Supplementary Materials

Characterisation of a Glucosamine/Glucosaminide *N*-Acetyltransferase of *Clostridium acetobutylicum*

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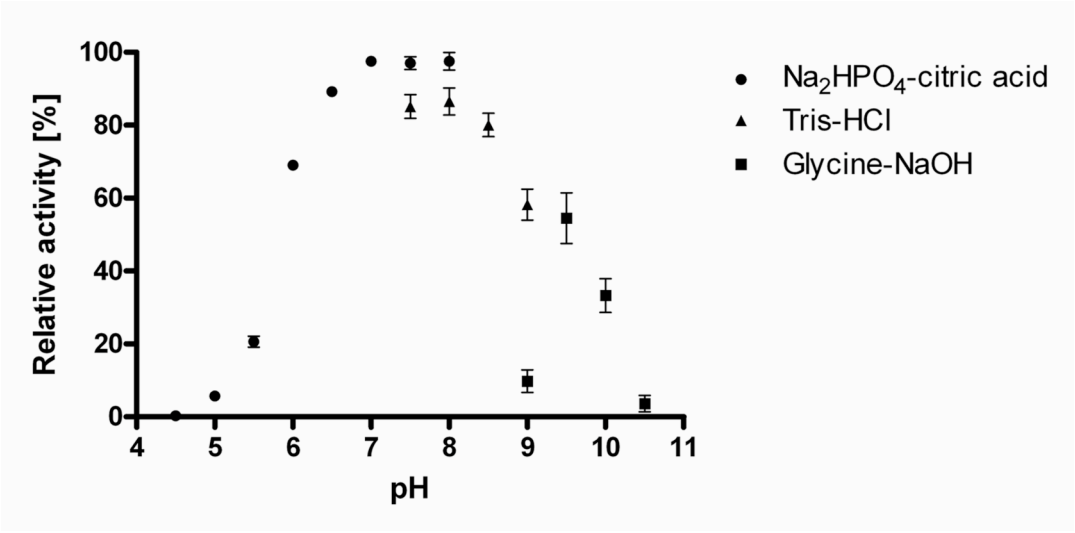

Fig. S1. The pH-activity profile of GlmA for the acetylation of GlcN with AcCoA. The enzyme activity was quantified by using radioactively labeled [acetyl-1-^{14}C]-CoA in the indicated buffers at a pH ranging from 4.5 to 10.5 (for details see Materials and Methods). Data are mean ± SE (n = 4).

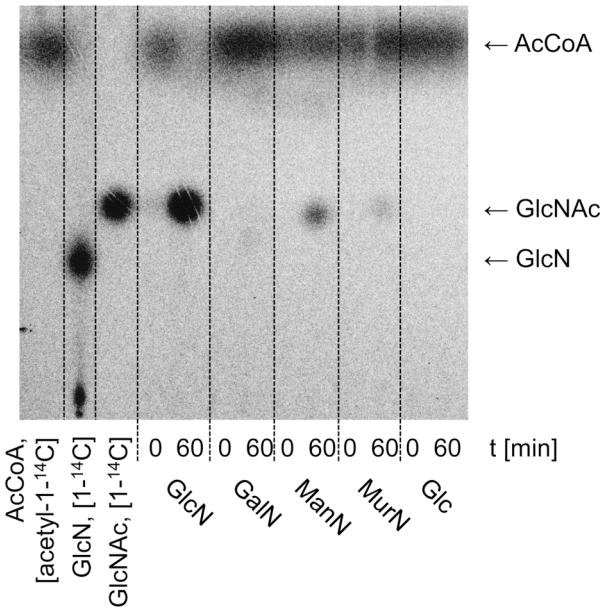

Fig. S2. GlcN is the preferred substrate of GlmA. An Acetylation of GalN and glucose (Glc) were observed in the assay using radioactively labeled [Acetyl-1-^{14}C]-CoA. The low amount of conversion with ManN and MurN may be due to GlcN contaminations in the chemicals since in

both cases a spot that runs like GlcNAc was detected and ManNAc as well as MurNAc would run different from that on TLC.

