a

	Vertical (first)	Horizontal (second)	
	W50 *porU*	W50 *porU*	-
	381 $porU_{C690A}$	W50 *porU*	-
	W50 *porV*	W50 *porU*	-
	W50 *porT*	W50 *porU*	-
	33277 *porQ*	W50 *porU*	-
	W50 *porZ*	W50 *porU*	-
	W50 *wbaP*	W50 *porU*	+

	Vertical (first)	Horizontal (second)	
	W50 *porU*	W50 *porU*	-
	W50 *porU*	381 $porU_{C690A}$	-
	W50 *porU*	W50 *porV*	-
	W50 *porU*	W50 *porT*	-
	W50 *porU*	33277 *porQ*	-
	W50 *porU*	W50 *porZ*	-
	W50 *porU*	W50 *wbaP*	+

b

	Vertical (first)	Horizontal (second)	
	W50 *porU*	381 $porU_{C690A}$	-
	381 $porU_{C690A}$	381 $porU_{C690A}$	-
	W50 *porV*	381 $porU_{C690A}$	-
	W50 *porT*	381 $porU_{C690A}$	-
	33277 *porQ*	381 $porU_{C690A}$	-
	W50 *porZ*	381 $porU_{C690A}$	-
	W50 *wbaP*	381 $porU_{C690A}$	+

	Vertical (first)	Horizontal (second)	
	381 $porU_{C690A}$	W50 *porU*	-
	381 $porU_{C690A}$	381 $porU_{C690A}$	-
	381 $porU_{C690A}$	W50 *porV*	-
	381 $porU_{C690A}$	W50 *porT*	-
	381 $porU_{C690A}$	33277 *porQ*	-
	381 $porU_{C690A}$	W50 *porZ*	-
	381 $porU_{C690A}$	W50 *wbaP*	+

c

	Vertical (first)	Horizontal (second)	
	W50 *porU*	W50 *porV*	-
	381 $porU_{C690A}$	W50 *porV*	-
	W50 *porV*	W50 *porV*	-
	W50 *porT*	W50 *porV*	-
	33277 *porQ*	W50 *porV*	-
	W50 *porZ*	W50 *porV*	-
	W50 *wbaP*	W50 *porV*	+

	Vertical (first)	Horizontal (second)	
	W50 *porV*	W50 *porU*	-
	W50 *porV*	381 $porU_{C690A}$	-
	W50 *porV*	W50 *porV*	-
	W50 *porV*	W50 *porT*	-
	W50 *porV*	33277 *porQ*	-
	W50 *porV*	W50 *porZ*	-
	W50 *porV*	W50 *wbaP*	+

d

	Vertical (first)	Horizontal (second)	
	W50 *porU*	W50 *porT*	-
	381 $porU_{C690A}$	W50 *porT*	-
	W50 *porV*	W50 *porT*	-
	W50 *porT*	W50 *porT*	-
	33277 *porQ*	W50 *porT*	-
	W50 *porZ*	W50 *porT*	-
	W50 *wbaP*	W50 *porT*	+

	Vertical (first)	Horizontal (second)	
	W50 *porT*	W50 *porU*	-
	W50 *porT*	381 $porU_{C690A}$	-
	W50 *porT*	W50 *porV*	-
	W50 *porT*	W50 *porT*	-
	W50 *porT*	33277 *porQ*	-
	W50 *porT*	W50 *porZ*	-
	W50 *porT*	W50 *wbaP*	+

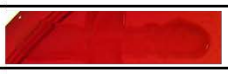
e

	Vertical (first)	Horizontal (second)	
	W50 *porU*	33277 *porQ*	-
	381 $porU_{C690A}$	33277 *porQ*	-
	W50 *porV*	33277 *porQ*	-
	W50 *porT*	33277 *porQ*	-
	33277 *porQ*	33277 *porQ*	-
	W50 *porZ*	33277 *porQ*	-
	W50 *wbaP*	33277 *porQ*	**W**

	Vertical (first)	Horizontal (second)	
	33277 *porQ*	W50 *porU*	-
	33277 *porQ*	381 $porU_{C690A}$	-
	33277 *porQ*	W50 *porV*	-
	33277 *porQ*	W50 *porT*	-
	33277 *porQ*	33277 *porQ*	-
	33277 *porQ*	W50 *porZ*	-
	33277 *porQ*	W50 *wbaP*	**W**

