Agrobacterium tumefaciens

motA fliM fliN fliG flhB At0565 At0566 flaD At0568 motB At0570 motD sltF At0573 flgE flgK flgL flaF flbT flgD fliQ flhA fliR At0583 flgJ

Agrobacterium sp.

motA fliM fliN fliG flhB Agro3968 Agro3969 Agro3971 flaD Agro3972 motB motC fliK sltF Agro3979 flgE flgK flgL flaF flbT flgD fliQ flhA fliR Agro4006 flgJ

Agrobacterium vitis

fliI Avi0743 fliH fliL flaA flaA flaA flaA flaB Avi0755 motB Avi0759 Avi0760 sltF Avi0763 flgE flgK flgL flaF flbT flgD fliQ flhA fliR Avi0774 flgJ

Bartonella clarridgeiae

fliF T420_1170 motB motC fliK sltF T420_1165 flgE flgK flgL flaF flbT flgD fliQ // flhA fliR T420_0428 flgJ T420_0430 T420_0431 T420_0432 flhB flgG fliG fliN T420_0436

Bartonella bacilliformis

flgJ BARBACKI156 BARBACKI157 BARBACKI154 flhB fliG fliG fliY BARBACKI150 fliM motB // fliF BARBACKI128 motB motC BARBACKI128 sltF BARBACKI123 flgE flgK flgL flaF flbT flgD fliQ

Bradyrhizobium japonicum

flgI Bll6870 flgH fliL fliP fliC2 fliC2 fliF Bll6863 motB motC motD sltF flgE flgK flgL fliC3 flbT flgD fliQ flhA fliR flgJ

Bradyrhizobium sp.

motE flgH fliL fliP fliC1 fliC2 fliC3 fliF SE23_14100 motB motC motD sltF flgE flgK flgL flaF flbT flaF flbT flgD fliQ flhA fliR SE_14240 flgJ

Brucella melitensis

fliC BMEA_B1139 motA motC fliK sltF BMEA_1134 flaE flgK flgL flaF flbT flgD fliQ fliR BMEA_1124 flgJ

Brucella suis

fliF BRA1145 motB motD sltF BRA1140 flgE flgK flgL flaF flbT flgD fliQ flhA fliR BRA1130 flgJ

Brucella abortus

flaF fliF BruAb2_1082 motB motC motD sltF BruAb2_1077 flgE flgK flgL flaF flbT flgD fliQ fliR BruAb2_1067 flgJ

Sinorhizobium meliloti

flgI motE flgH fliL fliP flaA flaB flaC flaD Smc03041 motB motC motD sltF Smc03046 flgE flgK flgL flaF flbT flgD fliQ flhA fliR Smc03056 flgJ

Sinorhizobium medicae

motE flgH fliL fliP fliC1 fliC2 fliC3 flaC Smed_0269 Smed_0270 motB motC motD sltF Smed_0275 flgE flgK flgL flaF flbT flgD fliQ flhA fliR Smed_0285 flgJ

Rhizobium sp.

flgI NGR_c2800 flgH flgL fliP fliCc fliCB fliCA flaD NGR_c2880 motB motC motD sltF NGRc_2930 flgE flgK flgL flaF flbT flgD fliQ flhA fliR NGR_c3030 flgJ

Ensifer fredii

flgH fliL fliP fliC1 USDA257_3170 fliC2 fliC3 fliC4 USDA257_3210 USDA257_3220 motB motC motD sltF USDA257_3270 flgE flgK flgL flaF flbT flgD fliQ flhA fliR USDA257_3370 flgJ

Sinorhizobium fredii

flgI motE flgH fliL fliP fliC1 fliC2 fliC3 fliC4 SFHH103_330 motB motC motD sltF SFHH103_335 flgE flgK flgL flaF flbT flgD fliQ flhA fliR SFHH103_345 flgJ

Mesorhizobium loti

fliE flgG flgA flhB flgI Mlr_2920 flgH fliL flgD flaA1 flaA2 fliF Mrl_2930 motB motC motD sltF Mrl_2935 flgE flgK flgL flaF flbT flgD fliQ flgJ