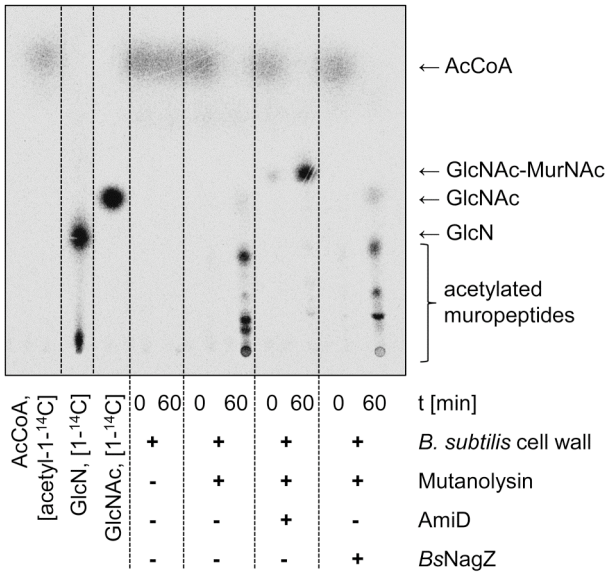

Fig. S3. Cell wall prepared from *B. subtilis* was degraded with the purified autolysins mutanolysin, AmiD and/or *Bs*NagZ. Terminal non-reducing GlcN residues of mutanolysin formed muropeptides had to be firstly acetylated by GlmA before the *N*-acetylglucosaminidase *Bs*NagZ is able to cleave it (compare with Fig. 7). Here, *Bs*NagZ was not added subsequently to the reaction mixture containing GlmA.

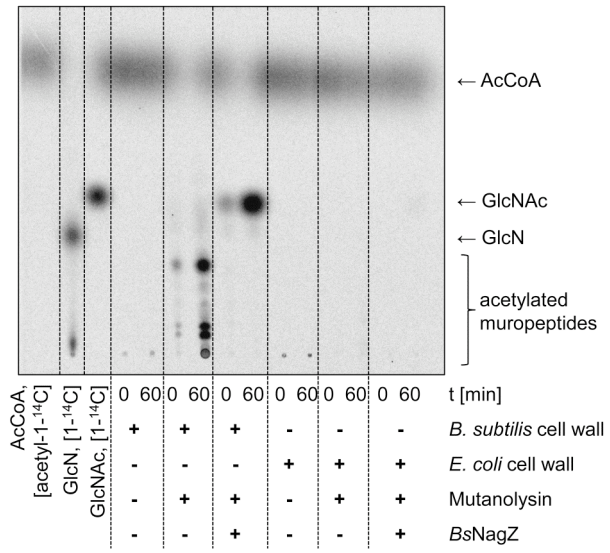

Fig. S4. Peptidoglycan fragments of *B. subtilis* containing deacetylated GlcN residues are a substrate for GlmA but not such fragments of *E. coli* that lack this modification. Radioactive

acetylation with GlmA using [acetyl-1-^{14}C]-CoA was conducted with cell wall preparations from *B. subtilis* and *E. coli* that were previously degraded with mutanolysin and/or *Bs*NagZ.

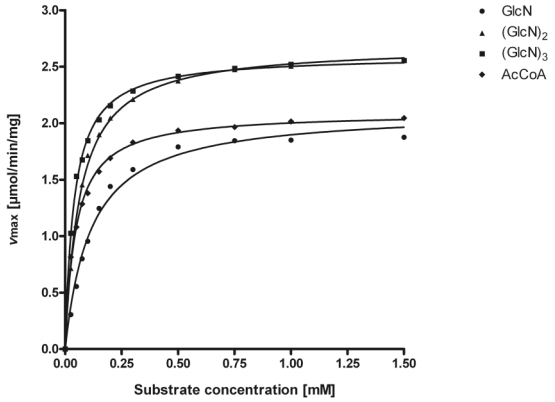

Fig. S5. Kinetics for GlmA with GlcN, chitobiose and chitotriose. The kinetic values of GlmA were determined by nonlinear regression. GlmA revealed a slightly lower v_{max} value for GlcN compared to the chitosans $(GlcN)_2$ and $(GlcN)_3$. The calculated K_m value of GlcN was about two to three fold higher than for chitosans consisting of more than one GlcN residue (cf. table 1). Data are mean of two independent experiments. Standard errors were below 5%.