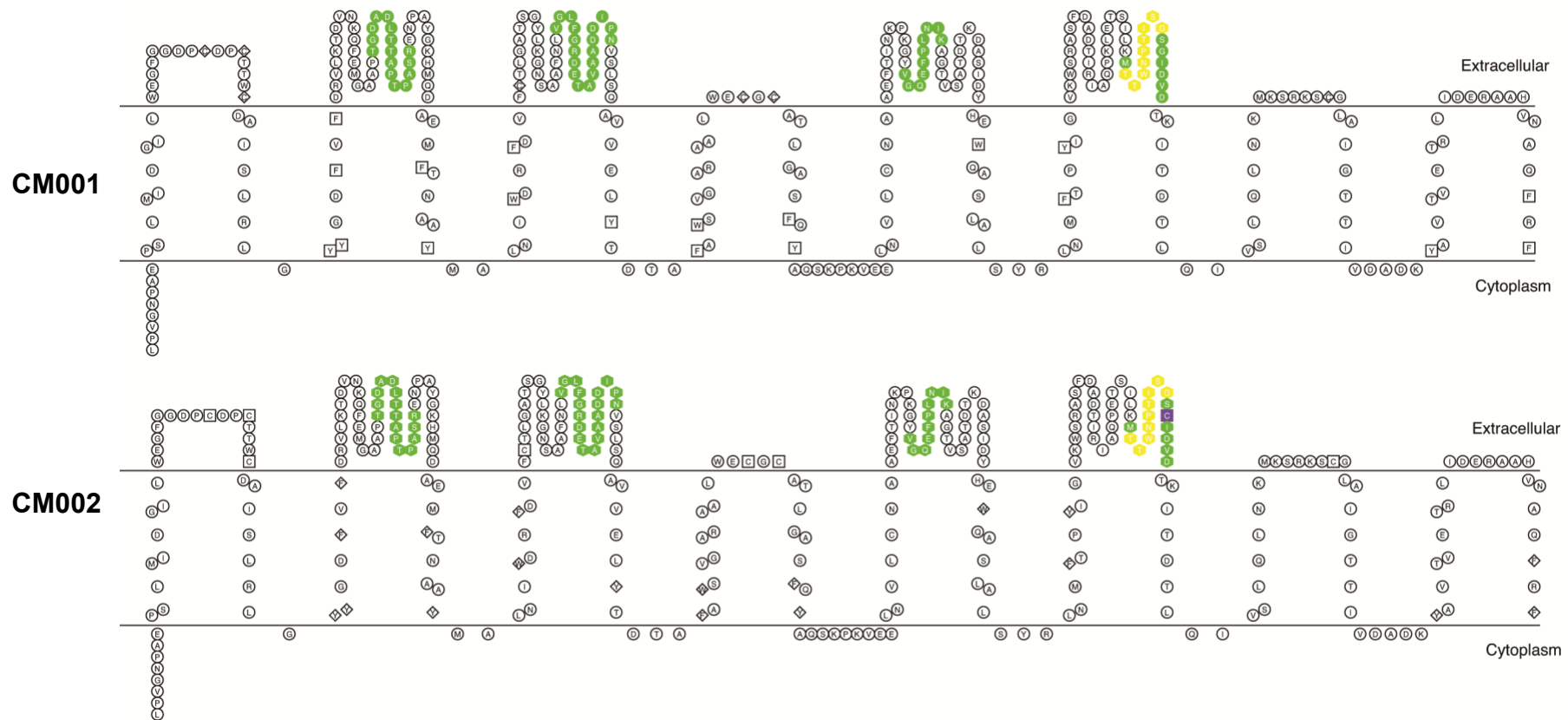
  

	190	200	210	220	230	240	250	260	270	280	290	300	310	320	330	340	350	360														
CM001	ELYTDTAF	AVS	VGARAAL	WECGCATL	GFQYAQ	SKPKVEE	LNVL	CNAAEFT	INKPKGY	VGGQEF	PLNIKAGT	VSATD	TKDAS	IDYHEW	QASLALS	YRLNMF	TPYIGV	KWSRAS	FDADT	IRIAQPK	LETSIL	KMT	TWNPT	ISGSG	IDVDT	KITD	TLQIV	SLQL	LNKMK	SRKSC	GLAIG	TITIV
CMUT3-5	ELYTDTAF	AVS	VGARAAL	WECGCATL	GFQYAQ	SKPKVEE	LNVL	CNAAEFT	INKPKGY	VGGQEF	PLNIKAGT	VSATD	TKDAS	IDYHEW	QASLALS	YRLNMF	TPYIGV	KWSRAS	FDADT	IRIAQPK	LETSIL	KMT	TWNPT	ISGSG	IDVDT	KITD	TLQIV	SLQL	LNKMK	SRKSC	GLAIG	TITIV
G0.1.1	ELYTDTAF	AVS	VGARAAL	WECGCATL	GFQYAQ	SKPKVEE	LNVL	CNAAEFT	INKPKGY	VGGQEF	PLNIKAGT	VSATD	TKDAS	IDYHEW	QASLALS	YRLNMF	TPYIGV	KWSRAS	FDADT	IRIAQPK	LETSIL	KMT	TWNPT	ISGSG	IDVDT	KITD	TLQIV	SLQL	LNKMK	SRKSC	GLAIG	TITIV
G28.38.1	ELYTDTAF	AVS	VGARAAL	WECGCATL	GFQYAQ	SKPKVEE	LNVL	CNAAEFT	INKPKGY	VGGQEF	PLNIKAGT	VSATD	TKDAS	IDYHEW	QASLALS	YRLNMF	TPYIGV	KWSRAS	FDADT	IRIAQPK	LETSIL	KMT	TWNPT	ISGSG	IDVDT	KITD	TLQIV	SLQL	LNKMK	SRKSC	GLAIG	TITIV
Nigg3-28	ELYTDTAF	AVS	VGARAAL	WECGCATL	GFQYAQ	SKPKVEE	LNVL	CNAAEFT	INKPKGY	VGGQEF	PLNIKAGT	VSATD	TKDAS	IDYHEW	QASLALS	YRLNMF	TPYIGV	KWSRAS	FDADT	IRIAQPK	LETSIL	KMT	TWNPT	ISGSG	IDVDT	KITD	TLQIV	SLQL	LNKMK	SRKSC	GLAIG	TITIV
CM006	ELYTDTAF	AVS	VGARAAL	WECGCATL	GFQYAQ	SKPKVEE	LNVL	CNAAEFT	INKPKGY	VGGQEF	PLNIKAGT	VSATD	TKDAS	IDYHEW	QASLALS	YRLNMF	TPYIGV	KWSRAS	FDADT	IRIAQ	PKLETS	ILKMT	TWNPT	ISGSG	IDVDT	KITD	TLQIV	SLQL	LNKMK	SRKSC	GLAIG	TITIV