Mesorhizobium ciceri

flgC fliE flgG flgA flgI Mesci_1952 flgH fliL fliP fliC1 fliC2 fliF Mesci_1944 motB motC fliK sltF Mesci_1939 flgE flgK flgL flaF flbT fliQ flgJ

Mesorhizobium opportunistum

flgC fliE flgG flgA flgI motE flgH fliL fliP fliC1 fliC2 fliF Mesop_2000 motB motC motD sltF Mesop_1995 flgE flgK flgL flaF flbT flgD fliQ flgJ

Methylobacterium radiotolerans

fliQ flhA fliR Mrad2831_1698 flgJ Mrad2831_1696 Mrad2831_1695 Mrad2831_1694 Mrad_2831_1693 flgB flgC fliE flgG flgA flgI Mrad2831_1686 flgH fliL fliP fliC fliF Mrad2831_1680 motB motC fliK sltF

Ochrobactrum anthropi

flgA flgI Oant_4206 flgH fliL fliP Oant_4202 fliC fliF Oant_4199 motB motC fliK sltF Oant_4194 flgE flgK flgL flaA flbT flgD fliQ flhA fliR Oant_4184 flgJ

Methylocella silvestris

flgG flgA flgI Msil_0030 flgH fliL fliP fliC fliF Msil_0036 motB motC Msil_0039 sltF Msil_0041 flgE flgK flgL flaF flbT flgD fliQ flhA fliR Msil_0051 flgJ

Polymorphum gilvum

flgA flgI motE flgH fliL fliP fliC fliF SL003B_3880 motB motC fliK sltF flgE flgK flgL flaF flbT fliQ SL003B3868 SL003B3867 SL003B3866 flhA fliR SL003B3863 flgJ

Pseudovibrio sp

flgA flgI motE flgH fliL fliP fliC1 fliC2 PSE_2614 fliF PSE_2616 motB motC motD sltF flgE flgK flgL flaF flbT flgD fliQ flhA fliR PSE_2630 flgJ

Rhizobium leguminosarum

fliP Rleg12_0329 Rleg12_0330 Rleg12_0331 fliC1 fliC2 fliC3 fliC4 Rleg12_0336 motB motC motD sltF Rleg12_0341 flgE flgK flgL flaF flbT flgD fliQ flhA fliR Rleg12_0351 flgJ

Rhizobium etli

RHE_CH664 flgH fliL fliP RHE_CH668 fliC1 fliC2 fliC3 fliC4 RHE_673 motB motC motD sltF RHE_678 flgE flgK flgL flaF flbT flgD fliQ flhA fliR RHE_688 flgJ

Agrobacterium radiobacter

fliL fliP Arad_0891 fliC1 fliC2 Arad_0897 Arad_0898 fliC3 fliC4 Arad_0901 motB motC motD sltF Arad_0907 flgE flgK flgL flaF flbT flgD fliQ flhA fliR Arad_0920 flgJ

Rhizobium tropici

flgH fliL fliP RTCIAT899_CH3330 fliC1 fliC2 RTCIAT899_CH3345 RTCIAT899_CH3350 fliC3 fliC4 RTCIAT899_CH3365 motB motC motD sltF RTCIAT899_CH3435 flgK flgL flaF flbT flgD fliQ flhA fliR RTCIAT899_CH3435 flgJ

Rhodomicrobium vannielii

fliC fliF Rvan_2570 motB motC fliK sltF Rvan_2565 flgF flgL fliI flgE flgK flgL flaF flbT flgD fliQ flhA fliR Rvan_2553 flgJ

Rhodopseudomonas palustris

flgG flgA flgI RPC_1100 flgH fliL fliP fliC1 fliF RPC_1106 motB motC motD sltF flgE flgK flgL flbT flaF flbT flgD fliQ flhA fliR RPC_1106 flgJ

Pannonibacter phragmitetus

flgG flgA flgI motE flgH fliL fliP fliC H163draft_01074 motB motC fliK sltF flgE flgK flgL flaF flbT flaF flbT flgD fliQ flhA fliR H163draft_01060 flgJ

Rhizobium phaseoli

motB motC motD sltF MYGdraft_0296 flgE flgK flgL flaF flbT flgD fliQ flhA fliR MYGdraft_02786 flgJ

