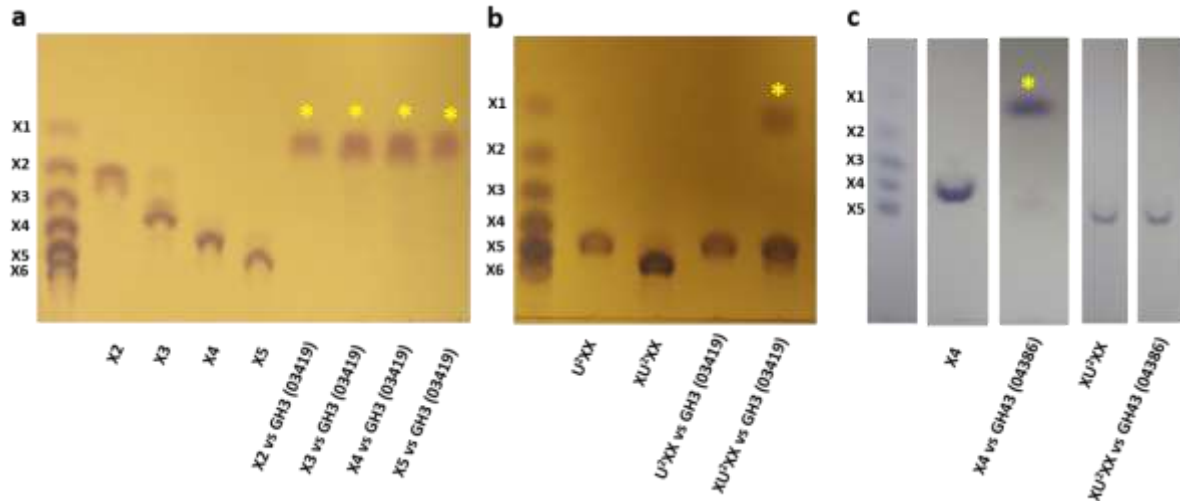
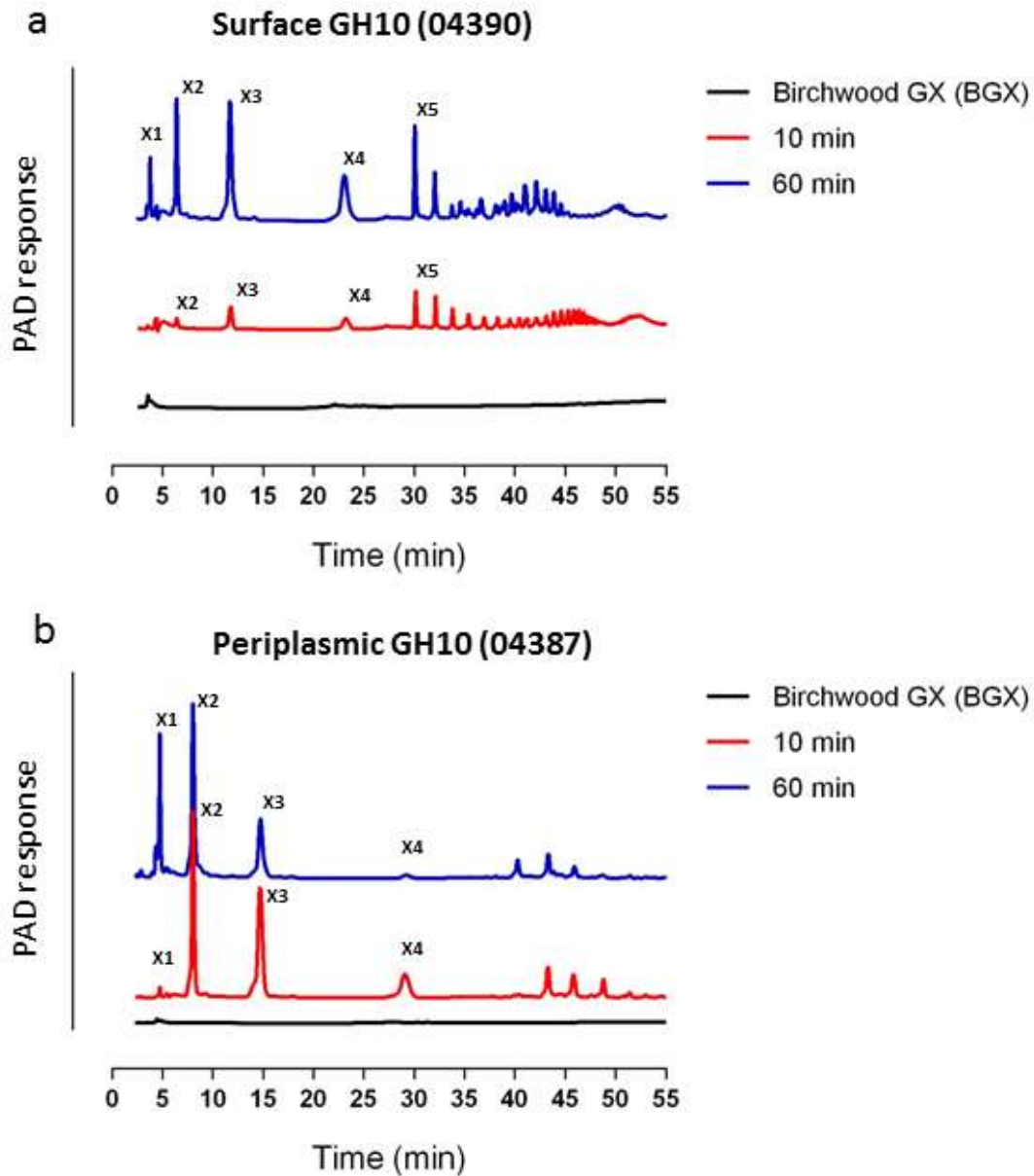


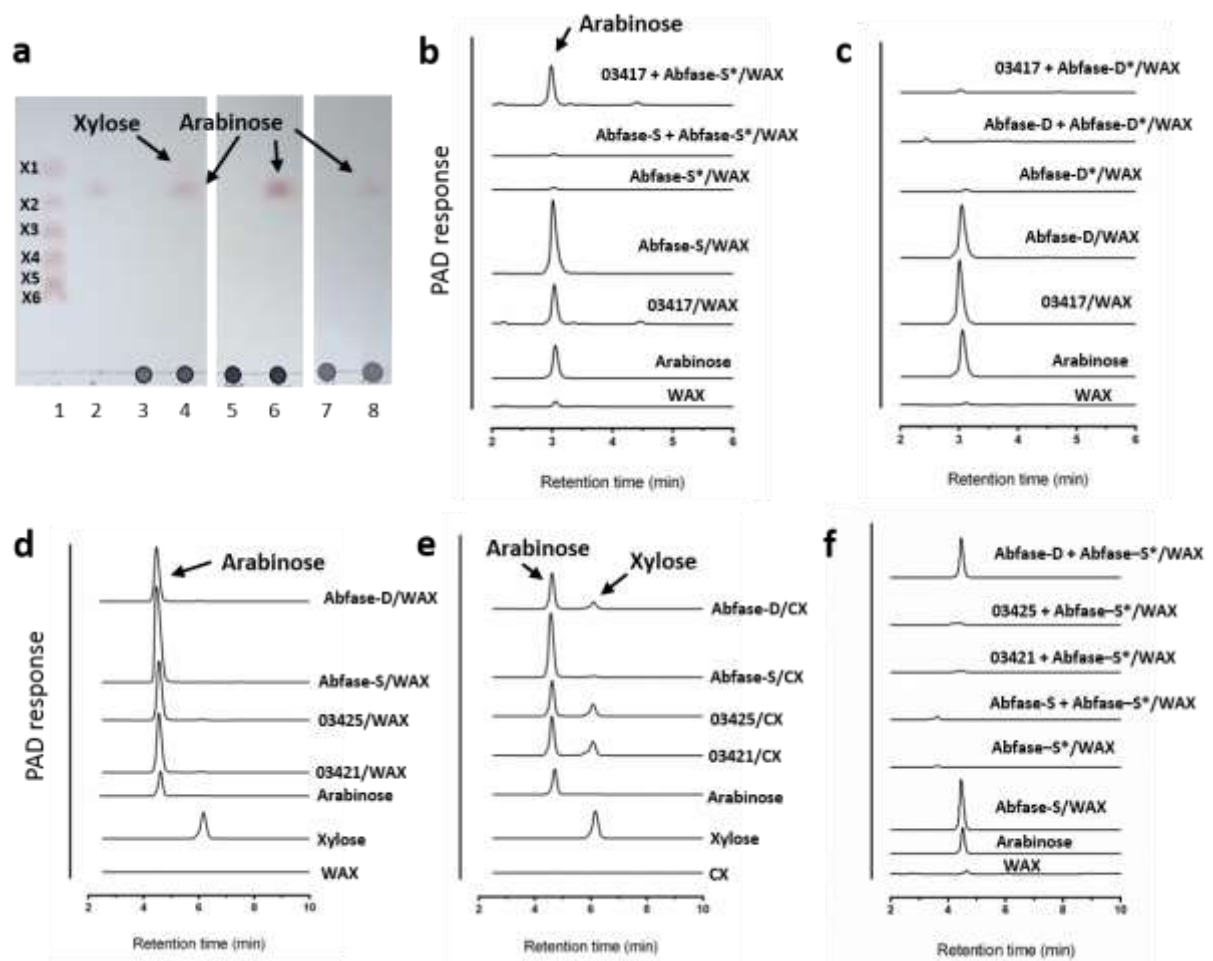
Supplementary Figure 1. The degradation of xylans by *B. ovatus* surface located enzymes. Wild type and mutants (whole PUL knockouts; PUL XylS and PUL XylI KO) of *B. ovatus* were grown on WAX or CX. The cells were then washed and incubated for 5 h at 37 °C with WAX (**a**) or BGX (**b**) under aerobic conditions and were thus metabolically inactive (no active glycan import). In addition, the recombinant form of the GH30 enzyme (BACOVA_03432) was incubated with BGX or WAX for 14 h. The reaction products were analysed by HPAEC-PAD and identified by reference to the following standards; arabinose (A) xylose (X1), xylobiose (X2), xylotriose (X3), xylotetraose (X4) and xylopentaose (X5).



