ECL_VgrG2 Eh49162 NIC22 Eh_e2355 Eh_e1105 ECL_M-7-X3	1 MLNR I T VQ LP V EGLLFWKLSGR EAMS ESFALT LT LLGT DAR I DR SK LLGQP VT VT I P TQNLLTP RY I NGK VT RV/ 1 MLNR I T VQ LP V EGLLFWKLSGR EAMS ESFALT LT LLGT DAR I DR SK LLGQP VT VT I P TQNLLTP RY I NGK VT RV/ 1 MLNR I T VQ LP V EGLLFWKLSGR EAMS ESFALT LT LLGT DAR I DR SK LLGQP VT VT I P TQNLTP RY I NGK VT RV/ 1 MLNR I T VQ LP V EGLLFWKLSGR EAMS ESFALT LT LLGT DAR I DR SK LLGQP VT VT I P TQNLTP RY I NGK VT RV/ 1 MLNR I T VQ LP V EGLLFWKLSGR EAMS ESFALT LT LLGT DAR I DR SK LLGQP VT VT I P TQNLTP RY I NGK VT RV/ 1 MLNR I T VQ LP V EGLLFWKLSGR EAMS ESFALT LT LLGT DAR I DR SK LLGQP VT VT I P TQNLTP RY I NGK VT RV/ 1 MLNR I T VQ LP V EGLLFWKLSGR EAMS ESFALT LT LLGT DAR I DR SK LLGQP VT VT I P TQNLTP RY I NGK VT RV/	75 75 75 75
ECL_VgrG2 Eh49162 NIC22 Eh_e2355 Eh_e1105 ECL_M-7-X3	76 V SAVELTGTRYAVYQLTVEPDLWPMKRDRNLR I FQGQTVPQIVKTLLGEHQVNVEDKLTGSYRVWDYCVQYQES 76 V SAVELTGTRYAVYQLTVEPDLWPMKRDRNLR I FQGQTVPQIVKTLLGEHQVNVEDKLTGSYRVWDYCVQYQES 76 V SAVELTGTRYAVYQLTVEPDLWPMKRDRNLR I FQGQTVPQIVKTLLGEHQVNEDKLTGSYRVWDYCVQYQES 76 V SAVELTGTRYAVYQLTVEPDLWPMKRDRNLR I FQGQTVPQIVKTLLGEHQVNLEDKLTGSYRVWDYCVQYQES 76 V SAVELTGTRYAVYQLTVEPDLWPMKRDRNLR I FQGQTVPQIVKTLLGEHQVNLEDKLTGSYRVWDYCVQYQES 76 V SAVELTGTRYAVYQLTVEPDLWPMKRDRNLR I FQGQTVPQIVKTLLGEHQVNLEDKLTGSYRVWDYCVQYQES	150 150 150 150
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ECL_VgrG2 Eh49162 NIC22 Eh_e2355 Eh_e1105 ECL_M-7-X3	226 LDDY DFRKP NAWL FQAQQNP A SP KP G S I DVY DWP GR F V ET GHA E FY AR I RQ ERWQ V E HQQ I QAT AT AAG I AP GH 226 LDDY D FR KP NAWL FQAQQNP A SP KP G S I DVY DWP GR F V ET GHA E FY AR I RQ ERWQ V E HQQ I QAT AT AAG I AP GH 226 LDDY D FR KP NAWL FQAQQNP A SP KP G S I DVY DWP GR F V ET GHA E FY AR I RQ ERWQ V E HQQ I QAT AT AAG I AP GH 226 LDDY D FR KP NAWL FQAQQNP A SP KP G S I DVY DWP GR F V ET GHA E FY AR I RQ ERWQ V E HQQ I QAT AT AAG I AP GH 226 LDDY D FR KP NAWL FQAQQNP A SP KP G S I DVY DWP GR F V ET GHA E FY AR I RQ ERWQ V E HQQ I QAT AT AAG I AP GH 226 LDDY D FR KP NAWL FQAQQNP A SP KP G S I DVY DWP GR F V ET GHA E FY AR I RQ ERWQ V E HQQ I QAT AT AAG I AP GH 226 LDDY D FR KP NAWL FQAQQNP A SP KP G S I DVY DWP GR F V ET GHA E FY AR I RQ ERWQ V E HQQ I QAT AT AAG I AP GH	300 300 300 300
ECL_VgrG2 Eh49162 NIC22 Eh_e2355 Eh_e1105 ECL_M-7-X3	301 FT LTNAP FF S DNG DY L VTAAGYH L E ENRYAS GEGET I HR I D FT V I PASVS FRPAQ STAWP RTY GPQTAK V V GPQ 301 FT LTNAP FF S DNG EY L VTAAGYH FE ENRYAS GEGET I HRTD FT V I PASVS Y RPAQ STAWP RTY GPQTAK V V GPQ 301 FT LTNAP FF S DNG EY L VTAAGYH FE ENRYAS GEGET I HRTD FT VI PASVS Y RPAQ STAWP RTY GPQTAK V V GPQ 301 FT LTNAP FF S DNG EY L VTAAGYH FE ENRYAS GEGET I HRTD FT VI PASVS Y RPAQ STAWP RTY GPQTAK V V GPQ 301 FT LTNAP FF S DNG EY L VTAAGYH FE ENRYAS GEGET I HRTD FT VI PASVS Y RPAQ STAWP RTY GPQTAK V V GPQ 301 FT LTNAP FF S DNG EY L VTAAGYH FE ENRYAS GEGET I HRTD FT VI PASVS Y RPAQ STAWP RTY GPQTAK V V GPQ 301 FT LTNAP FF S DNG EY L VTAAGYH FE ENRYAS GEGET I HRTD FT VI PASVS Y RPAQ STAWP RTY GPQTAK V V GPQ	375 375 375 375 375