Nitratireductor pacificus

flgA flgI NA2_19658 flgH fliL fliP fliC1 fliC2 fliF NA2_19623 motB motC motD sltF NA2_19598 flgE flgK flgL flaF flbT flgD fliQ flhA fliR H163draft_01060 flgJ

Pseudaminobacter salicylatoxidans

flgA flgI motE flgH fliL fliP fliC fliF BN85draft_03750 motB motC motD sltF BN85draft_03745 flgE flgK flgL flaF flbT flgD fliQ BN85draft_03735 flhA fliR BN85draft_03733 flgJ

Nitratireductor indicus

flgA flgI NA8A_15211 flgH fliL fliP fliC1 fliC2 fliF NA8A_15246 motB motC fliK sltF NA8A_15271 flgE flgK flgL flaF flbT flgD fliQ flhA fliR NA8A_15321 flgJ

Agrobacterium rhizogenes

fliC1 fliC2 ArA4draft_54170 motB motC motD sltF ArA4draft_54220 flgE flgK flgL flaF flbT flgD fliQ flhA fliR ArA4draft_54220 flgJ

Phaeobacter gallaeciencis

fliL Gal_03634 motE motA Gal_03637 sltF flhA fliR flhB Gal_03642 fliL2 flgH flgA flgG flgF fliQ fliE flgC flgB Gal_03653 fliI flbT flaF fliC flgN flgJ

Phaeobacter inhibens

fliL Phain_03506 motE motA Phain_03509 sltF flhA fliR flhB Phain_03514 fliL2 flgH flgA flgG flgF fliQ fliE flgC flgB Phain_03514 fliI flbT flaF fliC flgN flgJ

Rhodospirillum centenum

flgI flgJ RC_0206 fliC flaF flbT RC1_0202 RC1_0201 RC1_0200 RC1_0199 fliI flgB flgC fliE fliQ sltF RC1_0193 flhA fliR flhB flaA RC1_0187 fliM fliL fliF fliH

Rhodobacter sphaeroides fla1

flgB flgC flgD flgE flgF flgG flgH flgI flgJ flgK flgL sltF fleQ fliD fliC rpoN2 rps6092 motF flhB flhB fliR fliQ fliP fliO fliN fliM fliL

Rhodobacter sphaeroides fla2

RSP_1306 RSP_1308 fliH2 fliF2 fliL2 RSP_1314 RSP_1315 motA2 RSP_1318 sltF2 flhA2 fliR2 flhB2 fliJ2 fliL2 flgH2 flgA2 flgG2 fliF2 fliQ2 fliE2 flgC2 flgB2 fliI2 flgN2 flgJ2

Ruegeria pomeroyi
SPO_0168 sltF flhA fliR flhB SPO_0173 SPO_0174 flgH flgA flgG flgF fliQ fliE flgC flgB fliI SPO_0184 SPO_0185 SPO_0186 flgD fliK flgJ SPO_03460 flaA flaF

Roseobacter denitrificans
sltF flhA fliR flhB RD1_0153 RD1_0154 RD1_0155 RD1_0156 RD1_0157 RD1_0158 flaF fliC RD1_0161 flgJ fliK flgD

Roseobacter litoralis
flgF flgG flgA flgH RLO149_c043460 RLO149_c043470 RLO149_c043480 RLO149_c043490 RLO149_c043500 RLO149_c043510 RLO149_c043520 RLO149_c043530 RLO149_c043550 sltF flhA fliR flhB RLO149_c043600 RLO149_c043610 RLO149_c043620 RLO149_c043630 flbT flaF fliC flgN flgJ

Rubellimicrobium thermophilum
motE motA Ruthe_02331 Ruthe_02330 sltF flhA fliR flhB Ruthe_02325 fliL flgH Ruthe_02322 flgA flgG flgF fliQ fliE flgC flgB fliI Ruthe_02313 flbT flaF fliC Ruthe_02309 flgJ

Catellibacterium nectariphilum
fliL G481draft_00006 G481draft_00007 motA G481draft_00009 sltF flhA fliR flhB G481draft_00014 G481draft_00015 flgH flgA flgG flgF fliQ fliE flgC flgB fliI G481draft_00029 flbT flaF fliC G481draft_00029 flgJ

