Supplementary File S1.

Analysis of ompF, ompC and pldA expression

One of the mechanisms of OM permeability changing relies on a reciprocal expression of major porins, OmpF and OmpC, which is also regulated by many environmental factors [Mahendran et al., 2010; Moya-Torres et al., 2014; Novikova et al., 2011]. Therefore, expression of the *ompC* and *ompF* genes as well as the *pldA*, encoded membrane-bound phospholipase A in *Y. pseudotuberculosis* cells was studied in response to glucose and PEBH exposure. Expression of genes encoding porins OmpF, OmpC and phospholipase A in *Y. pseudotuberculosis* 488 was studied by qRT-PCR. Total RNA was isolated from late-exponential phase bacterial cultures (OD600 from1.1 to 1.5) using Aurum total RNA mini kit (Bio-Rad) according to the manufacturer's instructions. An additional DNase treatment was carried out with RNase-free DNase (Thermo Scientific) to remove any remaining genomic DNA. The RNA concentrations and purity were determined by electrophoresis and spectrophotometry at 260 nm. Purified total RNA (2 μg) were reverse transcribed into cDNA with MMLV RT Kit (Evrogen) and random hexamer primers (Evrogen) in accordance with the manufacturer's protocol. The cDNAs were subsequently used to quantify the relative level of *ompF*, *ompC* and *pldA* by qRT-PCR in a Light Cycler 96 (Roche). 16S rDNA gene was chosen as the endogenous reference for relative quantification. Gene-specific primers used in this experiment are shown in Table 1. Primer specificity and efficiency of amplification were validated as described previously [Livak and Schmittgen, 2005].

All RT-PCRs were carried out with HS GoTaq Polymerase (Promega) and dye Eva Green (Biotium) in accordance with the protocols. The following thermal cycling parameters were used for reaction: initial denaturation at 95 °C for 8 min; and 40 cycles of amplification and quantitation at 95 °C for 15 s, 55 °C for 10 s and 72 °C for 20 s followed by fluorescence reading. A melting curve was drawn at the end of the 40 cycles to evaluate the specificity of the PCR. To detect DNA contamination in the purified RNA, negative control reactions without reverse transcriptase were analyzed along with a «no template control» consisting of nuclease-free water instead of cDNA. All amplifications were carried out in duplicate from two different RNA preparations. Quantification for each target gene expression was determined by the $2^{-\Delta\Delta CT}$ method as described [Livak and Schmittgen, 2005] using the housekeeping gene, 16S rDNA.

Analysis of variance (ANOVA) was used to determine whether glucose or polyphenol had a statistically significant effect on the expression of porin *Y. pseudotuberculosis*. A P-value of less than 0.05 was considered significant. ANOVA was performed using Statistics (Tulsa, USA, 2008).

References

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Mahendran KR, Kreir M, Weingart H, Fertig N, Winterhalter M: Permeation of antibiotics through *Escherichia coli* OmpF and OmpC porins: screening for influx on a single-molecule level. J Biomol Screen 2010; 15:302–307.

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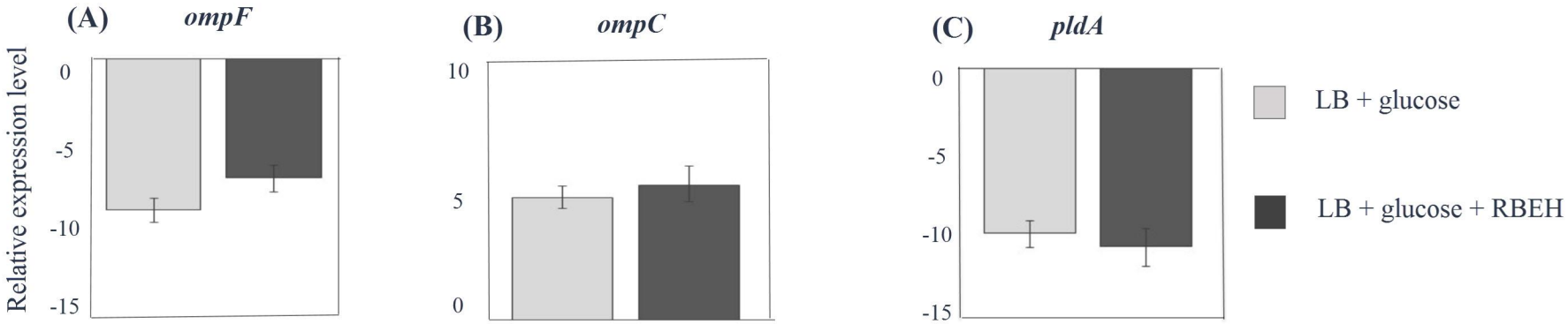
Supplementary Table S1. Primers used for RT-PCR analysis of *ompF, ompC* and *pldA* expression

Gene	Forward primer	Reverse primer
ompF	5’ ATGAATCACCACCGAACACT 3’	5’ CAAGACGGCAACGCAAC 3’
ompC	5’ TGACAGGAAGTTATCAGCACC 3’	5 ‘GACGGGAACCACGACAGT 3’
pldA	5’ CTTTCCCTATCTGGCGTGGT 3’	5 ‘GCTGTGGCTCGTAGTTGGTTT 3’
16S rDNA	5’ CTTGATTTCCCACCATTACG 3’	5’ ATTTAGCCGAGATGCTTTAG 3’



Supplementary Figure S1. Relative expression level of *ompF* (A), *ompC* (B) and *pldA* (C) in *Y. pseudotuberculosis* cells grown in glucose containing medium supplemented with and without PBEH. Transcription of target genes in culture grown in LB medium without glucose and PBEH was used as a control. Expression levels are shown as fold change compared to average expression level in control group. Columns represent the mean values of duplicate measurements from two different experiments ± standard deviation. All target genes had significant difference of expression level compared to control ($P < 0.05$) qRT-PCR analysis of the relative changes in expression of *ompF* (A), *ompC* (B) and *pldA* (C) in *Y. pseudotuberculosis* cells grown in glucose containing medium supplemented with and without PBEH.