MS4-O5 Substructure determination from SAD data using phase retrieval techniques

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The major current bottleneck for macromolecular structure solution from SAD data is determination of positions of the anomalously scattering atoms. The current programs for substructure determination are usually based on the "direct" methods developed for the structure solution of small molecules, obtaining the phase estimates from relations between intensities and phases of the reflections or from the Patterson function.

From a more general point of view, the X-ray crystallography phase problem belongs to the class of non-linear and non-convex inverse problems. Although no general solution is known for this class of inverse problems, they have been studied for decades and efficient phase retrieval algorithms were proposed for the special case of optical phase retrieval. Similarly to most "direct" methods implementations, the phase retrieval techniques perform iterative dual-space recycling. However, unlike the direct methods, the operations performed in either of the spaces alone cannot, even in principle, solve the phase problem.

Dumas&van der Lee (2008) showed that the simplest phase retrieval algorithm - charge flipping, as implemented in Superflip (Palatinus&Chapuis, 2007) - can be used for substructure determination from some SAD datasets. One disadvantage of this simple algorithm may be that it tends to diverge from the solution (Marchesini, 2007) for an inconsistent problem. Clearly, the problem of substructure determination from weak anomalous signals is strongly inconsistent due to the tiny signal-to-noise ratio of the data. The Relaxed Alternating Averaged Reflections (RAAR) algorithm (Luke, 2005) has been designed to elude the divergence while still retaining a high ability to escape local minima.

I have adapted the RAAR algorithm in a new program for Phase Retrieval from Anomalously Scattering Atoms (PRASA). The preliminary results show that the novel approach provides consistently better results than the charge flipping algorithm and is competitive with the state-of-the-art substructure determination programs. In the presentation, the advantages and disadvantages of this new approach will be discussed and its performance will be demonstrated on massive testing results on over 150 real SAD datasets.

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Keywords: anomalous scattering, substructure determination, SAD, phase retrieval, charge flipping, RAAR, phase problem

MS5 Structural information in drug design

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MS5-O1 Structural elucidation of ligand binding sites in family B GPCRs and their application in drug discovery

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G protein-coupled receptors (GPCRs) comprise one of the most important families of drug targets owing to the multitude of roles they fulfil across many different physiological processes. Despite the huge amount of investment in GPCR drug discovery there remains significant opportunities for identification of novel or better drugs. One way to achieve this goal is to utilise structure based drug design (SBDD) strategies. However, GPCRs are generally challenging proteins for crystallisation and structure determinations primarily due to their hydrophobic nature, low expression levels and conformational flexibility. To facilitate application of SBDD approaches to GPCRs Heptares uses its proprietary StaR technology to thermostabilise GPCRs in a single conformational state. The purified StaRs can then be used for crystallisation to yield X-ray structures with multiple ligands as well as used in biophysical screening techniques. Using the StaR approach, we have solved structures of multiple Class B GPCRs in both agonist and antagonist conformations. This has led to the elucidation of the orthosteric and allosteric binding sites. The structural insight into multiple ligand binding sites has significantly increased our ability to design novel drugs to modulate the activities of these receptors.

Keywords: GPCR, structure based drug design, family B receptors, glucagon receptor, GLP-1 receptor