written (in Fortran) to be easily portable to other computer platforms. This program has begun to play a major role in our studies by providing a tool with which measured diffraction data can be compared to 3D computer simulations.

The wide applicability of this program will be demonstrated by showing example diffraction patterns from simulated size-effect displacements in metallic alloys, mineral systems exhibiting oxygen vacancy order and cation relaxations, and conformational disorder in organic crystals. The program and a manual describing its use will be made freely available.

two components separating short range and long range coupling. The results show that the coupling of atomic movements in these highly ordered protein crystals is mostly short−ranged, similar to that in liquid rather than that in elastic solids. The total mean square atomic displacements for tetragonal lysozyme, triclinic lysozyme and 2 Zn rombohedral insulin are 0.25 $Å^2$, 0.13 $Å^2$ and 0.2 $Å^2$, respectively. About 90% of the total mean square displacements in these crystals are correlated over distances the size of one amino acid residue (~ 6 Å). Movements that are correlated over the distance the order of size of the protein molecule (~ 50 Å) account for about 10 % of the total displacements. Experiments are under way to measure diffuse scattering data from cubic insulin crystals at different pH and ionic strength, and to analyze these data in terms of the analytical model described above and empirical models about switching between conformational substates that are evident in high resolution structures.

MS−02.05.03   DIFFUSE SCATTERING AND INTRA− MOLECULAR FLEXIBILITY IN PROTEINS by J−P. Benoit, P.Faure and J.Doucet*, LURE and Laboratoire de Physique des Solides, Universite Paris−Sud, F91405 Orsay, France

The growing interest in the dynamical behaviour of proteins is motivated by the fact that in many cases the functional roles of these biological molecules do not only depend on their rigid three−dimensional structure but also on their deformability or flexibility. The atomic motions implicated in the macromolecular deformations cover indeed an extremely large range in amplitudes (sub−Angstroems to a few tenths of angstroems) and in duration from femto−s when only a few atoms are concerned up to seconds when thousands atoms move in a concerted (or correlated) way. Due to this enormous variety of motions, the experimental approach of the large panoply of techniques used. In order to overcome this difficulty calculation and simulation approaches have been developed. Nevertheless, up to now the likelihood and the effeciency of these theoritical approaches had only been undirectly tested through their ability to facilate protein structure determination and refinement by NMR or X−ray diffraction methods, or by comparisons of crystallographic B−factors with the mean square fluctuations of atoms.

We evidence here for the first time, direct correspondance between the intramolecular motions predicted by numerical simulations and X−ray diffuse scattering experimental observations.

MS−02.05.04 STUDIES OF DIFFUSE SCATTERING REVEAL LIQUID−LIKE DISORDER IN PROTEIN CRYSTALS, by Y. Li*, D.L.D. Caspar, B. Yu, Rosenstiel Basic Medical Sciences Research Center, Brandeis University, Waltham, MA 02254−9110, U.S.A., and J. B. Clarage, Department of Biochemistry and Cell Biology, Rice University, Houston, TX 77251,

Diffuse scattering from protein crsytals, notably haloes surrounding Bragg reflections, contain information about both the amplitude and correlation of atomic movements in the protein molecules. We have developed an analytical model for simulating diffuse scattering from protein crystals by representing the averaged Patterson function as the convolution of the peaks in the ideally ordered Patterson with a Gaussian whose variance is a function of the mean square atomic displacement and a correlation funciton which describes coupling between the movements. Based on this model we have simulated diffuse scattering data collected from lysozyme and insulin crystals in terms of an exponentially decaying correlation function which has

MS−02.05.05   MODELS FOR DIFFUSE SCATTERING FROM PROTEIN CRYSTALS   by Nobuhiro Go*[1], Kenji Mizuguchi[1] and Akinori Kidera[2], [1]Department of Chemistry, Faculty of Science, Kyoto University, Sakyo−ku, Kyoto 606, Japan and [2]Protein Engineering Research Institute, 6−2−3 Furuedai, Suita, Osaka 565, Japan