Saccharibacter floricola
fliL fliM F457draft_1865 fliC F457draft_1867 flgK flgL flgF flgG flgA flgH F457draft_1874 motE F457draft_1876 fliF fliG F457draft_1879 fliN F457draft_1881 fliP flhF flhA F457draft_1885 sltF flgI flgJ

Fig.S1 Flagellar context of lytic transglycosylases. Graphic representation of the flagellar context of 46 α-proteobacteria for lytic transglycosylases and single domain scaffolding *flgJ* genes. Flagellar LT Family 1F and 1F' (SltF$_{Bs}$, *brucella suis* (637332230), SltF$_{Ba}$*Brucella abortus* (637647151), SltF$_{Bm}$, *Brucella melitensis* (643747691), SltF$_{Oa}$, *Ochrobactrum anthropi* (640836883)SltF$_{Pp}$, *Pannonibacter phragmitetus* (2521738048), SltF$_{Pg}$, *Polymorphum gilvum* (2512365150), SltF$_{Rv}$, *Rhodomicrobium vannielii* (649746019), SltF$_{Ms}$, *Methylocella silvestris* (643463373), SltF$_{Psp}$, *Pseudovibrio sp* (2511538202), SltF$_{Mr}$, *Methylobacterium radiotolerans* (641627382), SltF$_{Bsp}$*Bradyrhizobium sp.* (2514088350), SltF$_{Bj}$*Bradyrhizobium japonicum*, (637374448), SltF$_{Rp}$, *Rhodopseudomonas palustris* (637924221), SltF$_{Bb}$, *Bartonella bacilliformis* (639842294), SltF$_{Bc}$, *Bartonella clarridgeiae* (2548760840), SltF$_{Mo}$, *Mesorhizobium opportunistum* (2503200023), SltF$_{Ml}$, *Mesorhizobium loti* (637075604), SltF$_{Mc}$, *Mesorhizobium