

# METHODS IN MOLECULAR BIOLOGY

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# Protein Cages

## Methods and Protocols

Edited by

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## Preface

In recent years, there has been a surge in research focused on proteins that form cage-like structures. While these multicomponent proteins have been of great interest to scientists studying protein folding and structural biology, they have also posed one of the next big challenges in protein design and engineering. Furthermore, because of their often nano-scaled size, they have caught the eye of materials scientists, and their interior cavities have been exploited by chemists and enzymologists to explore the possibility of enhancing reactivity by means of confinement. They also have been used to develop bioconjugation chemical reactions, some applications of which have been for the presentation of ligands multivalently and to attach directing functionality in order to control the delivery of drugs or imaging agents encapsulated inside the cage. Protein cages are currently inspiring diverse scientific disciplines and are therefore at the crossroads of extremely widely scoped research.

This volume emphasizes the geographic and scientific diversity of the field by highlighting the expertise of researchers based on three continents who are developing new techniques to help understand protein cages and to apply them to a variety of technologies. Following a nanomaterials direction, the chapter written by Tsvetkova and Dragnea provides procedures for the assembly of virus capsids around nanoparticles, whereas Uchida et al. describe methods to generate iron particles inside ferritin and organic polymers inside capsids. In addition, Pulsipher and Dmochowski present procedures for growing gold nanoparticles inside ferritins and assembling the proteins around the particles, and Sana and Lim describe how to measure relaxivities of protein cage encased iron nanoparticles. Protein cages have been the focus of protein design endeavors, and Ardejani and Orner describe the use of simple computational strategies to engineer more stable ferritins. As the field matures, developing improved methods for the production of these proteins is especially important, and the chapter by Rurup et al. describes how to isolate encapsulin proteins, and Lassila details procedures for the purification of bacterial microcompartments. Investigation into biochemical and physical properties of protein cages is also a large slice of this field and the development of novel approaches is required due to the unique challenges associated with this class of proteins. Cornell and Orner describe a technique to identify specific oligomerization states of protein cages by designing binding sites for a fluorescent reagent at key protein–protein interfaces, whereas Grove et al. present techniques to determine the role structural metal has on the assembly of a miniferritin, and Zhang and Ardejani describe how to apply Differential Scanning Calorimetry to the assembly of ferritins. Van Rosmalen et al. review the use of Atomic Force Microscopy to probe the structure and mechanical properties of protein cages, and Gibbons et al. present a theoretical model to understand the deformation and indentation of hollow protein nanostructures.

The chapters herein collectively provide an introduction to the rich world of protein cage research and specific techniques to understand and exploit this fascinating class of proteins. Moreover, it is hoped that this volume will help to inspire and further propel the current multidisciplinary enthusiasm in studying and discovering new applications for protein cages.

*London, UK*

*Brendan P. Orner*



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