ECL_VgrG2 Eh49162 NIC22 Eh_e2355 Eh_e1105 ECL_M-7-X3	376 ES I WT DK Y GR V K V K F HWDR L AK GDDT S SCWVR V S SAWAGQGY GGVQ I PRVGDE VVVDF I NGDP DR PI I T GR VY N 376 ES I WT DK Y GR V K V K F HWDR L AK GDDT S SCWVR V S SAWAGQGY GGVQ I PRVGDE VVVDF I NGDP DR PI I T GR VY N 376 ES I WT DK Y GR V K V K F HWDR L AK GDDT S SCWVR V S SAWAGQGY GGVQ I PRVGDE VVVDF I NGDP DR PI I T GR VY N 376 ES I WT DK Y GR V K V K F HWDR L AK GDDT S SCWVR V S SAWAGQGY GGVQ I PRVGDE VVVDF I NGDP DR PI I T GR VY N 376 ES I WT DK Y GR V K V K F HWDR L AK GDDT S SCWVR V S SAWAGQGY GGVQ I PRVGDE VVVDF I NGDP DR PI I T GR VY N 376 ES I WT DK Y GR V K V K F HWDR L AK GDDT S SCWVR V S SAWAGQGY GGVQ I PRVGDE VVVDF I NGDP DR PI I T GR VY N 376 ES I WT DK Y GR V K V K F HWDR L AK GDDT S SCWVR V S SAWAGQGY GGVQ I PRVGDE VVVDF I NGDP DR PI I T GR VY N	450 450 450 450
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ECL_VgrG2 Eh49162 NIC22 Eh_e2355 Eh_e1105 ECL_M-7-X3	601 TGGKLHLNT SGAK P GTTAP GAGHKGD I DAAVQAK F TTKGD	670 661 674 670
ECL_VgrG2 Eh49162 NIC22 Eh_e2355 Eh_e1105 ECL_M-7-X3	671 SY VYQNNSYNSDVMP F SEDVVKE I NK SP TLQTQLKDLKDKGWA I QP GAAGGGSYADTNNKL - I VMDP E F 662SNVP S SGYGK DVD SLVDK SP TMKND I ATLKKRGWT F E EG E AGKGT F ANRQTRV-I TVDKN-I 675 E DK F SK I S	721 719 738
ECL_VgrG2 Eh49162 NIC22 Eh_e2355 Eh_e1105 ECL_M-7-X3	739 M EDTATTVQTLAHEAGHATYP VAVDSSSKENFINSQ	5 774 Q 788 Q 791
ECL_VgrG2 Eh49162 NIC22 Eh_e2355 Eh_e1105 ECL_M-7-X3	792 I L S NGG I D I D I AG S A E N LKAY N S AY D KM Y N G E L S R I D A	827 828 797
ECL_VgrG2 Eh49162 NIC22 Eh_e2355 Eh_e1105 ECL_M-7-X3	845 GT N L N Y ND Y Y G G F Y G K	860 846 853 901

**Figure S1. Evolved VgrG proteins from** *Enterobacter* **species.** VgrG2 from *Enterobacter cloacae* ATCC 13047 was aligned with homologs from *Enterobacter hormaechei* ATCC 49162 (NCBI Refseq: WP\_006810934.1), *Enterobacter* sp. NIC22-4 (WP\_221813114.1), *Enterobacter hormaechei* e2355 (WP\_058700648.1), *Enterobacter hormaechei* e1105 (CZV11788.1), and *Enterobacter cloacae* M-7-X3 (WP\_165464354.1) using Clustal Omega and rendered with Jalview 2.11.2.6. Residues are colored according to sequence identity. The C-terminal extension domains from ATCC 49162, NIC22-4 and e1105 are predicted to fold into metallopeptidase domains by AlphaFold2. The effector domain from e2355 is homologous to *N*-acetylmuramidases (lysozyme), and the domain from M-7-X3 is a predicted LytD β-*N*-acetylglucosaminidase.