Supplementary Figure 2. Activity of the β -xylosidases BACOVA_03419 and BACOVA_04386. **a**, The surface GH3 β -xylosidase BACOVA_03419, at 1 μ M, was incubated with 1mM β 1,4-linked xylooligosaccharides for 16 h in 50 mM sodium phosphate buffer, pH 7.0, at 37 $^{\circ}$ C. **b**, BACOVA_03419 incubated with 1 mM U²XX and XU²XX (X is an undecorated xylose and U² signifies a xylose decorated with an α -D-(Me)glucuronic acid at O2¹). **c**, The cytoplasmic GH43 β -xylosidase, BACOVA_04386, was incubated with linear xylotetraose and XU²XX. The reactions conditions used in panels **b** and **c** were as described for **a**. X1-X5 are xylooligosaccharide standards and the xylose released is indicated by a yellow asterisk.

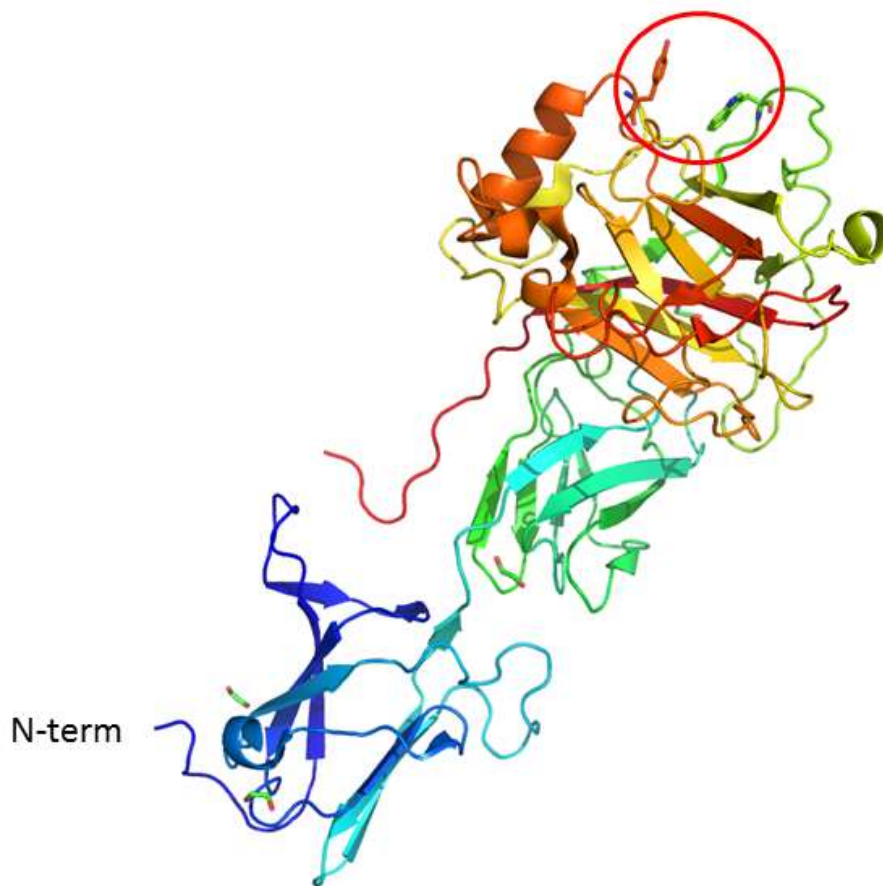


Supplementary Figure 3. The product profile of the surface and periplasmic GH10 xylanases against glucuronoxyylan. The surface (a, BACOVA_04390) and periplasmic (b, BACOVA_04387) GH10 xylanases at 200 nM were incubated with BGX (1% w/v) in 20 mM sodium phosphate buffer, pH 7.0, at 37 °C. The reaction products were subjected to HPAEC and identified by their co-migration with known xylooligosaccharide standards (X1-X5).

