method was applied to process X-ray diffraction patterns from well-oriented sols, which allowed us to extract layer-line amplitude distribution reliably. The phase data from the EM analyses were combined with X-ray amplitudes from the native and heavy atom derivatives of the filament to locate heavy atom binding positions in difference Fourier maps. The multiple isomorphous replacement phasing procedure is being applied to deduce electron density maps at higher resolution available in the X-ray data.

MS13.03.02 REFINEMENT OF THE F-ACTIN STRUCTURE AGAINST FIBER DIFFRACTION DATA. M. M. Tirion, K.C. Holmes, M. Lorenz. Department of Membrane Research and Biophysics, weizmann Institute of Science, Rehovot, Israel.

The crystal structure of G-actin permits development of atomic models of F-actin based on X-ray fiber diffraction images from aligned filaments. The initial atomic model of F-actin, obtained using only 4 structural parameters, reproduced most features of the X-ray and electron-microscopic data. However, the 7Å resolution X-ray fiber diffraction images contain sufficient data to refine more than 4 structural parameters. Refinement at atomic level is too fine-grained and would overfit the diffraction data. The challenge therefore, is to identify appropriate structural parameters which reduce the fiber diffraction residual while preserving the stereochemistry of actin.

We present two different methods: use of slow, normal modes of G-actin as structural refinement parameters, and a Monte-Carlo method to select appropriate subdomains in G-actin, for the refinement of F-actin. Both methods resulted in a significant reduction of the residual and showed similar trends in the structural changes from G- to F-actin, including the closure of the nucleotide binding pocket between the large and small domains. This finds strong support from biochemical experiments which show that polymerization of actin inhibits the nucleotide exchange rate.

MS13.03.03 STRUCTURE OF MUSCLE IN THE RELAXED STATE AND RIGOR LABELLED WITH MYOSIN S-1: IMPLICATIONS ABOUT FORCE PRODUCTION. Harford, J. J., Hudson, L., Denny, R., Squire, I. M., Biophysics Section, Imperial College, Prince Consort Rd, London, UK

The purpose of this project is to solve the structure of the A-band unit cell in relaxed fish muscle and to use this to follow the molecular movements in active muscle regulation and force generation. The first frame, relaxed muscle, has now been solved. Time-resolved X-ray diffraction patterns from contracting plaice fin muscle have been recorded at 1ms or 5ms time-intervals (depending on the region being studied) and these are then processed using CCP13 software and modelled using the known myosin head shape and actin filament structure. The solution for the first frame has revealed the organisation of myosin heads around the actin filaments in the hexagonal A-band unit cell. This frame is now being used as a starting structure to model successive frames through the time-series, thus producing a 'Movie' of the molecular processes involved in contraction. Already, from analysis of the resting and rigor muscle structures, it has been shown that myosin heads need to swing axially on actin by about 5 to 15nm in order to proceed from the resting to rigor states. It is presumed that the heads need to do this in active force generation as well. More recently we have recorded X-ray diffraction patterns to a resolution of about 1nm from chemically skinned plaice muscle in the rigor state labelled with exogenous myosin S-1 heads. Modelling of these patterns, now in progress, is revealing in much greater detail the changes in conformation of the myosin heads that accompany this transition from the relaxed to the rigor state.

MS13.03.04 STRUCTURAL DIVERSITY IN FILAMENTOUS BACTERIOPHAGES. Makowski, L., Institute of Molecular Biophysics, Florida State University, Tallahassee, FL 32306-3015, USA

X-ray and neutron diffraction data have been used to study the structure of filamentous bacteriophage M13 and Pf1, and chemically and genetically constructed variants of these particles. Ambiguity is intrinsic to the analysis of the structure of macromolecular assemblies using fiber diffraction. The ratio of data-to-model parameters for fiber diffraction is substantially lower than for x-ray crystallography. Even when the number of isomorphous derivatives are available, it may be not possible to obtain a unique structural solution from fiber diffraction data. Consequently, after a number of years, there continues to be substantial controversy in the literature about the filamentous bacteriophages M13 and Pf1. Differences among the molecular models for the viruses have substantial implications for our understanding of their membrane-mediated assembly, and for their use in phage display technology. It is now possible to obtain fiber diffraction data from a wide range of structural variants of filamentous bacteriophages that can provide rigorous tests of competing structural models. The relative positions and fidelity with which known structural features of the variants are reproduced in difference maps calculated on the basis of competing structural models provides a significant test of the competing models. The use of this strategy for exploring competing models of the coat proteins of filamentous bacteriophages Pf1 and M13 is demonstrated.

MS13.03.05 MOLECULAR DYNAMICS REFINEMENT WITHOUT DECONVOLUTION IN FIBER DIFFRACTION OF BIOLOGICAL POLYMERS. Gerald Stubbs and Hong Wang, Department of Molecular Biology, Vanderbilt University, Nashville, Tennessee 37235

There is useful information in a fiber diffraction pattern even beyond the resolution at which layer lines overlap. Most fiber diffraction studies involving large monomers have been limited to the non-overlapping region of the pattern, since objective determination of a molecular structure depends on complete phasing of separated diffraction intensities. Once a model has been constructed, however, there is no reason to discard higher resolution data. Makowski has shown that beyond the point where layer lines can be deconvoluted, the information content of a diffraction pattern continues to increase with resolution, although only linearly. The most powerful method of refinement of macromolecular structures currently available is molecular dynamics refinement, for example as used in our adaptation of the program X-PLOR to use fiber diffraction data. We have further modified X-PLOR to handle undeconvoluted data. In addition to atomic coordinates, global parameters such as specimen disorientation are refined. Testing so far has been against simulated and real helical virus data. Supported by NSF grant MCB 9506204.