Ultrafast X-ray Diffraction

Applications to Reaction Dynamics and Protein Structure Determination
The x-ray diffraction of protein crystals is the most reliable way to obtain the structure of those systems to which it applies. However there are problems

- It is currently unknown how to crystallize many proteins.
- Molecules in a crystal may change shape in a solvent environment.
- Membrane proteins are especially difficult to characterize for both of the above reasons.
Is it even possible to think about doing single protein molecule x-ray diffraction?

- The signal is measured by photon counting; a high intensity of the x-ray pulse is required for an acceptable signal-to-noise ratio.
- During the x-ray pulse, the protein will Coulomb explode, as some significant fraction of it’s component atoms are ionized. The nuclear motion happens on a longer time scale than probed by the x-ray diffraction.
Electrons are accelerated, and an undulator changes their path. This acceleration of a moving charge generates strongly linearly polarized light with a brilliance proportional to the number of electrons in the beam. Coherent light can be generated by bunching the electrons in the beam. This greatly reduces the intensity of the resulting x-ray pulse.
X-ray Free Electron Lasers

- Spontaneous Amplification of Spontaneous Emission (SASE)
- high brilliance
- coherent light
Fourth Generation X-ray Sources

- These new photon sources are poised to make new kinds of observations with spectroscopy possible.  
  - 80-8000eV (proposed up to 40keV)
  - $10^{12}$ photons/pulse
  - pulse duration < 230fs
  - strongly linearly polarized

- These sources can generate light capable of resolving atomic motions and electronic systems on a previously inaccessible scale.

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The intensity peaks of the x-ray diffraction pattern give directly the Fourier components of the charge density. However, there may be a phase difference between different Fourier components. This is called the “phase problem “ in X-ray crystallography.
Figure 3. X-rays scattered by a crystal form its diffraction pattern.

\( I(k) \propto 1/V_{uc} \int \rho(r)e^{-ik \cdot r} \, dr = F_k \)
Any function periodic in space with a unit cell length of $L$ can be written

\begin{equation}
    f(x) = \sum_{k} \left[ A_k \sin\left(\frac{2\pi k x}{L}\right) + B_k \cos\left(\frac{2\pi k x}{L}\right) \right]
\end{equation}

Note: each Fourier component has two parts, representing different boundary conditions on the unit cell. When the transform is written

\begin{equation}
    f(x) = \sum_{k} F_k e^{i k x}
\end{equation}

$F_k$ is in general complex.
Protein molecules in vacuo are obtained through electrospray injection. These are probed with an x-ray pulse. The proteins leave the source randomly oriented.
The authors propose two possible methods of dealing with the random orientation.

- A fixed-in-space experiment, in which an elliptically polarized nonresonant laser pulse is used to align the molecule.
- A different method of analysis that allows the contributions from different orientations to be filtered out.
In order to get the phase information, the intensity of x-ray diffraction pattern is sampled in a finer grid than the Nyquist frequency of the system (given by the inverse length of a unit cell).

Once the sampling at this frequency is performed, apparently there is enough information to extract the phase information from the modulus of the Fourier component.
Phase Problem

To reconstruct the phase,

- A random phase is chosen, and used to construct a charge density.

- Outside a carefully chosen envelope, the charge density is altered to be closer to zero, and negative parts of the charge density inside the envelope are reduced.

- This new charge density is used to make a new discrete Fourier transform. The phase information of the new charge transform is used with the $|F_k|$ information from the x-ray diffraction measurement.
Simulation

- Assumed a pulse width of 10fs with an intensity of $2 \cdot 10^{12}$ photons per pulse.
- Assumed $10^6$ random orientations of the proteins.
Images
For the future

- “Snapshots” of optically triggered reactions in time.
- Protein electronically excited states could be probed by exciting with an optical pulse before directly measuring the charge density with a following x-ray pulse.
Bibliography

- J. Miao, et. al. PNAS 98, 6641
- E. E. Lattman, PNAS 98, 6535 (2001)