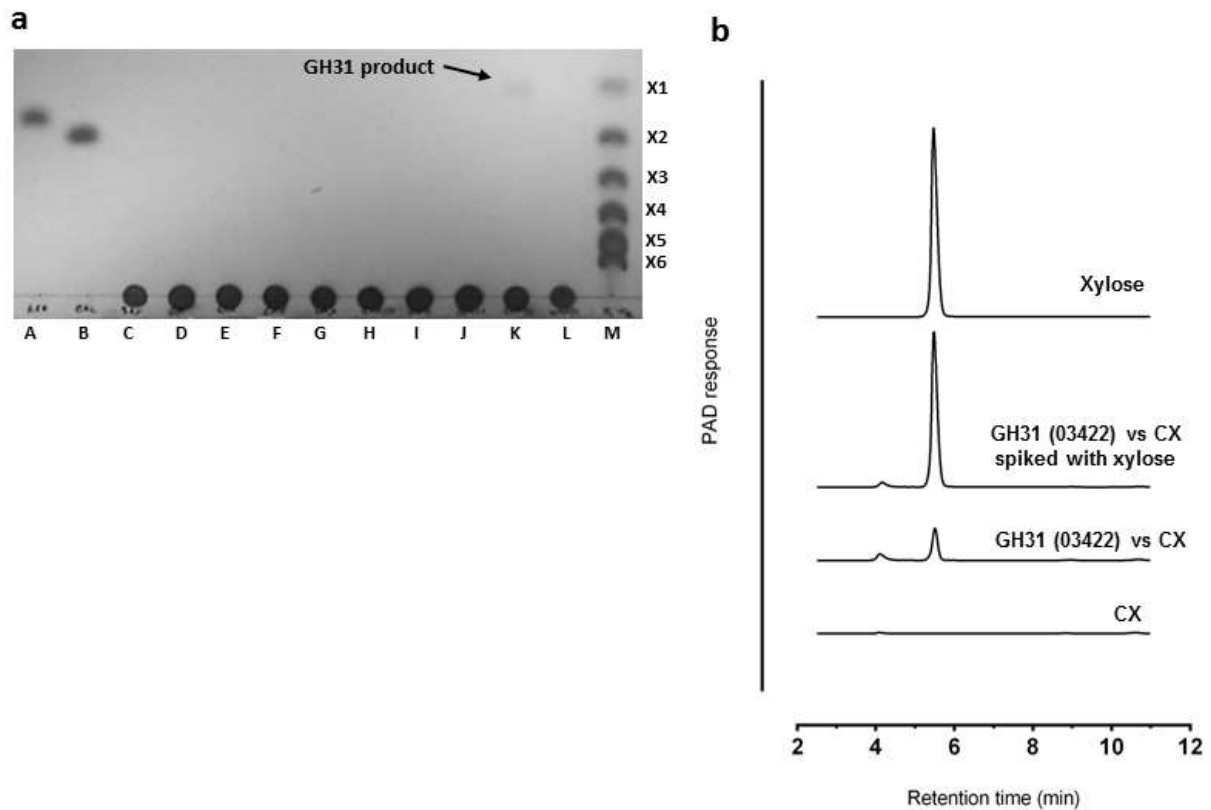


Supplementary Figure 4. Activity of the GH43 α -L-arabinofuranosidases BACOVA_03417, BACOVA_03421 and BACOVA_03425 against polysaccharides. The polysaccharide at 1% (w/v) was incubated with 1 μ M of enzyme at 37 $^{\circ}$ C for 16 h in 50 mM sodium phosphate buffer, pH 7.0. **a**, The products of BACOVA_03417 were analysed by TLC. The lanes contained the following: 1, xylooligosaccharide standards with a d.p. of 1-6; 2, arabinose standard; 3, corn xylan (CX); 4, CX + BACOVA_03417; 5, wheat arabinoxylan (WAX); 6, WAX + BACOVA_03417; 7, sugar beet arabinan; 8, sugar beet arabinan + BACOVA_03417. **b**, HPAEC analysis of the products released from WAX by BACOVA_03417 enzyme together with an arabinofuranosidase from *C. japonicus*² that targets either O2 or O3 single arabinose decorations (defined as Abfase-S). Abfase-S*/WAX denotes WAX pretreated with Abfase-S to remove all single arabinose decorations followed by boiling and then dialysis so that only the polysaccharide remained. This material was then

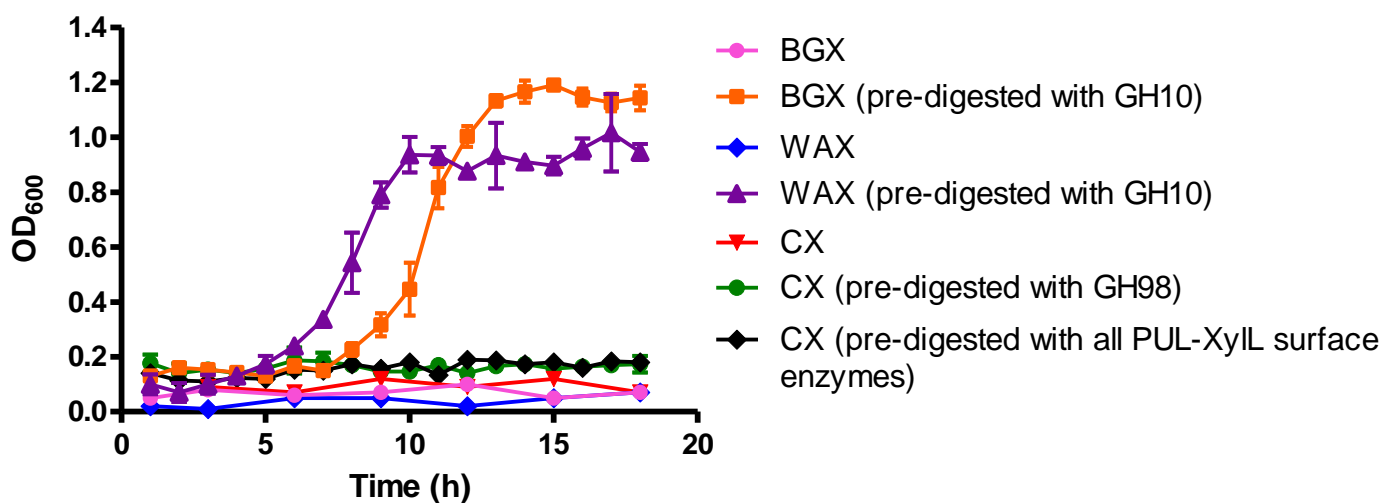
incubated with either additional Abfase-S (Abfase-S + Abfase-S*/WAX; shows that all the single arabinose units had been removed) or BACOVA_03417 (03417 + Abfase-S*/WAX; shows that BACOVA_03417 does not target single arabinose substitutions). **c**, HPAEC analysis of the products released from WAX by BACOVA_03417 together with a *Humicola* arabinofuranosidase³ that specifically targets arabinose units that are linked O3 to double substituted xylose residues (Abfase-D). Abfase-D*/WAX denotes WAX pretreated with Abfase-D to remove arabinose decorations from double substitutions, followed by boiling and then dialysis so that only the polysaccharide remained. This material was then incubated with either additional Abfase-D (Abfase-D + Abfase-D*/WAX; shows that all the double arabinose units had been removed) or BACOVA_03417 (03417 + Abfase-D*/WAX; shows that BACOVA_03417 also targets double arabinose substitutions as no product was released). **d**, and **e**, display HPAEC analysis of products generated by the single specific GH43 α -L-arabinofuranosidases encoded by PUL-XylL, BACOVA_03421 and BACOVA_03425, against WAX and CX, respectively. **f**, Shows that both BACOVA_03421 and BACOVA_03425 are single specific enzymes as neither releases arabinose from WAX after the polysaccharide has been pre-treated with *C. japonicus* Abfase-S (03421 + Abfase-S*/WAX and 03425 + Abfase-S*/WAX).



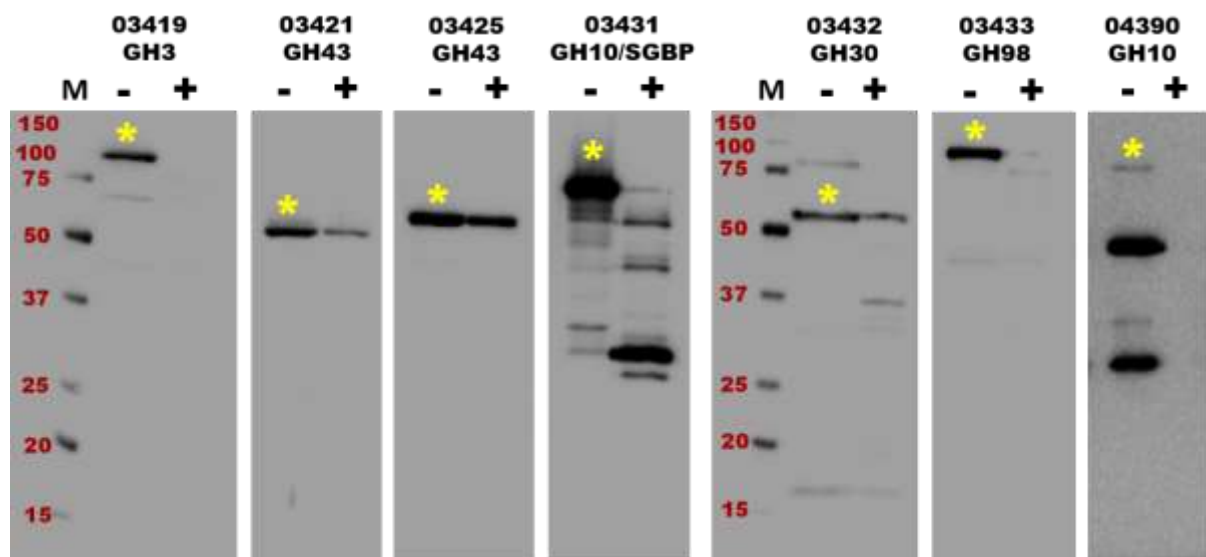
Supplementary Figure 5. Structure of the surface xylan binding protein BACOVA_04391 (PDB 3ORJ). The protein is displayed as a secondary structure schematic that is colour ramped from blue (N-terminus) to red (C-terminus). The three β -sandwich domains are coloured blue, green and orange/red. The likely xylan binding site with the opposing solvent exposed aromatic residues that are proposed to be important in ligand binding is circled in red, with the amino acid side chains shown in stick format. BACOVA_04391 is a lipoprotein and will thus be attached to the outer membrane via its N-term, providing significant spatial separation between the membrane and the xylan binding site.