ciceri* (649871818), SltF$_{Ps}$, *Pseudaminobacter salicylatoxidans* (2551926637), SltF$_{Ni}$, *Nitratireductor indicus* (2520373292), SltF$_{Np}$, *Nitratireductor pacificus* (2520250528), SltF$_{Agrh}$, *Agrobacterium rhizogenes* (2505292958), SltF$_{Agr}$, *Agrobacterium radiobacter* (643645133), SltF$_{Rt}$, *Rhizobium tropici* (2524419140), SltF$_{Rhe}$, *Rhizobium etli* (640437712), SltF$_{R}$, *Rhizobium phaseoli* (2549960670), SltF$_{Rhl}$, *Rhizobium leguminosarum* (2510372010), SltF$_{Agv}$, *Agrobacterium vitis* (643650334), SltF$_{Agsp}$, *Agrobacterium Sp.* (650739020), SltF$_{Agt}$, *Agrobacterium tumefaciens* (639296061), SltF$_{Rhsp}$, *Rhizobium sp.* (643824500), SltF$_{Sf}$, *Sinorhizobium fredii* (2517638777), SltF$_{Ef}$, *Ensifer fredii* (2515008689), SltF$_{Sm}$, *Sinorhizobium meliloti* (637181464), SltF$_{Sme}$, *Sinorhizobium medicae* (640789209). SltF$_{Pi}$, *Phaeobacter inhibens* (2574253765), SltF$_{Pga}$, *Phaeobacter gallaeciensis* (2558539010), SltF$_{Sp}$, *Ruegeria pomeroyi* (637287661), SltF$_{Rt}$, *Rubellimicrobium thermophilum* (2521341176), SltF2$_{Rs}$, *Rhodobacter sphaeroides* WS8N, (651575303), SltF$_{Cn}$, *Catellibacterium nectariphilum* (2525538211), SltF$_{Rl}$, *Roseobacter litoralis* (2510237871), SltF$_{Rd}$, *Roseobacter denitrificans* (639633682), SltF$_{Sf}$, *Saccharibacter floricola* (2519014188), SltF$_{Rce}$, *Rhodospirillum centenum* (643411101), SltF1$_{Rs}$, *Rhodobacter sphaeroides* WS8N (651573991).
