BESPA: Software Tools for Three-Dimensional Structure Reconstruction from Single Particle Images of Proteins

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1 Introduction

Single particle analysis is a straightforward method for structural studies of protein and biological macromolecules developed in image analysis on electron microscopy [1]. It is a predominant structural probe for proteins which decline to crystallize and exceed the limit in molecular weight for NMR analysis. Furthermore, it allows to examine structural changes and molecular complex of proteins at work, which will become a great advantage for the next generation in structural biology after static structures of proteins are all determined.

Our system, named BESPA (Backplane for Electron microscopic Single Particle Analysis) will be packaged soon featuring new algorithms and techniques together with practically useful programs. The single particle analysis has also raised topics for a computational science both in practical algorithm and in establishment of theoretical background.

2 Software System

Fig. 1 gives an overview of computational tasks for the single particle analysis in our system.

For statistically satisfactory results and resolution improvements, more than orders of thousands of particles are required in analysis. However, the automatic pick-up of particle with existing algorithms still depends on the quality of images and the experimental conditions. Besides computational trials for new algorithm, we developed a user-friendly interactive program to pick up particles from large micrographs. It is an editor for the particle coordinates that allows us to keeps track of their raw images throughout analysis.

In the two dimensional image analysis, averaging images in the same orientation is the critical task to obtain a correct result. Since the signal to noise ratio often reaches to unity, the classification fails very easily in existing method. We are improving our robust method for clustering the observed particle images without references. Classification was achieved in a bottom-up manor and an iterative alignment method was employed to derive class averages as characteristic views.

Our three dimensional reconstruction is based on the general back projection framework from observed projections of object as a computational tomography. The initial three dimensional model is build by the estimated Euler angle for the obtained characteristic using the common line theory. Bootstrapping from this initial model, the assignment of angles are refined through an iterative alignment to the re-projection of the model.

There are two major software packages available for general single particle analysis, SPIDER [2] and IMAGIC [3]. Both systems are confirmed traditional foundation of computations, but their

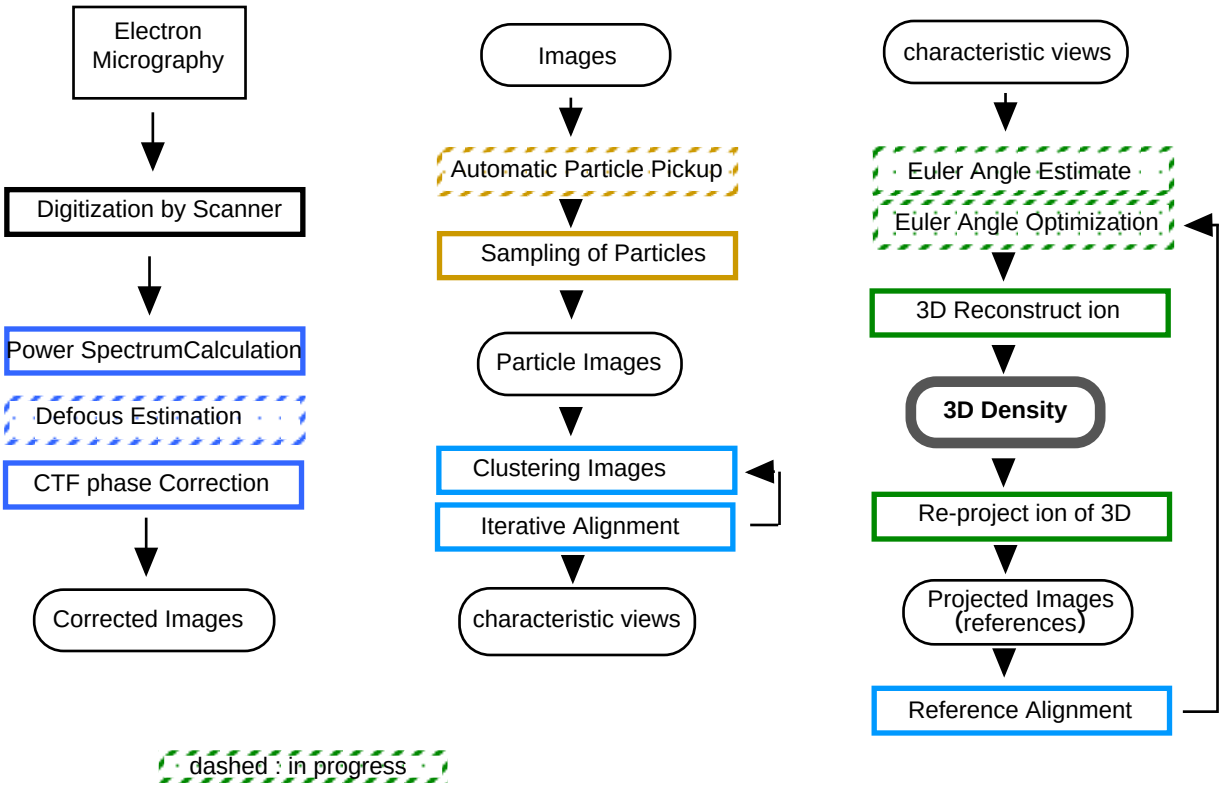

Figure 1: Overview of single particle analysis.

legacy programming environment with FORTRAN is inadequate for young scientists to implement new algorithms in modern computational techniques.

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