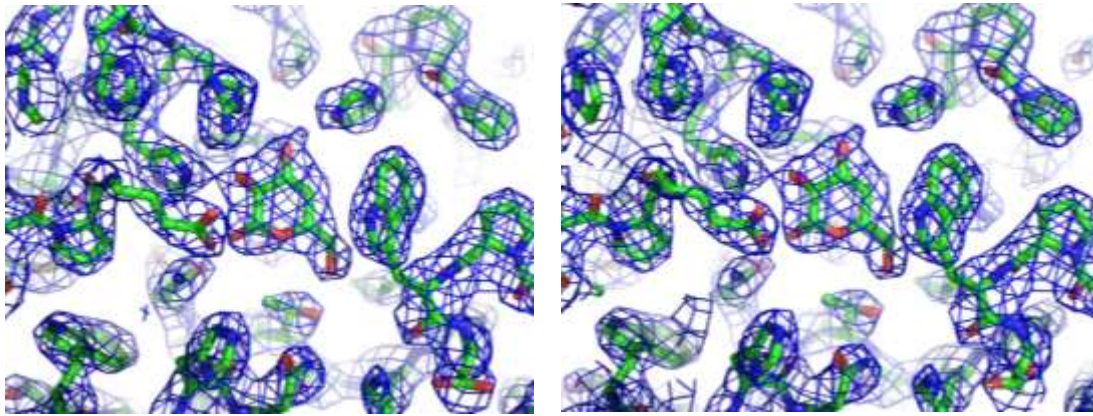
Supplementary Figure 6. Activity of the GH31 enzyme BACOVA_03422. **a**, TLC of rye AX (C, H), BGX (D, G) oat spelt AX (E, I), CX (F, K) and WAX (G, L) all at 5 mg/ml, incubated for 16 h in 20mM sodium phosphate, pH 7.0, at 37 °C in the absence (lanes C-G) or presence (lanes H-L) of 1 μ M BACOVA_03422 GH31. Standards were arabinose (lane A), galactose (lane B) and xylooligosaccharides with a d.p. of 1-6 (lane M). Only incubation of the enzyme with CX (lane K) released a reaction product, which co-migrated with xylose. **b**, The product derived from CX was spiked with xylose and analysed by HPAEC-PAD.



Supplementary Figure 7. Growth of *Bifidobacterium adolescentis* on pre-digested and undigested xylans. Pre-digested indicates the xylan was digested to completion with the surface enzymes from the two *B. ovatus* xylan PULs prior to addition to minimal media at 0.5% final. GH10 = BACOVA_04390 (PUL-XyIS); GH98 = BACOVA_03433 (PUL-XyIL). All surface enzymes from PUL-XyIL = BACOVA_03419 GH3, BACOVA_03421 GH43, BACOVA_03432 GH30 and BACOVA_03433 GH98. Data are averages and standard deviations of 3 separate growths.



Supplementary Figure 8. Western blots of *B. ovatus* cells grown on WAX, either untreated with proteinase K (-) or incubated with 2 mg/ml proteinase K for 16 h (+). The blots were probed with antibodies against the *B. ovatus* enzymes indicated. The yellow asterisk indicates the band at the predicted molecular weight of the target protein. Note that for BACOVA_04390 the major bands visible appear to be processed forms of the full length enzyme. Lanes labelled M are the molecular weight markers (sizes shown in kDa).



Supplementary Figure 9. Stereo image of a portion of the electron density map of BACOVA_03438 GH95. The PDB model is represented as sticks with carbon atoms in green, oxygen atoms in red and nitrogen atoms in blue.

Supplementary Table 1. Kinetics of PUL-XylL and PUL-XylS β -xylosidases against linear and substituted xylooligosaccharides

Substrate	GH3 (03419)	GH43 (04386)
	k_{cat}/K_m ($s^{-1} mM^{-1}$)	k_{cat}/K_m ($s^{-1} mM^{-1}$)
Xylobiose (X_2)	0.06 ($\pm 3.3 \times 10^{-5}$) ^a	0.4 ($\pm 8.5 \times 10^{-4}$)
Xylotriose (X_3)	0.04 ($\pm 1.4 \times 10^{-4}$)	2.2 ($\pm 7.3 \times 10^{-3}$)
Xyloetraose (X_4)	0.06 ($\pm 2.3 \times 10^{-5}$)	2.4 ($\pm 5.1 \times 10^{-5}$)
Aldotriuronic acid (U^2XX) ^b	ND ^c	ND
Aldopentauronic acid (XU^2XX)	0.14 ($\pm 1.7 \times 10^{-4}$)	ND
Arabinotetrasaccharide (XA^3X) ^d	0.03 ($\pm 2.9 \times 10^{-5}$)	ND

^aValues are from a single experiment and errors are derived from the fit of the linear regression to the data.

^b U^2 = (Me)GlcA substituted on the O2 of xylose².

^cND = no activity detected.

^d A^3 = arabinose substituted on the O3 of xylose.

Supplementary Table 2. Kinetics of PUL-XylS and PUL-XylL GH10 and GH30 endo-xylanases against xylan and xylooligosaccharides

Substrate	GH30 (03432)	GH10 (03431)	GH10 (04387)	GH10 (04390)
	k_{cat}/K_m ($s^{-1} mg^{-1} ml$)	k_{cat}/K_m ($s^{-1} mg^{-1} ml$)	k_{cat}/K_m ($s^{-1} mg^{-1} ml$)	k_{cat}/K_m ($s^{-1} mg^{-1} ml$)
Birch glucuronoxylan (BGX)	3.8 (± 0.13) ^a	ND	150 (± 16.7)	4.7 (± 0.39)
			Relative activity	Relative activity
Xylotriose (X_3)	ND ^b	ND	0.001	UQ ^c
Xyloetraose (X_4)	ND	ND	0.15	0.0003
Xylopentaose (X_5)	ND	ND	1.1	0.004
Xylohexaose (X_6)	ND	ND	1	0.03

^aValues are from a single experiment and errors are derived from the fit of the linear regression to the data.

^bND = no activity detected.

^cUQ = activity too low to quantify.

Supplementary Table 3. ITC analysis of the SusD-likes and surface glycan binding proteins from the xylan PULs binding to xylans and xylooligosaccharides

Protein	Ligand	K _a (M ⁻¹)	ΔG (kcal mol ⁻¹)	ΔH (kcal mol ⁻¹)	TΔS (kcal mol ⁻¹)	n
PUL-XylS						
SusD-like (BACOVA_04392)	BGX ^a	3.7 × 10 ³ (±0.5) ^b	-4.9	-15.4 (±1.2)	-10.5	1.0 ^c
	WAX	2.8 × 10 ³ (±0.2)	-4.7	-10.2 (±0.4)	-5.5	1.0
	CX	NB ^d	-	-	-	-
	X6	8.0 × 10 ² (±1.3)	-4.0	- ^e	-	-
	≤X5	NB	-	-	-	-
SGBP ^f (BACOVA_04391)	BGX	2.0 × 10 ⁴ (±1.2)	-5.5	-10.8 (±1.4)	-6.3	1.0
	WAX	1.5 × 10 ⁴ (±0.6)	-5.7	-10.2 (±1.3)	-4.5	1.0
	CX	NB	-	-	-	-
	X6	1.8 × 10 ⁴ (±0.3)	-5.8	-11.2 (±3.8)	-5.4	0.85 (±0.2)
	X5	9.1 × 10 ³ (±0.7)	-5.4	-8.9 (±0.2)	-3.5	1.0 (±0.2)
	X4	1.3 × 10 ³ (±0.1)	-4.2	- ^e	-	-
≤X3	NB	-	-	-	-	
PUL-XylL						
SusD-like_1 (BACOVA_03427) ^g	BGX	8.4 × 10 ⁴ (±0.9)	-6.7	-9.2 (±0.5)	-2.9	1.0
	WAX	NB	-	-	-	-
	CX	1.5 × 10 ⁵ (±0.1)	-7.1	-8.1 (±0.2)	-1.0	1.0
	≤X6	NB	-	-	-	-
SGBP (BACOVA_03431; inactive GH10)	BGX	1.3 × 10 ⁴ (±0.1)	-5.6	-9.9 (±0.8)	-4.3	1.0
	WAX	NB	-	-	-	-
	CX	1.5 × 10 ⁴ (±0.1)	-5.7	-17.3 (±0.8)	-11.6	1.0
	≤X6	NB	-	-	-	-

^aBGX = birchwood glucuronoxylan, WAX = wheat arabinoxylan, CX = corn glucuronoarabinoxylan.

^bData are averages and standard deviations of between two and three independent titrations.

^cThe data with xylans was fit with a molar concentration of ligand such that n=1.

^dNB – no binding detected. ^eAffinity too low to obtain accurate thermodynamic data. ^fSGBP – surface glycan binding protein. ^gThe other SusD-like from PUL-XylL, BACOVA_03429, could not be tested as the protein did not express in a recombinant form.

