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MAID: A NEW SOFTWARE ROUTINE THAT AUTOMATES THE FITTING OF PROTEIN ELECTRON DENSITY MAPS

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The approach used by MAID is basically an automation of the steps that a skilled investigator would use in the classical fitting approach. It requires, as input, just the map and the amino acid sequence and it outputs the final structure without any user intervention. The completeness of the final structure is relatively independent of the map resolution. The program was tested on the unaveraged 2.5 Å selenomethionine multiple wavelength anomalous dispersion electron density map that was originally used to solve the structure of the 291 residue protein human heart short chain L-3-hydroxyacyl-CoA dehydrogenase. Inputting just the map density and the amino acid sequence, MAID fit 80 percent of the residues with a r.m.s.d. error of 0.43 Å for the main chain atoms and 1.0 Å for all atoms, without any user intervention. When tested on a higher quality 1.9 Å SMAD map, MAID correctly fit 100 percent of the residues. A major advantage of the MAID fitting procedure is that it maintains ideal bond lengths and angles and constrains φ-ς angles to the appropriate Ramachandran regions. The program compiles and runs on most Linux and Unix compilers. It is freely available at www.msi.umn.edu/(tilda)levitt.

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USE OF XENON IN PRACTICE

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Xenon derivatisation is classically performed with hydrophilic additives (e.g. Glycerol) to prevent nucleation of ice when the cryoprotectant buffer containing the crystal is flash cooled. The difficulties of obtaining a suitable cryoprotectant buffer, in which crystals are stable for a period of several minutes, are, however, rarely reported. Crystals of porcine pancreatic elastase (PPE) have been used to demonstrate that cryoprotection using dry paraffin oil [1] or panjellytm [2] allows with equal or better efficacy derivatisation under a xenon atmosphere prior to shock cooling [3]. Although the xenon k or l-edges are not readily accessible, the anomalous signal of xenon is appreciable even at remote energies. Phasing procedures for xenon derivatised PPE crystals, using data sets measured at three wavelengths easily accessible on a tunable beamline, have been investigated. The importance of highly redundant data in measuring precise anomalous differences is demonstrated and it is shown that an siras procedure yields a better phase set than those generated by sas or pseudo-mad procedures. Crystals derivatised by soaking in bromide solutions have been subsequently derivatised under a xenon atmosphere. Intensity data collected below the bromine absorption edge is used to determine the xenon position and the resultant phase information used to determine the bromine substructure from data collected above the bromine absorption edge. This method would appear to have general applicability where large substructures need to be determined.

References

[1] Riboldi-Tunnicliffe, A. & Hilgenfeld, R. (1999). J. Appl. Cryst. 32, 1003-1005. [2] Panjikar, S. (2001). Ph.D. Thesis.
[3] Panjikar, S. & Tucker, P. A. (2002). J. Appl. Cryst. 35, 117-119.

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COORDINATE ERROR ESTIMATION OF A SET OF FREE ATOMS

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When a set of free atoms is available and undergoes an interpretation process with the aim of identifying molecular fragments with known stereochemistry, an estimate of the accuracy of the positions of the free atoms is required. This coordinate error estimate is used for the calibration of the likelihood function used in the interpretation, which is analogous to the use of a σA estimate in Maximum likelihood refinement. In order to get a good performance of the pattern recognition algorithm, an accurate estimate of the coordinate error is thus vital. Existing methods to estimate the coordinate error such as the DPI or σA may fail to give correct estimates when the error on the structure is large. We have developed a method that is based on real space properties of the free atoms placed to overcome these problems. A set of distributions has been derived that describes the occurrence of the magnitude of nearest neighbor distances in proteins, given a Gaussian error on the coordinates. When a free atom structure is available, for every atom the distance to its closest neighbor is obtained. This set of interatomic distances and the available distributions are used to calculate likelihoods for a set of proposed variances. The best estimate of the variance of the error model is the variance with the highest likelihood. The error estimation procedure gives fairly accurate estimates over a wide range of applied errors and thus seems suitable for calibration of the likelihood functions used in the interpretation of free atoms.

Keywords: MODEL BUILDING MAP INTERPRETATION MAXIMUM LIKELIHOOD

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SULFUR PHASING AND DIRECT CRYSTALLOGRAPHY

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As a means of improving the throughput and cost-effectiveness of protein crystal structure determination an approach, we term 'Direct Crystallography', is being pursued. Direct Crystallography aims at reducing the number of intermediate steps in going from gene to structure, Its ultimate goal is to determine structures directly from unaltered native crystals using the sulfur and/or metal atoms naturally present in the protein as phasing probes. To achieve this objective, several new and innovative approaches are being pursued. These include: the use of improved X-ray optics and detector technologies, automation of crystal placement/alignment, data collection procedures for collecting highly redundant data in a reasonable time frame, developing new algorithms for detecting, extracting and using weak signals from diffraction data, improving computer software for phasing protein structures, and using single wavelength diffraction data collected at both synchrotron and home sources.

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