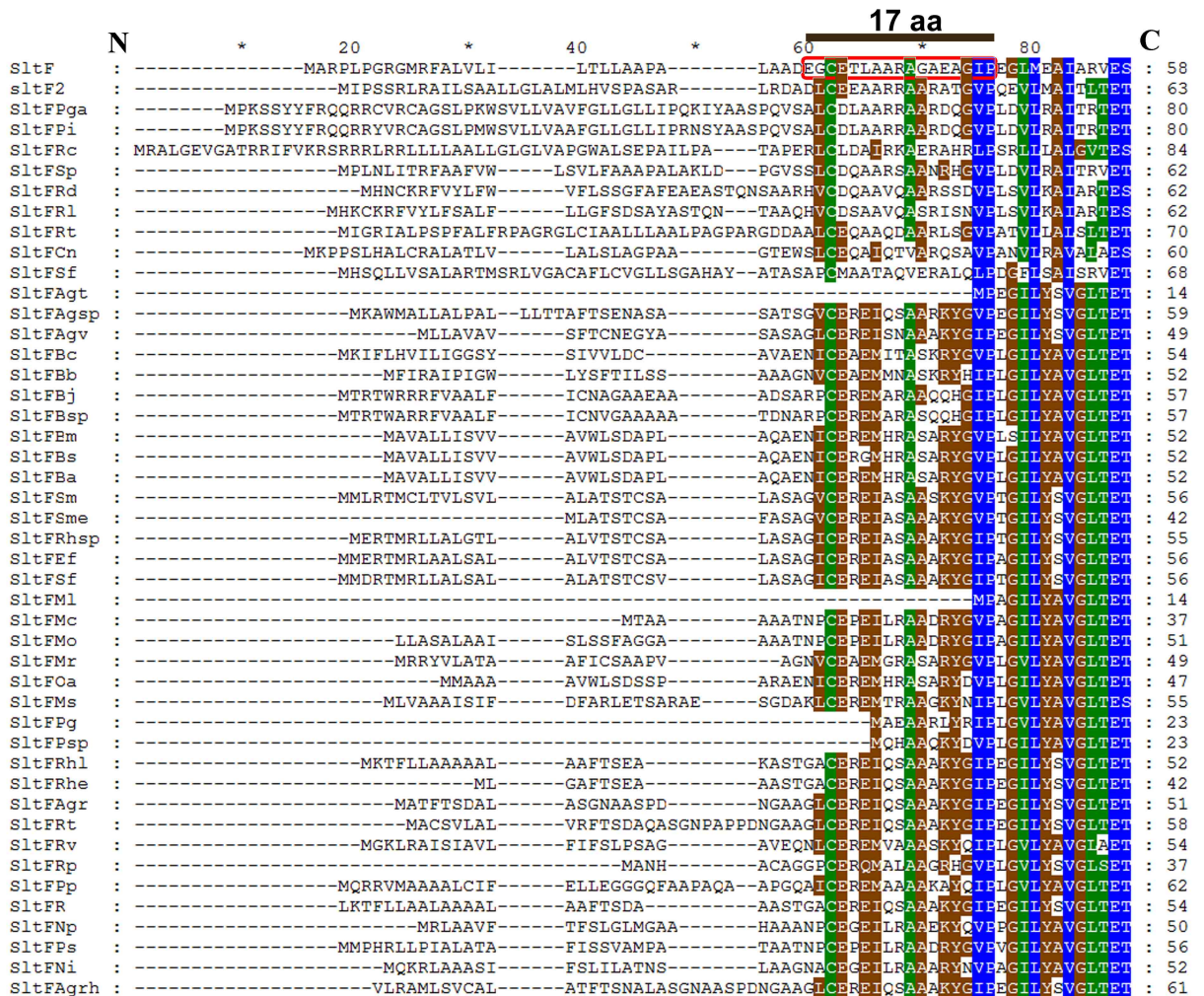
Fig.S2 N-terminus alignment flagellar lytic transglycosylases. SltF1$_{Rs}$Rhodobacter sphaeroides WS8N (651573991), SltF2$_{Rs}$Rhodobacter sphaeroides WS8N (651575303), SltF$_{Pi}$Phaeobacter inhibens (2574253765),SltF$_{Pga}$Phaeobacter gallaeciensis (2558539010), SltF$_{Sp}$Ruegeria pomeroyi (637287661),SltF$_{Rt}$Rubellimicrobium thermophilum (2521341176), SltF$_{Cn}$Catellibacterium nectariphilum (2525538211),SltF$_{Rl}$Roseobacter litoralis (2510237871), SltF$_{Rd}$Roseobacter denitrificans (639633682), SltF$_{Sf}$Saccharibacter floricola (2519014188), SltF$_{Rc}$Rhodospirillum centenum (643411101), SltF$_{Bs}$ brucella suis (637332230), SltF$_{Ba}$Brucella abortus (637647151),SltF$_{Bm}$Brucella melitensis (643747691), SltF$_{Oa}$Ochrobactrum anthropi (640836883),SltF$_{Pp}$Pannonibacter phragmitetus (2521738048), SltF$_{Pg}$Polymorphum gilvum (2512365150), SltF$_{Rs}$Rhodomicrobium vannielii (649746019), SltF$_{Ms}$Methylocella silvestris (643463373), SltF$_{Psp}$Pseudovibrio sp (2511538202),SltF$_{Mt}$Methylobacterium radiotolerans (641627382), SltF$_{Btp}$Bradyrhizobium sp. (2514088350), SltF$_{Bj}$Bradyrhizobium japonicum (637374448),SltF$_{Rp}$Rhodopseudomonas palustris,(637924221), SltF$_{Bb}$Bartonella