Supplementary Table 4. Kinetics of PUL-XylL double and single specific α -L-arabinofuranosidases against arabinoxylans

Enzyme	substrate	k_{cat} (s^{-1})	K_m (mM)	k_{cat}/K_m ($s^{-1}mM^{-1}$)
GH43 (03417 - double)	Wheat arabinoxylan (WAX)	3.3 (± 0.15) ^a	0.22 (± 0.004)	15
GH43 (03421 - single)	WAX	7.0 (± 0.6)	0.13 (± 0.005)	54
GH43 (03425 - single)	WAX	11.0 (± 0.8)	0.18 (± 0.006)	61
GH43 (03417 - double)	Corn xylan (CX)	2.7 (± 0.2)	0.09 (± 0.002)	30
GH43 (03421 - single)	CX	5.8 (± 0.4)	0.95 (± 0.003)	6
GH43 (03425 - single)	CX	9.3 (± 1.1)	2.9 (± 0.01)	3

^aValues are averages and standard deviations of three independent experiments.

Supplementary Table 5. Kinetics of PUL-XylS GH67 vs *C. japonicus* GlcA67A⁴ α -glucuronidases

Substrate	<i>Bo</i> GH67 (04385)	<i>Cj</i> GlcA67A
	k_{cat}/K_m ($s^{-1}mM^{-1}$)	k_{cat}/K_m ($s^{-1}mM^{-1}$)
Aldotetrauronic acid (U ² XX)	270 (± 0.23) ^o	320 (± 0.28)

^oValues are from a single experiment and errors are derived from the fit of the linear regression to the data.

Supplementary Table 6. Kinetics of PUL-XylL GH31 vs *C. japonicus* Xyl31A⁵ α -xylosidases

Substrate	<i>Bo</i> GH31 (03422)	<i>Cj</i> Xyl31A
	k_{cat}/K_m ($s^{-1}mM^{-1}$)	k_{cat}/K_m ($s^{-1}mM^{-1}$)
Xyloglucan heptasaccharide (XXXG)	0.02 (± 0.003) ^o	403 (± 0.26)
Corn xylan (CX)	58 (± 0.02)	0.65 (± 0.003)

^oValues are from a single experiment and errors are derived from the fit of the linear regression to the data.

Supplementary Table 7. Activity of wild type and mutant forms of PUL-XylI α -L-galactosidase vs *B. bifidum* α -L-fucosidase AfcA⁶ GH95s

Substrate	WT <i>Bo</i> GH95 (03438)	<i>Bb</i> GH95 (AfcA)	<i>Bo</i> GH95 E501A	<i>Bo</i> GH95 T370H
	k_{cat}/K_m (s ⁻¹ mM ⁻¹)	k_{cat}/K_m (s ⁻¹ mM ⁻¹)		
Corn xylan (CX)	26 (\pm 0.51) ^a	0.02 (\pm 0.006)	ND ^b	ND
	Relative activity	Relative activity		Relative activity
2'fucosyl-lactose ^c	1	3700	-	-
2-chloro-4-nitrophenyl α -L-fucose ^d	1	-	-	0.13

^aValues are from a single experiment and errors are derived from the fit of the linear regression to the data.

^bND = no activity detected.

^cValues for fucosyl lactose are relative activity at 0.25mM sugar as the K_m of the *Bb*GH95 for this substrate was too low to measure a valid k_{cat}/K_m .

^dValues for 2-chloro-4-nitrophenyl α -L-fucose are relative activity vs 5mM substrate.

Supplementary Table 8. List of Bacteroidetes species searched for the presence of XylS and XylL PULs

Organism name	NCBI taxonomy ID	IMG/M HMP project ID
<i>Aequorivita sublithicola</i> DSM 14238	746697	2509276063
<i>Algoriphagus marincola</i> DSM 16067	1120966	2523533625
<i>Algoriphagus vanfongensis</i> DSM 17529	1120968	2523231017
<i>Alistipes finegoldii</i> DSM 17242	679935	2509601035
<i>Alistipes indistinctus</i> YIT 12060	742725	2513237277
<i>Alistipes onderdonkii</i> DSM 19147	1120974	2517572213
<i>Alistipes putredinis</i> DSM 17216	445970	641736205
<i>Alistipes shahii</i> WAL 8301	717959	650377904
<i>Alistipes</i> sp. HGB5	908612	2512047053
<i>Alloprevotella rava</i> F0323	679199	2513237313
<i>Anaerophaga thermohalophila</i> DSM 12881	886379	2515154051
<i>Anaerophaga thermohalophila</i> str. Valhall	1144273	2515154050
<i>Arenibacter certesii</i> DSM 19833	1121011	2523231023
<i>Bacteroides barnesiae</i> DSM 18169	1121094	2517572120
<i>Bacteroides caccae</i> ATCC 43185	411901	640963023
<i>Bacteroides cellulosilyticus</i> DSM 14838	537012	643886111
<i>Bacteroides clarus</i> YIT 12056	762984	651324010
<i>Bacteroides coprocola</i> DSM 17136	470145	642791613
<i>Bacteroides coprophilus</i> DSM 18228	547042	643886197
<i>Bacteroides coprosuis</i> DSM 18011	679937	2503982039
<i>Bacteroides dorei</i> 5_1_36	556260	646206258
<i>Bacteroides dorei</i> DSM 17855	483217	642979370
<i>Bacteroides eggerthii</i> 1_2_48FAA	665953	649989912
<i>Bacteroides eggerthii</i> DSM 20697	483216	642979334
<i>Bacteroides finegoldii</i> DSM 17565	483215	642979319
<i>Bacteroides fluxus</i> YIT 12057	763034	651324011
<i>Bacteroides fragilis</i> 3_1_12	457424	645058788
<i>Bacteroides fragilis</i> 638R	862962	650377910
<i>Bacteroides fragilis</i> NCTC 9343	272559	637000024
<i>Bacteroides fragilis</i> YCH46	295405	637000025
<i>Bacteroides gallinarum</i> DSM 18171	1121096	2519899680
<i>Bacteroides graminisolvens</i> DSM 19988	1121097	2524614553
<i>Bacteroides helcogenes</i> P 36-108	693979	649633012
<i>Bacteroides intestinalis</i> DSM 17393	471870	642791621
<i>Bacteroides massiliensis</i> DSM 17679	1121098	2515154074
<i>Bacteroides oleiciplenus</i> YIT 12058	742727	2534681837
<i>Bacteroides ovatus</i> 3_8_47FAA	665954	651324012
<i>Bacteroides ovatus</i> ATCC 8483	411476	641380449
<i>Bacteroides ovatus</i> NLAE-zl-C34	1145612	2513020027
<i>Bacteroides ovatus</i> NLAE-zl-H304	1201624	2515154064

<i>Bacteroides ovatus</i> SD CC 2a	702444	647000212
<i>Bacteroides ovatus</i> SD CMC 3f	702443	647000213
<i>Bacteroides plebeius</i> DSM 17135	484018	642979351
<i>Bacteroides propionificiens</i> DSM 19291	1121099	2517093002
<i>Bacteroides pyogenes</i> DSM 20611	1121100	2524614554
<i>Bacteroides salanitronis</i> DSM 18170	667015	649633013
<i>Bacteroides salyersiae</i> WAL 10008	1121101	2531839250
<i>Bacteroides</i> sp. 1_1_14	469585	648861000
<i>Bacteroides</i> sp. 1_1_30	457387	651324013
<i>Bacteroides</i> sp. 1_1_6	469586	646206272
<i>Bacteroides</i> sp. 2_1_16	469587	647533110
<i>Bacteroides</i> sp. 2_1_22	469588	647533111
<i>Bacteroides</i> sp. 2_1_33B	469589	647533112
<i>Bacteroides</i> sp. 2_1_56FAA	665938	651324014
<i>Bacteroides</i> sp. 2_1_7	457388	645058782
<i>Bacteroides</i> sp. 2_2_4	469590	646206266
<i>Bacteroides</i> sp. 20_3	469591	648861001
<i>Bacteroides</i> sp. 3_1_19	469592	648861002
<i>Bacteroides</i> sp. 3_1_23	457390	648861003
<i>Bacteroides</i> sp. 3_1_33FAA	457391	647533113
<i>Bacteroides</i> sp. 3_1_40A	469593	649989913
<i>Bacteroides</i> sp. 3_2_5	457392	646206273
<i>Bacteroides</i> sp. 4_1_36	457393	649989914
<i>Bacteroides</i> sp. 4_3_47FAA	457394	646206274
<i>Bacteroides</i> sp. 9_1_42FAA	457395	646206263
<i>Bacteroides</i> sp. D1	556258	646206264
<i>Bacteroides</i> sp. D2	556259	645951811
<i>Bacteroides</i> sp. D20	585543	647533114
<i>Bacteroides</i> sp. D22	585544	648861004
<i>Bacteroides</i> sp. HPS0048	1078089	2529293266
<i>Bacteroides stercoris</i> ATCC 43183	449673	641736196
<i>Bacteroides thetaiotaomicron</i> NLAE-zl-C523	1145805	2513020028
<i>Bacteroides thetaiotaomicron</i> NLAE-zl-H207	1201516	2515154063
<i>Bacteroides thetaiotaomicron</i> NLAE-zl-H492	1201827	2515154067
<i>Bacteroides thetaiotaomicron</i> VPI-5482	226186	637000026
<i>Bacteroides uniformis</i> ATCC 8492	411479	641380447
<i>Bacteroides vulgatus</i> ATCC 8482	435590	640753008
<i>Bacteroides vulgatus</i> PC510	702446	647000214
<i>Bacteroides xylanisolvens</i> NLAE-zl-C29	1145558	2513020025
<i>Bacteroides xylanisolvens</i> NLAE-zl-G310	1189854	2514885005
<i>Bacteroides xylanisolvens</i> NLAE-zl-G421	1189970	2514885006
<i>Bacteroides xylanisolvens</i> NLAE-zl-P352	1146212	2513020030
<i>Bacteroides xylanisolvens</i> NLAE-zl-P393	1146256	2513020031
<i>Bacteroides xylanisolvens</i> NLAE-zl-P727	1146614	2513020033