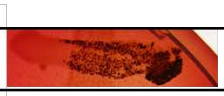
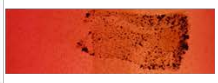
f

	Vertical (first)	Horizontal (second)	
	W50 *porU*	W50 *porZ*	-
	381 $porU_{C690A}$	W50 *porZ*	-
	W50 *porV*	W50 *porZ*	-
	W50 *porT*	W50 *porZ*	-
	33277 *porQ*	W50 *porZ*	-
	W50 *porZ*	W50 *porZ*	-
	W50 *wbaP*	W50 *porZ*	**W**

	Vertical (first)	Horizontal (second)	
	W50 *porZ*	W50 *porU*	-
	W50 *porZ*	381 $porU_{C690A}$	-
	W50 *porZ*	W50 *porV*	-
	W50 *porZ*	W50 *porT*	-
	W50 *porZ*	33277 *porQ*	-
	W50 *porZ*	W50 *porZ*	-
	W50 *porZ*	W50 *wbaP*	**W**

g

	Vertical (first)	Horizontal (second)	
	W50 *porU*	W50 *wbaP*	+
	381 $porU_{C690A}$	W50 *wbaP*	+
	W50 *porV*	W50 *wbaP*	+
	W50 *porT*	W50 *wbaP*	+
	33277 *porQ*	W50 *wbaP*	W
	W50 *porZ*	W50 *wbaP*	W
	W50 *wbaP*	W50 *wbaP*	-

	Vertical (first)	Horizontal (second)	
	W50 *wbaP*	W50 *porU*	+
	W50 *wbaP*	381 $porU_{C690A}$	+
	W50 *wbaP*	W50 *porV*	+
	W50 *wbaP*	W50 *porT*	+
	W50 *wbaP*	33277 *porQ*	W
	W50 *wbaP*	W50 *porZ*	W
	W50 *wbaP*	W50 *wbaP*	-