We have developed a new theoretical framework for the study of X−ray diffuse scattering from protein crystals, in which we start from a general equation and introduce a series of approximate models, appropriate for analyses of experimental data obtainable with different degrees of precision. When a high precision data are available, we can employ essentially the same model used for the recently developed method, NM−REF (Kidera, A.&Go, N.(1992) *J. Mol. Biol.* 225, 457−475), of refinement of protein structure from the usual Bragg diffraction. Further approximation is based on the assumption that the covariance matrix of atomic displacements can be expressed by using a relatively simple empirical correlation function. The formalism using the correlation function retains information about atomic details and allows us to introduce a variety of models, in which (1) the effect of higher order scattering is included, (2) intra−and intermolecular correlations can be distinguished, and (3) amplitudes of fluctuations can be atom−dependent. By calculating diffuse−scattering patterns from a human lysozyme crystal, we have examined the assumptions used in these models and discussed important factors that determine the general feature of the scattering patterns. The higher order scattering is shown to make a signficant contribution at high resolutions. It is also shown that the resulting patterns are sensitive to changes in correlation lengths of about 1A, but they are also affected sensitively by the form of the correlation function. Only the 'average' value of the intra− and intermolecular correlation lengths seems to determine the gross feature of the pattern.

MS−02.05.06 DIFFUSE SCATTERING IN ELECTRON DIFFRACTION FROM MOLECULAR ORGANIC- AND PROTEIN-CRYSTALS; ANALYSIS OF CRYSTAL-CRYSTAL PHASE TRANSITIONS. By Douglas L. Dorset*, Electron Diffraction Dept., Medical Fndn. of Buffalo, Inc., 73 High St., Buffalo, NY 14203 USA

Electron diffraction patterns from molecular organic crystals often contain a pronounced directional diffuse component

which is related to the continuous transform of the component molecules in the crystal unit cell. Such details in e.d. patterns from anthracene taken by Charlesby, Finch and Wilman in 1939, in fact, led to the use of such information by some crystallographers for structure analysis. Generally there are two origins to this diffuse signal: thermal motion and static displacement. In polymethylene chain compounds such as the n-paraffins and polyethylene, for example, both components can be found in respective regions of the reciprocal lattice. The thermal signal is observed in the projection down the chain axes and is found to be extinguished when the samples are cooled to 40K. A much stronger diffuse pattern in the projection onto the molecular axes (epitaxially crystallized samples), however, is not diminished even when the specimen is cooled to 4 K. Models based on disordered molecular packings indicate that its origin is due to small, residual longitudinal chain translations which persist in the lamellar layers when the material is crystallized through the conformationally-disordered structure ('rotator' phase for certain chain lengths) to the lowest energy methylene subcell structure. This type of disorder fits well with the larger longitudinal chain translations observed in the reverse process, when paraffin crystals are heated toward the melting point. Diffuse scattering can also be useful for following crystal-crystal transitions in two-dimensional protein crystals. For example, the onset of a protein trimer recrystallization in hexagonal layers of the Omp F porin from *E. coli* reconstituted in DMPC was first identified by the diffuse scattering signal found in the computed Fourier transform of an electron microscope image (or seen in electron diffraction patterns). Subsequent changes in the diffuse signal, and finally its sharpening into directional streaks which then, in time, split into discrete spots, correspond to the crystallization of a polar orthorhombic cell in the less anisotropic hexagonal layer. Cross-correlation analysis on images also indicates that the total sampled area can contain recrystallized orthorhombic subareas oriented in any of three possible directions. This is the first direct observation of cooperative rotational diffusion of a membrane protein in a lipid bilayer.