<i>Bacteroides xylanisolvens</i> NLAE-zl-P732	1146620	2513020034
<i>Bacteroides xylanisolvens</i> NLAE-zl-P736	1146624	2513020035
<i>Bacteroides xylanisolvens</i> SD CC 1b	702447	647000215
<i>Bacteroides xylanisolvens</i> XB1A	657309	650377911
<i>Bacteroidetes bacterium</i> SCGC AAA027-G08	938698	2524023173
<i>Bacteroidetes bacterium</i> SCGC AAA027-N21	938709	2524023177
<i>Balneola vulgaris</i> DSM 17893	1121104	2515154152
<i>Barnesiella intestinihominis</i> YIT 11860	742726	2529292932
<i>Barnesiella viscericola</i> DSM 18177	880074	2509276067
<i>Belliella baltica</i> DSM 15883	232259	2509276058
<i>Butyricimonas synergistica</i> DSM 23225	1121129	2515154026
<i>Capnocytophaga canimorsus</i> Cc5	860228	650716019
<i>Capnocytophaga cynodegmi</i> DSM 19736	926551	2513237123
<i>Capnocytophaga gingivalis</i> ATCC 33624	553178	643886113
<i>Capnocytophaga ochracea</i> DSM 7271	521097	644736338
<i>Capnocytophaga ochracea</i> F0287	873517	649989920
<i>Capnocytophaga</i> sp. oral taxon 329 str. F0087	706436	651324019
<i>Capnocytophaga</i> sp. oral taxon 338 str. F0234	888059	651324020
<i>Capnocytophaga sputigena</i> ATCC 33612	553177	642979305
<i>Cellulophaga algicola</i> DSM 14237	688270	649633031
<i>Cellulophaga lytica</i> DSM 7489	867900	649633032
<i>Chitinophaga pinensis</i> DSM 2588	485918	644736340
<i>Chryseobacterium caeni</i> DSM 17710	1121285	2524614876
<i>Chryseobacterium daeguense</i> DSM 19388	1121286	2523533536
<i>Chryseobacterium gregarium</i> DSM 19109	1121287	2523533548
<i>Chryseobacterium palustre</i> DSM 21579	1121288	2523533629
<i>Croceibacter atlanticus</i> HTCC2559	216432	648028020
<i>Cyclobacterium marinum</i> DSM 745	880070	2506381023
<i>Cytophaga fermentans</i> DSM 9555	869213	2509601038
<i>Dyadobacter fermentans</i> DSM 18053	471854	644736356
<i>Dysgonomonas gadei</i> ATCC BAA-286	742766	651324026
<i>Dysgonomonas mossii</i> DSM 22836	742767	651324027
<i>Echinicola pacifica</i> DSM 19836	1121859	2515154083
<i>Echinicola vietnamensis</i> DSM 17526	926556	2509276020
<i>Emticicia oligotrophica</i> DSM 17448	929562	2506485001
<i>Flavobacteria bacterium</i> BAL38	391598	640612204
<i>Flavobacteria bacterium</i> BBFL7	156586	638341093
<i>Flavobacteria bacterium</i> MS024-2A	487796	643886118
<i>Flavobacteria bacterium</i> MS024-3C	487797	643886079
<i>Flavobacteria bacterium</i> MS190-1F	913852	2507262045
<i>Flavobacteriaceae bacterium</i> 3519-10	531844	644736368
<i>Flavobacteriaceae bacterium</i> JJC	512012	2515154008
<i>Flavobacterium columnare</i> ATCC 49512	1041826	2511231122
<i>Flavobacterium daejeonense</i> DSM 17708	1121887	2523533615

<i>Flavobacterium filum</i> DSM 17961	1121889	2523533606
<i>Flavobacterium gelidilacus</i> DSM 15343	1121891	2523533607
<i>Flavobacterium johnsoniae</i> UW101	376686	644736369
<i>Flavobacterium psychrophilum</i> JIP02 86	402612	640753027
<i>Flavobacterium rivuli</i> DSM 21788	1121895	2519103183
<i>Flavobacterium soli</i> DSM 19725	1121897	2522572162
<i>Flavobacterium</i> sp. CF136	1144313	2513020052
<i>Flavobacterium subsaxonicum</i> DSM 21790	1121898	2524614667
<i>Flavobacterium tegetincola</i> DSM 22377	1121901	2524023138
<i>Flectobacillus major</i> DSM 103	929703	2509601041
<i>Flexibacter litoralis</i> DSM 6794	880071	2509276059
<i>Flexithrix dorotheae</i> DSM 6795	1121904	2515154145
<i>Gramella echinicola</i> DSM 19838	1121931	2524614734
<i>Gramella forsetii</i> KT0803	411154	639633025
<i>Gramella portivictoriae</i> DSM 23547	1121932	2524614725
<i>Haliscomenobacter hydrossis</i> DSM 1100	760192	2504756004
<i>Joostella marina</i> DSM 19592	926559	2509276026
<i>Krokinobacter</i> sp. 4H-3-7-5	983548	650716043
<i>Lacinutrix</i> sp. 5H-3-7-4	983544	650716044
<i>Leadbetterella byssophila</i> DSM 17132	649349	649633063
<i>Maribacter</i> sp. HTCC2170	313603	648028027
<i>Marivirga tractuosa</i> DSM 4126	643867	649633065
<i>Mucilaginibacter paludis</i> DSM 18603	714943	2506520020
<i>Muricauda ruestringensis</i> DSM 13258	886377	2505679007
<i>Myroides injenensis</i> M09-0166	1008457	2523231007
<i>Myroides odoratimimus</i> CCUG 10230	883150	2513237331
<i>Myroides odoratimimus</i> CCUG 12901	883152	2522572175
<i>Myroides odoratimimus</i> CCUG 3837	883153	2523231006
<i>Myroides odoratimimus</i> CIP 101113	883154	2522572177
<i>Myroides odoratimimus</i> CIP 103059	883155	2522572176
<i>Myroides odoratus</i> DSM 2801	929704	2506520050
<i>Niabella aurantiaca</i> DSM 17617	1122605	2515154175
<i>Niabella soli</i> DSM 19437	929713	2506783006
<i>Niastella koreensis</i> GR20-10	700598	2506520016
<i>Odoribacter laneus</i> YIT 12061	742817	2513237280
<i>Odoribacter splanchnicus</i> DSM 20712	709991	649633078
<i>Olivibacter sitiensis</i> DSM 17696	1122621	2515154027
<i>Ornithobacterium rhinotracheale</i> DSM 15997	867902	2509601001
<i>Owenweeksia hongkongensis</i> DSM 17368	926562	2508501098
<i>Paludibacter propionigenes</i> WB4	694427	649633080
<i>Parabacteroides distasonis</i> ATCC 8503	435591	640753039
<i>Parabacteroides johnsonii</i> DSM 18315	537006	642979358
<i>Parabacteroides merdae</i> ATCC 43184	411477	640963016
<i>Parabacteroides</i> sp. D13	563193	646206279

