

## Structural analysis of Dps from *Staphylococcus aureus*

Yoshikazu Tanaka<sup>1</sup>, Min Yao<sup>1</sup>, and Nobuhisa Watanabe<sup>1</sup>

<sup>1</sup>Division of Biological Sciences, Graduate School of Science, Hokkaido University, Sapporo 001-0021, Japan

### Introduction

Iron is an essential cofactor of many enzymes. Although iron is abundant in the world, it is easily oxidized to  $\text{Fe}^{3+}$ , which is insoluble and inactive form for living cells, under physiological pH and oxidizing conditions. Then, the cellular uptake system of iron in organism is well developed to acquire enough amount of active form of iron from environment.

When pathogenic bacteria infects human, they must acquire iron from host cells, and that causes sepsis. Then, factors relating to acquisition of iron might be important target for drug design. Dps is thought to act as a storage protein of iron. Interestingly Dps is also thought to relate to DNA protection from oxidative damage. In this study, we have solved the structure of Dps to elucidate its biological importance from a structural viewpoint.

### Experiments and Results

Dps was expressed in *E. coli* B21(DE3) grown in LB medium, and purified by Ni-NTA affinity chromatography and size exclusion chromatography. Crystals of Dps suitable for X-ray diffraction analysis were grown in 100 mM Tris-HCl (pH7.0), 200 mM magnesium chloride, and 10% polyethylene glycol 8000.

X-ray diffraction data were collected at the beamline BL-6A of Photon Factory, under cryogenic condition (100 K) using flash cooling technique. Crystal was soaked into the crystallization buffer containing 30% glycerol for 1 min. before flash cooling. The dataset up to a resolution of 1.85 Å was collected and processed with HKL2000 program suite. The data processing statistics are given in Table 1. The crystals belong to the space group  $P321$  with unit cell parameters  $a = b = 143.4$  Å, and  $c = 88.3$  Å. The crystal structure of Dps was determined by molecular replacement method using the structure of Dlp1 from *Bacillus anthracis* (1JI5) as a search probe. The asymmetric unit contains four molecules of Dps, corresponding to the  $V_M$  value of  $3.67 \text{ \AA}^3 \text{ Da}^{-1}$ . Monomeric structure of Dps is a four helix bundle structure and four molecules which located in an asymmetric unit are crystallographically assemble into dodecamer.

\*yoshikazu@castor.sci.hokudai.ac.jp



Figure 1 Crystals of Dps

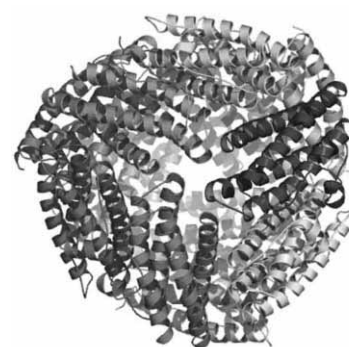


Figure 2 Dodecamer structure of Dps

Table 1. Data processing statistics

Space group	$P321$
Cell dimensions (Å)	$a=b= 143.4, c= 88.3$
Beamline	BL-6A
Resolution (Å)	50-1.85(1.92-1.85)
Wavelength (Å)	0.9798
$R_{\text{sym}}$ (%)	6.4 (28.0)
Completeness (%)	100 (100)
Observed reflections	2059803
Unique reflections	89128
Multiplicity	11.2(10.6)