bacilliformis (639842294),SltF$_{Bc}$Bartonella clarridgeiae (2548760840),SltF$_{Mo}$Mesorhizobium opportunistum (2503200023), SltF$_{Ml}$Mesorhizobium loti (637075604),SltF$_{Mc}$Mesorhizobium ciceri (649871818),SltF$_{Ps}$Pseudaminobacter salicylatoxidans (2551926637), SltF$_{Ni}$Nitratireductor indicus (2520373292),SltF$_{Np}$Nitratireductor pacificus (2520250528), SltF$_{Agrh}$Agrobacterium rhizogenes (2505292958),SltF$_{Agr}$Agrobacterium radiobacter (643645133), SltF$_{Rt}$Rhizobium tropici (2524419140), SltF$_{Rhe}$Rhizobium etli (640437712), SltF$_{Rhl}$Rhizobium phaseoli (2549960670), SltF$_{Rhl}$Rhizobium leguminosarum (2510372010), SltF$_{Agv}$Agrobacterium vitis (643650334),SltF$_{Agsp}$, Agrobacterium Sp. (650739020),SltF$_{Agt}$Agrobacterium tumefaciens (639296061), SltF$_{Rhsp}$Rhizobium sp.(643824500), SltF$_{Sf}$Sinorhizobium fredii (2517638777),SltF$_{Ef}$Ensifer fredii (2515008689),SltF$_{Sm}$Sinorhizobium meliloti (637181464), SltF$_{Sme}$Sinorhizobium medicae (640789209).Access numbers for each sequence are in accordance with IMG (Integrated Microbial Genomes : https://img.jgi.doe.gov/) or with GenBank. In black are shown the 17 residues of SltF1 from R. sphaeroides studied in this work.
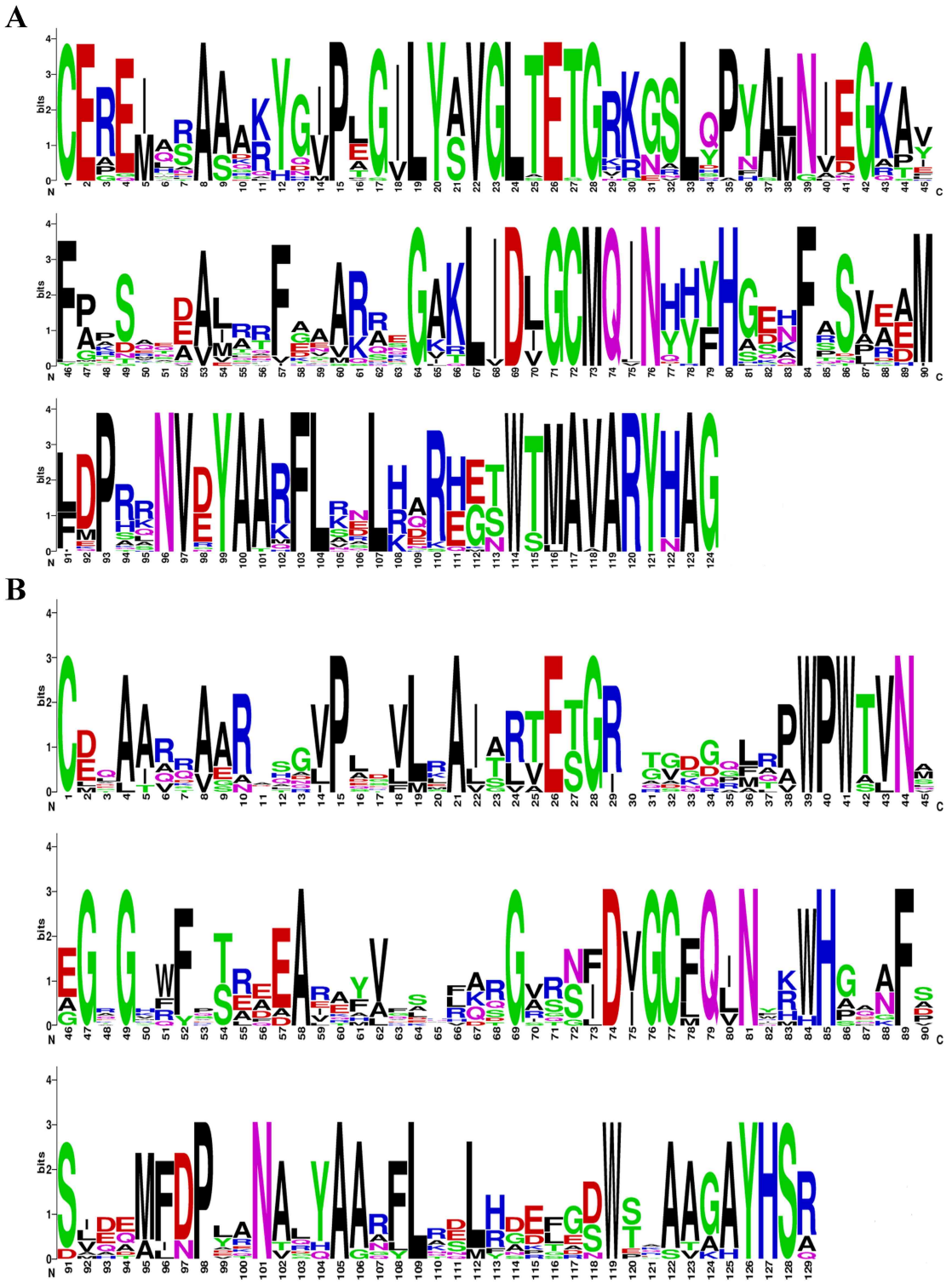