Var003	ELYTDTAF	AVS	VGARAAL	WECGCATL	GFQYAQ	SKPKVEE	LNVL	CNAAEFT	INKPKGY	VGGQEF	PLNIKAGT	VSATD	TKDAS	IDYHEW	QASLALS	YRLNMF	TPYIGV	KWSRAS	FDADT	IRIAQ	PKLETS	ILKMT	TWNPT	ISGSG	IDVDT	KITD	TLQIV	SLQL	LNKMK	SRKSC	GLAIG	TITIV
Var004	ELYTDTAF	AVS	VGARAAL	WECGCATL	GFQYAQ	SKPKVEE	LNVL	CNAAEFT	INKPKGY	VGGQEF	PLNIKAGT	VSATD	TKDAS	IDYHEW	QASLALS	YRLNMF	TPYIGV	KWSRAS	FDADT	IRIAQ	PKLETS	ILKMT	TWNPT	ISGSG	IDVDT	KITD	TLQIV	SLQL	LNKMK	SRKSC	GLAIG	TITIV
Nigg	ELYTDTAF	AVS	VGARAAL	WECGCATL	GFQYAQ	SKPKVEE	LNVL	CNAAEFT	INKPKGY	VGGQEF	PLNIKAGT	VSATD	TKDAS	IDYHEW	QASLALS	YRLNMF	TPYIGV	KWSRAS	FDADT	IRIAQ	PKLETS	ILKMT	TWNPT	ISGSG	IDVDT	KITD	TLQIV	SLQL	LNKMK	SRKSC	GLAIG	TITIV
Nigg2MCR	ELYTDTAF	AVS	VGARAAL	WECGCATL	GFQYAQ	SKPKVEE	LNVL	CNAAEFT	INKPKGY	VGGQEF	PLNIKAGT	VSATD	TKDAS	IDYHEW	QASLALS	YRLNMF	TPYIGV	KWSRAS	FDADT	IRIAQ	PKLETS	ILKMT	TWNPT	ISGSG	IDVDT	KITD	TLQIV	SLQL	LNKMK	SRKSC	GLAIG	TITIV
Weiss	ELYTDTAF	AVS	VGARAAL	WECGCATL	GFQYAQ	SKPKVEE	LNVL	CNAAEFT	INKPKGY	VGGQEF	PLNIKAGT	VSATD	TKDAS	IDYHEW	QASLALS	YRLNMF	TPYIGV	KWSRAS	FDADT	IRIAQ	PKLETS	ILKMT	TWNPT	ISGSG	IDVDT	KITD	TLQIV	SLQL	LNKMK	SRKSC	GLAIG	TITIV
Var001	ELYTDTAF	AVS	VGARAAL	WECGCATL	GFQYAQ	SKPKVEE	LNVL	CNAAEFT	INKPKGY	VGGQEF	PLNIKAGT	VSATD	TKDAS	IDYHEW	QASLALS	YRLNMF	TPYIGV	KWSRAS	FDADT	IRIAQ	PKLETS	ILKMT	TWNPT	ISGSG	IDVDT	KITD	TLQIV	SLQL	LNKMK	SRKSC	GLAIG	TITIV
Var001.1	ELYTDTAF	AVS	VGARAAL	WECGCATL	GFQYAQ	SKPKVEE	LNVL	CNAAEFT	INKPKGY	VGGQEF	PLNIKAGT	VSATD	TKDAS	IDYHEW	QASLALS	YRLNMF	TPYIGV	KWSRAS	FDADT	IRIAQ	PKLETS	ILKMT	TWNPT	ISGSG	IDVDT	KITD	TLQIV	SLQL	LNKMK	SRKSC	GLAIG	TITIV

