

# Crystal Structure of the *Bacillus subtilis* GlvA Protein

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## Introduction

The National Institutes of Health (NIH) Protein Structure Initiative is exploring the range of biologically important protein structures. As a part of the Midwest Center for Structural Genomics, we are determining representative structures of members of evolutionarily conserved sequence families. These structures also furnish insight into the biological functions of these proteins. The *Bacillus subtilis* GlvA protein is a member of family GH4 of the glycoside hydrolases. It catalyzes the hydrolysis of phosphorylated, alpha-linked glucosyl disaccharides.

## Methods and Materials

Crystals of the seleno-methionine substituted GlvA protein were transferred to a mixture of Paratone-N and paraffin oil. Excess mother liquor around the crystals was removed, and the crystals were cooled to 100K in a cold nitrogen stream. The protein was crystallized in space group I222 with cell dimensions of  $a = 82.6 \text{ \AA}$ ,  $b = 102.0 \text{ \AA}$ , and  $c = 144.8 \text{ \AA}$ . X-ray diffraction data were collected at 100K at DND-CAT beamline station 5-ID-B station at the APS with a Mar165 charge-coupled device (CCD) detector. A single-wavelength anomalous diffraction (SAD) data set was collected near the peak wavelength of the Se absorption edge (0.979 Å). The data were integrated and merged with DENZO/SCALEPACK. The 2.05-Å resolution data set was used for both phasing and refinement, as is described in more detail elsewhere [1].

## Results and Discussion

The Se atom positions were determined from the SAD data by using the SOLVE program. The initial phases were then improved, and an initial, partial interpretation of the electron density map was carried out by using the program RESOLVE. The structure of GlvA revealed a surprising degree of similarity to the hydroxy-acid dehydrogenases, such as lactate or malate dehydrogenases. The structure reveals the presence of NAD, Mn, and glucose 6-phosphate ligands copurified and crystallized with the protein.

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## References

[1] S.S. Rajan, X. Yang, F. Collart, V.L.Y. Yip, S.G. Withers, A. Varrot, J. Thompson, G.J. Davies, and W.F. Anderson (submitted for publication, 2003).