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SAXS study of capsid assembly of bacteriophage T5

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1 Introduction

Capsids of the tailed double-stranded DNA (dsDNA) bacteriophages assemble in a tightly regulated process involving hundreds of copies of a major capsid protein organized in a regular repeating pattern of hexameric and pentameric capsomers on an icosahedrally symmetric lattice.

Despite the vast array of icosahedral capsid morphologies observed, including prolate icosahedral geometries, every capsid of the tailed dsDNA bacteriophage family detailed to date appears to exploit a common fold—that first described for phage HK97 (Wikoff W.R. et al. (2000) Science, 289, 2129-2133; Helgstrand C. et al. (2003) J. Mol. Biol., 334, 885-899). The study of the principles guiding assembly of these structurally related subunits into different but specific icosahedral geometries with high fidelity as well as the identification of sequential stages of the capsid assembly are the challenges of modern structural biology. We developed procedure for mutant phages production to freeze some steps of T5 capsid assembly and here present SAXS pattern of native capsid and mutant one.

2 Experimental

We obtained mutants of phage T5, which allow us to isolate the phage T5 capsids at different stages of assembly. The filled capsids (wt and Δdec) were purified by centrifugation in a CsCl gradient, and the empty capsids (php_S122A, php_mut1-2/F, php_S122am and dec_am1) were purified by centrifugation in a gradient of glycerol (10-40%; v/v). Synchrotron X-ray measurements were done on a small-angle camera BL-6A (Photon Factory, Tsukuba) using PILATUS 100K detector. The range of scattering vectors Q=0.01-0.25 $Å^{-1}$.

3 Results

Scattering patterns for filled capsid (wt and Δdec) and empty capsids (php_S122A, php_mut1-2/F, php_S122am, dec_am1) are shown in Fig.1. One can see well ordered structure of full capsid and great disturbance of this structure in the case of empty ones. The values of radius of gyration (R_g) were calculated from the initial part of scattering curve and was found to be in average 46 nm for full capsid and 36 nm for empty one. It means that empty capsid is essentially smaller the full one. We plan to recover the structure from scattering patterns and obtain more detailed information about structural changes.

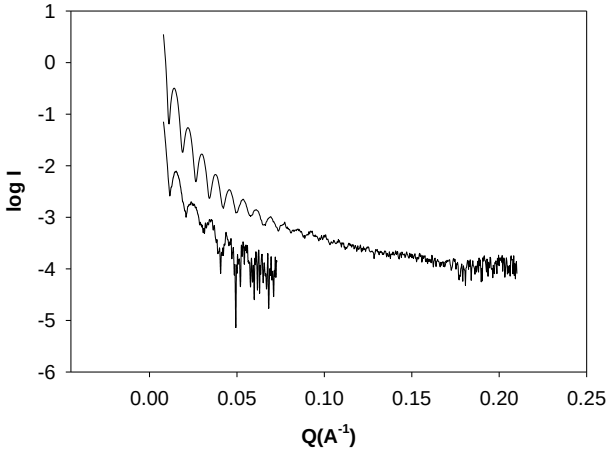

Fig.1 The dependence log I versus Q for capsids wt (upper) and php_S122A (below).

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