<i>Paraprevotella clara</i> YIT 11840	762968	2513237390
<i>Paraprevotella xylaniphila</i> YIT 11841	762982	651324083
<i>Pedobacter glucosidilyticus</i> DSM 23534	540906	2523533576
<i>Pedobacter heparinus</i> DSM 2366	485917	644736398
<i>Pedobacter oryzae</i> DSM 19973	468059	2523231054
<i>Pedobacter saltans</i> DSM 12145	762903	649633082
<i>Pedobacter</i> sp. BAL39	391596	640963036
<i>Polaribacter irgensii</i> 23-P	313594	638341152
<i>Polaribacter</i> sp. MED152	313598	638341218
<i>Porphyromonas asaccharolytica</i> DSM 20707	879243	2504756018
<i>Porphyromonas asaccharolytica</i> PR426713P-I	910312	649989985
<i>Porphyromonas bennonis</i> DSM 23058	1122971	2517093003
<i>Porphyromonas endodontalis</i> ATCC 35406	553175	643886148
<i>Porphyromonas gingivalis</i> ATCC 33277	431947	642555148
<i>Porphyromonas gingivalis</i> TDC60	1030843	650716072
<i>Porphyromonas gingivalis</i> W83	242619	637000209
<i>Porphyromonas gulae</i> DSM 15663	1122972	2518645603
<i>Porphyromonas macacae</i> DSM 20710	1122974	2517093006
<i>Porphyromonas somerae</i> DSM 23386	1122975	2515154109
<i>Porphyromonas uenonis</i> 60-3	596327	643886142
<i>Porphyromonas uenonis</i> DSM 23387	1122976	2528311143
<i>Prevotella albensis</i> DSM 11370	1122978	2526164514
<i>Prevotella amnii</i> CRIS 21A-A	679191	648276713
<i>Prevotella amnii</i> DSM 23384	1122979	2518645615
<i>Prevotella baroniae</i> DSM 16972	1122980	2526164515
<i>Prevotella bergensis</i> DSM 17361	585502	645951825
<i>Prevotella bivia</i> DSM 20514	868129	2509601036
<i>Prevotella bivia</i> JCVIHMP010	553171	647000292
<i>Prevotella bryantii</i> B14	752555	648276714
<i>Prevotella bryantii</i> C21a	1280705	2524614710
<i>Prevotella buccae</i> ATCC 33574	873513	649989986
<i>Prevotella buccae</i> D17	575611	647533195
<i>Prevotella buccalis</i> ATCC 35310	679190	647000293
<i>Prevotella copri</i> DSM 18205	537011	643886200
<i>Prevotella dentalis</i> DSM 3688	908937	2509276066
<i>Prevotella denticola</i> CRIS 18C-A	944557	651324088
<i>Prevotella denticola</i> DSM 20614	1122982	2526164516
<i>Prevotella denticola</i> F0289	767031	650716073
<i>Prevotella disiens</i> FB035-09AN	866771	648276715
<i>Prevotella histicola</i> F0411	857291	2513237391
<i>Prevotella intermedia</i> 17	246198	2501416924
<i>Prevotella intermedia</i> DSM 20706	1122984	2523231016
<i>Prevotella loescheii</i> DSM 19665	1122985	2518645604
<i>Prevotella maculosa</i> DSM 19339	1122986	2521172638

<i>Prevotella maculosa</i> OT 289	999422	2513237350
<i>Prevotella marshii</i> DSM 16973	862515	648276716
<i>Prevotella melaninogenica</i> ATCC 25845	553174	648028051
<i>Prevotella melaninogenica</i> D18	575612	647533196
<i>Prevotella micans</i> DSM 21469	1122987	2515154159
<i>Prevotella micans</i> F0438	883158	2513237310
<i>Prevotella multififormis</i> DSM 16608	888743	649989987
<i>Prevotella multisaccharivorax</i> DSM 17128	688246	2503754046
<i>Prevotella nanceiensis</i> DSM 19126	1122988	2514885007
<i>Prevotella nigrescens</i> ATCC 33563	997352	651324089
<i>Prevotella oralis</i> ATCC 33269	873533	649989988
<i>Prevotella oralis</i> HGA0225	1203550	2541047008
<i>Prevotella oris</i> C735	563008	648861012
<i>Prevotella oris</i> DSM 18711	1122989	2517572237
<i>Prevotella oris</i> F0302	649760	645951806
<i>Prevotella oulorum</i> F0390	702438	2513237263
<i>Prevotella pallens</i> ATCC 700821	997353	651324090
<i>Prevotella paludivivens</i> DSM 17968	1122990	2515154072
<i>Prevotella ruminicola</i> 23	264731	646564560
<i>Prevotella salivae</i> DSM 15606	888832	649989989
<i>Prevotella</i> sp. AGR2160	1280674	2524614711
<i>Prevotella</i> sp. C561	563031	2513237323
<i>Prevotella</i> sp. oral taxon 299 str. F0039	575614	647533197
<i>Prevotella</i> sp. oral taxon 306 str. F0472	1095752	2513237374
<i>Prevotella</i> sp. oral taxon 317 str. F0108	575615	647533198
<i>Prevotella</i> sp. oral taxon 472 str. F0295	619693	647000294
<i>Prevotella stercorea</i> DSM 18206	1002367	2513237318
<i>Prevotella tanneriae</i> ATCC 51259	626522	645951840
<i>Prevotella timonensis</i> CRIS 5C-B1	679189	647000295
<i>Prevotella timonensis</i> DSM 22865	1122992	2523533627
<i>Prevotella veroralis</i> DSM 19559	1122993	2517572238
<i>Prevotella veroralis</i> F0319	649761	645951836
<i>Proteiniphilum acetatigenes</i> DSM 18083	1123008	2518645612
<i>Psychroflexus torquis</i> ATCC 700755	313595	638341165
<i>Psychroflexus tropicus</i> DSM 15496	1123035	2518645613
<i>Rhodothermus marinus</i> DSM 4252	518766	646311949
<i>Rhodothermus marinus</i> SG0.5JP17-171	762569	2506520042
<i>Rhodothermus marinus</i> SG0.5JP17-172	762570	2505679078
<i>Riemerella anatipestifer</i> ATCC 11845 = DSM 15868	693978	2512564079
<i>Riemerella anatipestifer</i> RA-CH-1	1228997	2518645536
<i>Riemerella anatipestifer</i> RA-CH-2	1271752	2521172679
<i>Riemerella anatipestifer</i> RA-GD	992406	651053063
<i>Riemerella columbina</i> DSM 16469	1123058	2517434010
<i>Rikenella microfusus</i> DSM 15922	880526	2509601020

<i>Robiginitalea biformata</i> HTCC2501	313596	646311950
<i>Rudanella lutea</i> DSM 19387	1089547	2517093013
<i>Runella slithyformis</i> DSM 19594	761193	2505679030
<i>Salinibacter ruber</i> DSM 13855	309807	637000250
<i>Salisaeta longa</i> DSM 21114	1089550	2517093009
<i>Saprospira grandis</i> str. Lewin	984262	2512564032
<i>Sediminibacterium</i> sp. OR43	1076522	2509887033
<i>Sediminibacterium</i> sp. OR53	925409	2516143025
<i>Segetibacter koreensis</i> DSM 18137	1123248	2516143113
<i>Solitalea canadensis</i> DSM 3403	929556	2509276062
<i>Sphingobacterium</i> sp. 21	743722	2505679080
<i>Sphingobacterium spiritivorum</i> ATCC 33300	525372	643886135
<i>Sphingobacterium spiritivorum</i> ATCC 33861	525373	645058810
<i>Spirosoma linguale</i> DSM 74	504472	646311954
<i>Spirosoma luteum</i> DSM 19990	1123276	2515154133
<i>Spirosoma panaciterrae</i> DSM 21099	1123277	2515154167
<i>Spirosoma spitsbergense</i> DSM 19989	1123278	2515154151
<i>Tannerella forsythensis</i> ATCC 43037	203275	2512047036
<i>Tannerella</i> sp. 6_1_58FAA_CT1	665949	2513237267
<i>Thermonema rossianum</i> DSM 10300	1313301	2523533595
<i>Weeksella virosa</i> DSM 16922	865938	650377986
<i>Xylanibacter oryzae</i> DSM 17970	915438	2509276029
<i>Zunongwangia profunda</i> SM A87	655815	646564591

Supplementary Table 9. Cloning primers and *E. coli* expression vectors used in this study.