Fig.S3 Amino acid frequency analysis. Frequency analysis of residues within the catalytic site of SltFfrom subfamily 1F (Panel A) y 1F` (Panel B). This was achieved using WebLogo v2.8 (Crooks GE, Hon G, Chandonia JM, Brenner SE. 2004, WebLogo: A sequence logo generator, *Genome Research*, 14:1188-1190)
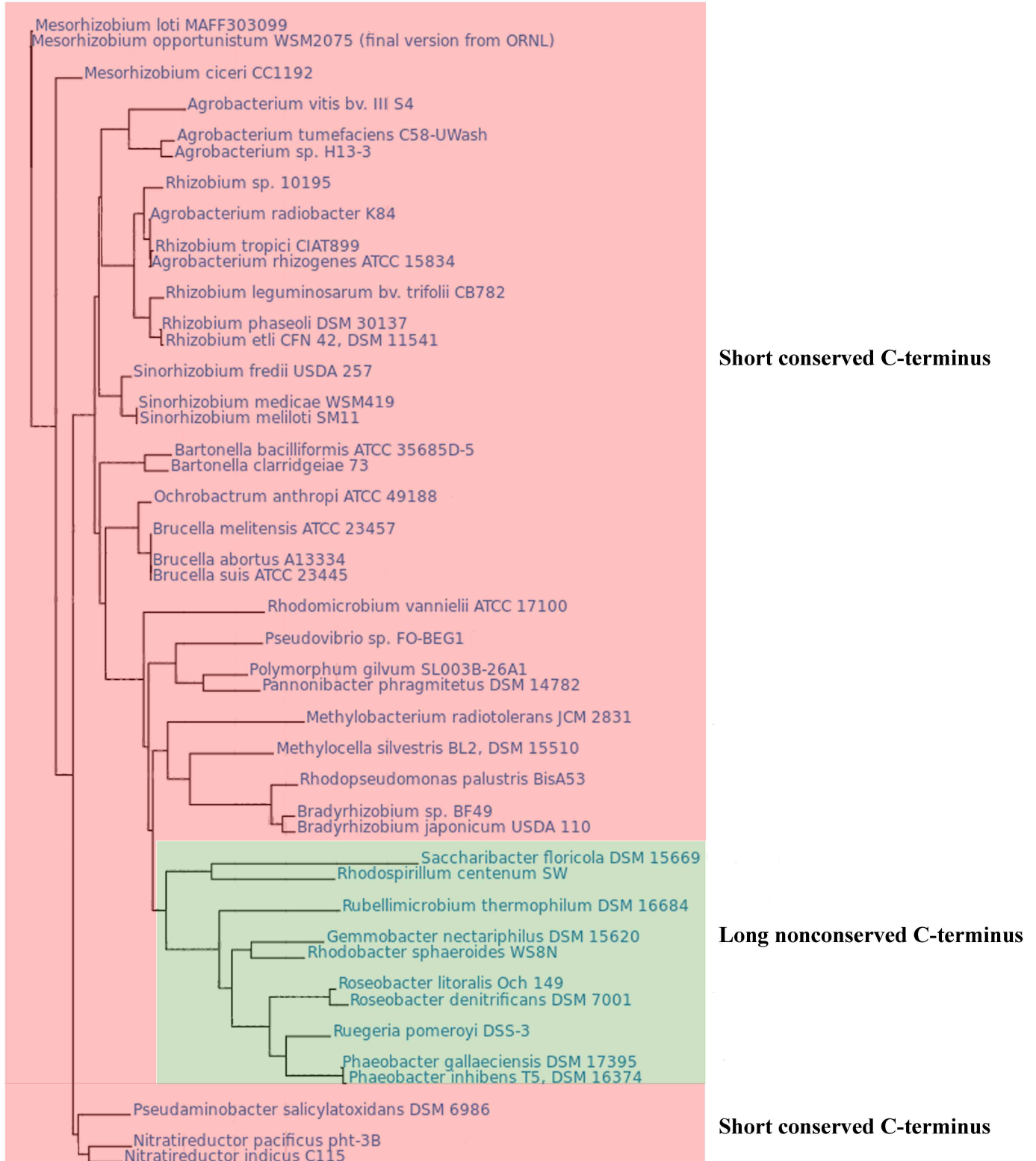
Fig.S4 Phylogenetic tree based on 16S ribosomal RNA sequences. The phylogenetic tree was built with sequences from 46 α-proteobacteria that posses single domain SltFs.

Table S1 Sec signal sequence prediction for Transglycosylases found in this work. Access numbers correspond to the data base from IMG. Signal sequence was predicted with SignalP V 4.1 Nielsen H. 2017. Predicting Secretory Proteins with SignalP. Methods Mol Biol Clifton NJ 1611:59–73.

SltF proteins	Signal sequence*
639296061	✓
650739020	✓
643650334	✓
2548760840	✗
639842294	✓
637374448	✓
2514088350	✓
643747691	✓
637332230	✓
637647151	✓
637181464	✓
640789209	✓
643824500	✓
2515008689	✓
2517638777	✓
637075604	✗
649871818	✓
2503200023	✓
641627382	✓
640836883	✓

643463373	✗
2512365150	✗
2511538202	✗
2510372010	✓
640437712	✓
643645133	✓
2524419140	✓
649746019	✓
637924221	✓
2521738048	✗
2549960670	✓
2520250528	✓
2551926637	✓
2520373292	✓
2505292958	✓
2558539010	✗
2574253765	✗
643411101	✗
SltF	✓
SltF2	✓
637287661	✓
639633682	✓
2510237871	✓
2521341176	✓

2525538211	✓
2519014188	✓

* The software used was SignalP V 4.1