PS04.09.21 CRYSTALLOGRAPHIC STUDY OF A FELINE CALICIVIRUS. Lan Zhou, Ming Luo, Department of Microbiology, Center for Macromolecular Crystallography, University of Alabama at Birmingham, Birmingham, AL USA

Feline calicivirus (FCV) is a nonenveloped, spherical animal virus of the Calicivirus family. It contains a positive sense, single-stranded RNA genome and one major capsid protein. The virion has a diameter of 405 Å and exhibits T=3 icosahedral symmetry. The FCV crystals belong to an orthorhombic crystal system with unit-cell dimensions a=889.0 Å, b=998.3 Å, c=437.6 Å. Based on the Vm value (3.4 $Å^3$/Da), it was estimated that there is one FCV particle in one crystallographic asymmetric unit, which implies the presence of 60-fold noncrystallographic redundancy.

The orientations of the FCV particles in the unit cell were determined with a self-rotation as well as the locked rotation function using the GLRF program. Data between 15-7 Å and 7-4 Å, respectively, were used to search for 5-, 3- and 2-fold axes. Two sets of 5-, 3-, and 2-fold noncrystallographic symmetry axes were obtained. The orientations of the 5-, 3-, and 2-fold noncrystallographic symmetry axes of each set were those expected for an icosahedron. This indicated that two different particle orientations exist in the crystallographic asymmetric unit. Combining this with the packing consideration and the Vm value, it was concluded that there is one unique particle content from two differently oriented FCV particles in one crystallographic asymmetric unit. The precise particle orientations were refined by optimizing the fit between the directions of the observed self-rotation function peaks from one of the virus particle and the corresponding axes of a standard icosahedron.

In order to precisely define the particle positions in the unit cell, heavy atom derivatives of FCV crystals are in pursuit.

Receptors/Signal Transduction

MS04.10.01 CRYSTAL STRUCTURE OF A COMPLEX BETWEEN THE EXTRACELLULAR DOMAIN OF THE HUMAN ERYTHROPOIETIN RECEPTOR AND AN AGONIST PEPTIDE. Oded Livnah†, Enrico A. Stura†, Dana L. Johnson§, Linda K. Jolliffe§, Ian A. Wilson†, †The Scripps Research Institute, Dept. of Molecular Biology, 10666 North Torrey Pines Rd., La Jolla, CA 92037, §The R. W. Johnson Pharmaceutical Research Institute, Drug Discovery Research, 1000 Route 202, Box 300, Raritan, NJ 08869

A 20 residue cyclic peptide has been shown to be capable of effecting dimerization of a class 1 cytokine receptor and is able to mimic the biological activity of the natural hormone ligand erythropoietin (EPO). The crystal structure of the complex between the extracellular domain of the erythropoietin receptor (EPOR) and this peptide, unrelated in sequence to erythropoietin was determined at 2.8 Å resolution in an orthorhombic crystal form having the space group $P2_12_12_1$ with cell dimensions a=59.2Å, b=75.5Å, c=132.2Å with two receptor molecules in the assymetric unit. Initial MIRAS phases were calculated to a resolution of 3.1 Å using the program package PHASES and had a mean figure of merit of 0.64 (25.0-3.1 Å). Phases were refined in PHASES using the solvent flattening protocol. The structure was refined using the program package X-PLOR to an R-value of 0.21 without any solvent molecules added.

Each receptor molecule consists of two domains with fibronectin type-III folds having a 90° angle between them. The structure of the complex consists of an almost perfect two-fold 2 receptor:2 peptide assembly. The dimerization assembly differs markedly from the human growth hormone receptor complex, another class 1 cytokine receptor of known structure, and suggests that more than one type of dimerization may be sufficient to induce signal transduction and cell proliferation. The functional mimic of a biological protein ligand by an unrelated molecule has previously appeared an unreachable goal. This EPOR complex suggests future progress towards the design of a non-peptidic small molecule drugs may be possible.

MS04.10.02 THREE-DIMENSIONAL STRUCTURE OF HUMAN INTERFERON-γ COMPLEXED TO ITS CELL RECEPTOR. Daniel Thiel[1], Marie le Du[1], Richard Walter[1], Allan D'Arcy[2], Christiane Chene[2], Michael Fontoulakis[2], Gianni Garotta[2], Fritz Winkler[2] and Steve Ealick[1]. Section of Biochemistry, Molecular and Cell Biology, Cornell University, Ithaca, NY 14853 and Pharma Research, F. Hoffman-La Roche Ltd, 4002 Basel, Switzerland.

Interferon-γ (IFN-γ) was originally identified because of its ability to "interfere" with viral replication after infection. Since then IFN-γ has been shown to be responsible for many types of immune responses. IFN-γ is secreted by activated T-cells and natural killer cells and functions by binding to a specific high affinity cell receptor (Rα) and a second low affinity receptor (Rβ). We have used X-ray crystallography to determine the structure of IFN-γ complexed to its high affinity receptor (Rα) at 2.7 Å resolution. The complex crystallizes in space group C2 with a = 202 Å, b = 114 Å, c = 74 Å and β = 117° and with one complex per asymmetric unit. The structure was determined by MAD phasing in which the eight methionines of IFN-γ were substituted by selenomethionine. Data were measured at four different wavelengths using frozen crystals on CHESS beamline F-2. The complex consists of one IFN-γ dimer, two ordered receptors and one disordered receptor; the IFN-γ and ordered receptors show approximate two-fold symmetry. The IFN-γ dimer is made up of identical subunits with six α helices each. The subunits are interdigitated such that four helices from one subunit form a cleft that cradles the C-terminal helix from the other subunit. Each receptor molecule contains two 100 amino acid domains joined by a hinge. Each of these domains contains seven β strands with a fribronectin type III fold. In the crystal structure, the receptor molecules are truncated at the point where they normally attach to the cell membrane. Most of the interactions between IFN-γ and the two ordered Rα's occur between an N-terminal helix, C-terminal helix and the loop connecting helices A and B of IFN-γ and the N-terminal domain of the receptor. There is no interaction between these two Rα chains within one complex. A third partially disordered Rα chain was observed in the electron density maps. The C-terminal domain of the disordered receptor packs against the C-terminal domain of an ordered receptor chain. The N-terminal domain of the third receptor is almost completely disordered and is located near the IFN-γ molecule. Several pieces of evidence suggest that the disordered Rα chain may be mimicking an Rβ chain. This observation has lead to a hypothetical model of a ternary IFN-γ/Rα/Rβ complex.

MS04.10.03 REFINED STRUCTURES OF HETEROTRIMERIC G PROTEIN COMPLEXES AT HIGH RESOLUTION. John Sondek*, Andrew Bohm*, David G. Lambright#, Nikolai P. Skiba‡, Joseph P. Noel¶, Heidi E. Hamm‡, Paul B. Sigler*, *Dept. of Molecular Biophysics & Biochem. and Howard Hughes Medical Institute, Yale University, New Haven, CT 06510, ‡Dept. of Physiology & Biophysics, U. of Illinois, Chicago IL 60612 #Prog. in Mol. Med., U. of Massachusetts Med. Center, Worchester MA ¶Structural Bio. Lab., The Salk Institute, La Jolla, CA 92037

Many signaling cascades use heterotrimeric G proteins coupled to seven-helical transmembrane receptors to convert extra-