Supplementary Fig. 1 Reciprocal cross overs of two strains

First strain was applied vertically and the second strain applied in one horizontal motion from left to right on BHI-T BA plate and incubated 14 days anaerobically. Reciprocal crosses between (**a**) W50 *porU* and one of seven strains (W50 *porU*, 381 $porU_{C690A}$, W50 *porV*, W50 *porT*, 33277 *porQ*, W50 *porZ*, W50 *wbaP*); (**b**) 381 $porU_{C690A}$; (**c**) W50 *porV*; (**d**) W50 *porT*;(**e**) 33277 *porQ*; (**f**) W50 *porZ*; (**g**) W50 *wbaP*. +, obvious pigmentation; W, weak pigmentation; -, no pigmentation.

```
RgpB_33277      MKKNFSRIVSIVAFSSLLGGMAFAQPAERGRNPQVRLLSAEQSMSKVQFRMDNLQFTDVQ 60
RgpB_381        MKKNFSRIVSIVAFSSLLGGMAFAQPAERGRNPQVRLLSAEQSMSKVQFRMDNLQFTDVQ 60
                ************************************************************

RgpB_33277      TSKGVAQVPTFTEGVNISEKGTPILPILSRSLAVSETRAMKVEVVSSKFIEKKDVLIAPS 120
RgpB_381        TSKGVAQVPTFTEGVNISEKGTPILPILSRSLAVSETRAMKVEVVSSKFIEKKDVLIAPS 120
                ************************************************************

RgpB_33277      KGVISRAENPDQIPYVYGQSYNEDKFFPGEIATLSDPFILRDVRGQVVNFAPLQYNPVTK 180
RgpB_381        KGVISRAENPDQIPYVYGQSYNEDKFFPGEIATLSDPFILRDVRGQVVNFAPLQYNPVTK 180
                ************************************************************

RgpB_33277      TLRIYTEIVVAVSETAEAGQNTISLVKNSTFTGFEDIYKSVFMNYEATRYTPVEEKENGR 240
RgpB_381        TLRIYTEIVVAVSETAEAGQNTISLVKNSTFTGFEDIYKSVFMNYEATRYTPVEEKENGR 240
                ************************************************************

RgpB_33277      MIVIVAKKYEGDIKDFVDWKNQRGLRTEVKVAEDIASPVTANAIQQFVKQEYEKEGNDLT 300
RgpB_381        MIVIVAKKYEGDIKDFVDWKNQRGLRTEVKVAEDIASPVTANAIQQFVKQEYEKEGNDLT 300
                ************************************************************

RgpB_33277      YVLLVGDHKDIPAKITPGIKSDQVYGQIVGNDHYNEVFIGRFSCESKEDLKTQIDRTIHY 360
RgpB_381        YVLLVGDHKDIPAKITPGIKSDQVYGQIVGNDHYNEVFIGRFSCESKEDLKTQIDRTIHY 360
                ************************************************************

RgpB_33277      ERNITTEDKWLGQALCIASAEGGPSADNGESDIQHENVIANLLTQYGYTKIIKCYDPGVT 420
RgpB_381        ERNITTEDKWLGQALCIASAEGGPSADNGESDIQHENVIANLLTQYGYTKIIKCYDPGVT 420
                ************************************************************

RgpB_33277      PKNIIDAFNGGISLVNYTGHGSETAWGTSHFGTTHVKQLTNSNQLPFIFDVACVNGDFLF 480
RgpB_381        PKNIIDAFNGGISLVNYTGHGSETAWGTSHFGTTHVKQLTNSNQLPFIFDVACVNGDFLF 480
                ************************************************************

RgpB_33277      SMPCFAEALMRAQKDGKPTGTVAIIASTINQSWASPMRGQDEMNEILCEKHPNNIKRTFG 540
RgpB_381        SMPCFAEALMRAQKDGKPTGTVAIIASTIDQYWAPPMRGQDEMNEILCEKHPNNIKRTFG 540
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RgpB_33277      GVTMNGMFAMVEKYKKDGEKMLDTWTVFGDPSLLVRTLVPTEMQVTAPANISASAQTFEV 600
RgpB_381        GVTMNGMFAMVEKYKKDGENMLDTWTVFGDPSLLVRTLVPTEMQVTAPANISASAQTFEV 600
                ********************:***************************************

RgpB_33277      ACDYNGAIATLSDDGDMVGTAIVKDGKAIIKLNESIADETNLTLTVVGYNKVTVIKDVKV 660
RgpB_381        ACDYNGAIATLSDDGDMVGTAIVKDGKAIIKLNESIADETNLTLTVVGYNKVTVIKDVKV 660
                ************************************************************

RgpB_33277      EGTSIADVANDKPYTVAVSGKTITVESPAAGLTIFDMNGRRVATAKNRMVFEAQNGVYAV 720
RgpB_381        EGTSIADVANDKPYTVAVSGKTITVESPAAGLTIFDMNGRRVATAKNRMVFEAQNGVYAV 720
                ************************************************************

RgpB_33277      RIATEGKTYTEKVIVK  736
RgpB_381        RIATEGKTYTEKVIVK  736
                ****************
```

Supplementary Fig. 2 Alignment of RgpB protein sequences from strains 33277 and 381

Protein sequence alignment for RgpB from 33277 and 381 strains was generated by the Clustal Omega program from EMBL-EBI Web Services (https://www.ebi.ac.uk/Tools/msa/clustalo/). Yellow highlighted amino acids indicate sequence mismatches between the two strain sequences. Underlined peptides represent strain-specific RgpB tryptic peptides (three for 33277 and two for 381).

Supplementary Table 1 Unique peptide sequences for the same regions of RgpB between 33277 and 381 strains[a]

Peptide ID[b]	Sequence[c]	RgpB		RgpA	
		33277	381	33277	381
1	K/DGEK/MLDTWTVFGDPSLLVR	x		x	x
1*	K/DGEN MLDTWTVFGDPSLLVR		x		
9	K/DGKPTGTVAIIASTINQSWASPMR	x		x	x
9*	K/DGKPTGTVAIIASTIDQYWAPPMR		x		

[a]A "x" represents that the peptide is present.

[b]Peptide ID numbers "1" and "9" correspond to the RgpA/B peptides shown in **Fig. 5**. Numbers with an asterisk are peptides unique to RgpB from strain 381.

[c]Highlighted amino acid residues indicate a mismatch in the same region of RgpB between 33277 and 381 strains.