Gene	Vector	Forward primer	Reverse primer
BACOVA_03417	pET22b ^a	CTTATGGAATTCGACGTGCCGATATGTCAGATGTCGCGCAG ^b	CTCCCGCTCGAGCATTGACGCATCACAGAACGAATAATCAAAGG ^c
BACOVA_03419	pET28b	CACTACCATATGAAGCAATTGCTGCCTTACCAGGAC	CGACCGCTCGAGTTACTGTACGTTTATTACAATCG
BACOVA_03421	pRSET A ^c	CGCGGATCCAATATGGAGGTGAGTGGC	CCCAAGCTTATAACCAACAGATGTTGCATACAG
BACOVA_03422	pRSET A	CGCGGATCCAATATCAGAAAACATCGTC	CCCAAGCTTAACTTTACAGAAATCGCTTTCCATTATAATC
BACOVA_03423	pET21a	CGCGGATCCAATAAACAGATAACAAGTCTGATG	CCCAAGCTTTTTGGTTATTAATACTCCACCATTGCAAGG
BACOVA_03424	pRSET A	CGCGGATCCAATCCTATTATACAAAACAAAATATACTG	CCCAAGCTTTTCAAATTTCCAATAATCCAGATTGAAAAGTTTG
BACOVA_03425	pRSET A	CGCGGATCCAATCAGAAAGAAACAACAATTACCG	CCCAAGCTTTTTGCAAGTAGCGTATAATCCAATTGTAGTAC
BACOVA_03427	pET22b	GCGCGGGGATCCACTGACTATTTGGACAATCTCCATATAG	GAAACGCTCGAGCCATCCCGATTCTGGCCAAAG
BACOVA_03429	pET22b	GTACAGGGATCCTTCGAGCCGGCCATCGAGAAC	GCGGGCTCGAGTTTCCACCTCTATTTGTACAAGGTTG
BACOVA_03430	pET22b	GCTGAAGGATCCGAAAGTAGTGACAATGAATTCCTG	CATGAACTCGAGTTTTCTCACTACTTCGAGTGACTTAC
BACOVA_03431	pET21a	CGCGGATCCGACGAATATAATTGCCAGCTTC	CCCAAGCTTTTCAATCCGTCCACTATACCTTCATAC
BACOVA_03432	pET21a	CGCGGATCCTGTTCCGGAGGGGAAG	CCCAAGCTTAAATGTCAATCTGACTGAAGTTATACTG
BACOVA_03433	pET21a	CGCGGATCCTGTATTCTTGGTTCTGTAAAGATGAC	CCCAAGCTTTTTCTTTCGATAACAATGTTGTCGAG
BACOVA_03434	pET21a	CGCGGATCCGCTGCGGAACCATTTGTCACTTTTATATC	CCGCTCGAGATGCGTAACCTTTCGATAAGGACTTTC
BACOVA_03436	pRSET A	CGCGGATCCAACCCGATTGTGCAAACTTGC	CCCAAGCTTCTGTAAACTTGTACCAATCAAAGTTAAAC
BACOVA_03438	pET21a	CGCGGATCCGACCTGAAACTGTGGTATAGTC	CCCAAGCTTTGTTTTCTTCGATTGTATTTACAATATAGG
BACOVA_03449	pET28b	CGCGGAATTCATGGCTGAACAATTTGTAATTCACACCG	CTCCCGCTCGAGTCTACTTTCTCTTACTTTTCGATTTC
BACOVA_04385	pRSET A	CGCGGATCCGATGGAAGTGCCTTGTG	CCCAAGCTTCTTACTAAACCCATAAGTGAGCATTTC
BACOVA_04386	pET21a	CGCGGATCCAAAACAGAAAAAGATATTTAGTTCC	CCCAAGCTTTTCATCTTTTCCCTCGATAGTAATGATTTC
BACOVA_04387	pRSET A	CGCGGATCCGATGGAAGTTCGCTGAAAAAG	CCCAAGCTTTTTTGTTTTTTTCAGCCTTTTGATAATCAGG
BACOVA_04390	pET21a	CGCGGATCCATGGAGTGGTATAAGACCTAC	CCCAAGCTTTTCAAATCTCCGGTAAAGTCTC
BACOVA_04391	pET22b	GTTATAGGATCCGACGATAATGGCGGAAGTTCTGTG	CATGAACTCGAGTTCTGCATCCTCTTCTCGTCC
BACOVA_04392	pET22b	GCGCGGAATTCGCAAGTACTTCTCGATTCTG	GCGGGCTCGAGGTTAAACAATCGGTTATTCTGTCTTC

^apET vectors are from Novagen

^bRestriction site underlined

^cpRSET vector is from Life Technologies

Supplementary Table 10. Primers used for site directed mutagenesis.

Gene	Residue change	Forward primer	Reverse primer
BACOVA_03433	E501A	GCCTTCTGTGTGCCG <u>GCG</u> CACGGTCCCATCACGG	CCGTGATGGGACCGT <u>GCG</u> CCGGCGACACAGAAGGC
BACOVA_03433	T370H	CACCTGGGATAGCAAATAC <u>CAT</u> ATCAATATCAATACAG	CTGTATTGATATTGAT <u>ATG</u> GATTGCTATCCCAGGGTG
BACOVA_03438	E361A	TCTTGGTTTCAATTATTG <u>TGC</u> CAGTTTTGGGGTATGATG	CATCATACCCCAAACTGGG <u>CACA</u> ATAATTGAAACCAAGA
BACOVA_03438	D467A	TCAGTACGGCATCCGTTT <u>GCG</u> CAATGCGGTGGACAGAGG	TCTGTCCAGCCGATT <u>GCG</u> GAAACGGATGCCGTACT

^aBases changed are underlined

Supplementary Table 11. Primers used for qRT-PCR.

Gene	Forward primer	Reverse primer
BACOVA_03426	CCCTTCCATTAGCTCCAATTG	GCGACGGTGAATTGATGCTGATGA
BACOVA_03428	CATCTCCATATACTACGTTCTTTTGC	CGGAACTTTCATACGTGGACTTAGC
BACOVA_04393	ATTGTTTCGCCTGTCTGATTACTCT	ATGAACCGTAATAAAGTAAATCGCTGGA

Supplementary Table 12. Crystal data statistics and refinement details

Data statistics*	
BACOVA_03438 GH95	
Beamline	IO2
Date	22/06/13
Wavelength (Å)	0.9796
Resolution (Å)	49.31-2.81 (2.90-2.81)
Space group	P2 ₁ 2 ₁ 2 ₁
Unit-cell parameters	
a (Å)	53.69
b (Å)	179.1
c (Å)	205.2
α, β, γ (°)	90.0,90.0,90.0
Unit-cell volume (Å ³)	1973256
Solvent content (%)	53
No. of measured reflections	336786
No. of independent reflections	48329
Completeness (%)	97.6(74.5)
Redundancy	7.0(3.0)
Rmerge (%)	11.8(48.3)
<I>/<σ(I)>	10.8(2.0)
Refinement statistics*	
Rwork (%)	18.64
Rfree [#] (%)	22.80
No. of non-H atoms	
No. of protein atoms	12531
No. of ion atoms	13
No. of ligand atoms	24
R.m.s. deviation from ideal values	
Bond length (Å)	0.011
Bond Angle (°)	1.5
Average B factor (Å ²)	
Protein	43
Ligand	24
Ions	68
Ramachandran plot [†] , residues in allowed and most favoured regions (%)	99.75

*(Values in parenthesis are for the highest resolution shell).

[#]5% of the randomly selected reflections excluded from refinement.

[†]Calculated using MOLPROBITY.

Supplementary Table 13. Primers used to create PUL knockouts and tagged strains.

Primer name	Sequence
Δ PUL-XylS – left flank 1	GCGG TCGAC AGGCTTCACGTTCCATTTAGG ^A
Δ PUL-XylS – left flank 2	ATAAAGATATTCCTTCTACGGAAC
Δ PUL-XylS – right flank 1	GCG TCTAG AGGCTTTCTCGCCGGAGTT
Δ PUL-XylS – right flank 2	CTGTGTGGAAAAAGGAGAAAGG
Δ PUL-XylL – left flank 1	GCGG TCGAC CCGGTATGATTGGTATCGGTGC
Δ PUL-XylL – left flank 2	GTTTTTTTATAACTGGCTACATTATC
Δ PUL-XylL – right flank 1	GCG TCTAG AAAAGATACAGTGATTGGCATTGC
Δ PUL-XylL – right flank 2	GAGGATGGGAAGAGCTTAC
Tag11 F	ATG TCG CCA ATT GTC ACT TTC TCA
Tag 1 F	ATG CCG CGG ATT TAT TGG AAG AAG
Universal Reverse	CAC AAT ATG AGC AAC AAG GAA TCC
ATT1 F	CCT TTG CAC CGC TTT CAA CG
ATT1 R	TCA ACT AAA CAT GAG ATA CTA GC

^aBold are restriction sites; either Sall or XbaI for cloning into pExchange

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