PS–02.05.07 THE DISORDERED STRUCTURE OF MULLITE USING DIFFUSE X-RAY SCATTERING. By B. D. Butler*, T. R. Welberry, and R. L. Withers, Research School of Chemistry, Australian National University, Canberra A.C.T. 0200, Australia

A description of the three dimensional disordered arrangement of oxygen vacancies in a mullite of composition $Al_2(Al_{2+2x}Si_{2-2x})O_{10-x}$ where x=0.4 has been developed which is consistent with the measured diffraction data. The structure of this material was modelled using Monte Carlo techniques where the oxygen vacancies were allowed to interact via a set of pair energies. Cations in adjacent tetrahedrally coordinated sites were given displacements that depended only on the local arrangement of these oxygen vacancies. A calculation of the diffraction pattern from this model crystal compared favourably with measured diffraction data. Not only can this model describe the origin of the observed incommensurate diffraction maxima in the $2c^*$ reciprocal section but it is also consistent with many broader diffuse diffraction features that have been observed in other reciprocal sections. In addition to large repulsive $2 \cdot 110\bar{0}$ and [110] interactions that are required to

satisfy certain cation bonding requirements, it was found that unequal vacancy repulsive interactions were required along $\cdot 100\bar{0}$ and $\cdot 010\bar{0}$. Attractive vacancy–vacancy interactions along $2 \cdot 112\bar{0}$ and $\cdot 001\bar{0}$ were also necessary but in the latter case the magnitude of the interaction is such that the probability of having an $\cdot 001\bar{0}$ vacancy pair was near that of a random vacancy distribution.

PS–02.05.08 ATOMIC FORCE MICROSCOPY HELPS DOMAIN STRUCTURE DETERMINATIONS BY X-RAY AND THERMAL ANALYSES. By N. Masaki*, Fac. of Pharm. Sci., Kyoto Univ., Kyoto, Japan, and Y. Yoshimura, Fac. of Sci. &. Eng., Ritsumeikan Univ., Kyoto, Japan, and H. Kado, K. Yokoyama and T. Tohda, Central Res. Labs., Matsushita Elec. &. Ind. Co. Ltd., Moriguchi, Japan, and N. Man and M. Sakiyama, Fac. Sci., Osaka Univ., Toyonaka, Japan.

Crystal structure of phenytoin (diphenyl hydantoin $C_3N_2H_2O_2 \cdot 2(C_6H_5)$, a = 6.230, b = 13.581, c = 15.532 Å, $Pn2_1a$, Z = 4) of famous anticonvulsant drug for epilepsie had been determined at room temperature by Camerman &. Camerman[1]. We found a reversible phase transition of the crystal of phenytoin at 183.5°K by X-ray diffraction photographs and confirmed by thermal analysis. Satellite reflections in diffraction pattern and λ-shaped curve of thermal analysis convinces of that it is a typical order-disorder phase transition caused by domain structure formation in lower temperature phase. Atomic force microscopy (AFM) revealed the domain structure even at room temperature. Therefore domain structure is inherent in phenytoin crystals through across the phase transition temperauture. X-ray pattern in high temperature phase can not detect the phase boundary. This is the third application of AFM for X-ray analysis followed structure determination[2] and space group determination[3]. New technique of domain structure analysis by X-ray diffraction and AFM will be discussed.

① Camerman, A. & Camerman, N. (1971). Acta Cryst. B27, 2205.
② Masaki, N. et al. (1992) Ultramicroscopy, 42-44, 1148. Chem. Pharm. Bull, 39, 1899.
③ Masaki, N. et al. (1992) Asian Crystallographic Association Conference Abstract. 14s31 (Singapore, Nov.)

PS–02.05.09 MEASUREMENT AND ANALYSIS OF X-RAY DIFFUSE SCATTERING FROM PROTEIN CRYSTALS, By Bin Yu, Donald L. D. Caspar, Youli Li, Rosenstiel Basic Medical Research Center, Brandeis University, Waltham, MA 02254-9110, U.S.A.

Analysis of diffuse X-ray scattering provides important information about correlations of atomic movements in protein crystals. Computer modeling of such correlations, either based on analytical considerations or empirical observation, can be tested by comparison with 3-D diffuse scattering pattern. Data taken from tetrahedral lysozyme crystals on synchrotron at Brookhaven National Laboratory show strong modulations in its diffuse scattering patterns. We are conducting experiments to find differences in diffuse scattering resulting