	370	380		
CM001	DADKYAVT	VETRLID	ERAHHVNAQ	FRF
CMUT3-5	DADKYAVT	VETRLID	ERAHHVNAQ	FRF
G0.1.1	DADKYAVT	VETRLID	ERAHHVNAQ	FRF
G28.38.1	DADKYAVT	VETRLID	ERAHHVNAQ	FRF
Nigg3-28	DADKYAVT	VETRLID	ERAHHVNAQ	FRF
CM006	DADKYAVT	VETRLID	ERAHHVNAQ	FRF
Var003	DADKYAVT	VETRLID	ERAHHVNAQ	FRF
Var004	DADKYAVT	VETRLID	ERAHHVNAQ	FRF
Nigg	DADKYAVT	VETRLID	ERAHHVNAQ	FRF
Nigg2MCR	DADKYAVT	VETRLID	ERAHHVNAQ	FRF
Weiss	DADKYAVT	VETRLID	ERAHHVNAQ	FRF
Var001	DADKYAVT	VETRLID	ERAHHVNAQ	FRF
Var001.1	DADKYAVT	VETRLID	ERAHHVNAQ	FRF

**Fig. S3.** Sequence alignment of the predicted *C. muridarum* CM001 and CM006 MOMP porin with other published sequences. Sequences were derived from the following strains; CMUT3-5 (4), G0.1.1, G28.38.1(5), Nigg3-28 (6), Var001, Var001.1, Var003, Var004 (7), Nigg (8), Nigg2MCR(9), Weiss (10). Amino acid ambiguities, substitutions or deletions are indicated in red.



**Fig. S4.** Cartoon depicting the effect of MOMP sequence differences on 2-dimensional protein conformation for *C. muridarum* CM001 (top) and CM002 (bottom). CM002 serves as a representative example of all sequenced clonal isolates derived from *C. muridarum* Nigg. The models were prepared using the Topo2 program (Johns S.J., TOPO2, Transmembrane protein display software, <http://www.sacs.ucsf.edu/TOPO2/>) with data for predicted extracellular and cytoplasmic loops, and transmembrane spanning regions of *C. muridarum* MOMP (11). The variable domains (VD1-4) are indicated in green, the conserved amino acids within VD4 are indicated in yellow and the glycine to cysteine substitution in VD4 present in CM002 is indicated in purple.

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