

## CURRICULUM VITAE

First Name: Takemasa

Last Name: Kawashima

Citizenship: Japanese

Immigration Status Permanent resident of the USA.

Research Interests Structure-function studies of macromolecules in biological systems, in particular signal transduction involving G-proteins.

Experience and Skills:

Laboratory and Research

- Maintenance of research laboratory operation, including troubleshooting and performing minor repairs of laboratory equipment, computer hardware and software, and preparing samples for biochemical, biophysical and chemical analyses.
- Practice of basic molecular biology protocols, including nucleic acid extraction and cell transformation/transfection, preparation of screening media, and induction of gene construct expression.
- Production of eukaryotic proteins using baculovirus and insect cells.
- Isolation of proteins from biological tissues and cell cultures, using affinity, ion-exchange and size-exclusion chromatography techniques, analysis of purification steps by gel electrophoresis, western-blot, UV-VIS spectroscopy, and multi-angle scattering, as well as filter-binding assays involving radio-labeled chemicals.
- Screening of crystallization conditions for macromolecules, manually or semi-automatically in multi-well plate format, configuration and operation of X-ray optics for optimum beam delivery onto sample, collection and processing of X-ray diffraction data, determination of crystal structure using various phasing methods (e.g. multiple isomorphous replacement, anomalous diffraction, and molecular replacement).
- High manual dexterity allowing for, e.g., soldering of modern surface-mounted electronic components, recovery of small organs from laboratory animals, to cleavage of sub-milimeter size crystals under microscope.

Computer and Information Technology

- Using and troubleshooting common computer operating systems (Windows, MacOS, Unix variants), software and hardware.
- Programming using compiled languages and scripts (e.g. C/C++, JavaScript, Python, Node.js); quickly adaptable to new languages and development

## CV (continued)

environments.

- Performing basic administration of networked resources in multi-user environments, including networked database servers and web interfaces.
- Extracting data from binary files manually.

### Languages

- Fluent English, French and Japanese; basic German.
- Translation of patents, clinical trials reports, scientific articles, technical and non-technical documents, as well as web site, software and video game resource files.

### Education:

#### **1981-1984 Secondary Education: Lycée Louis-Le-Grand, Paris, France**

July 1984    Baccalauréat série C (high school diploma with emphasis on maths and physics)

#### **1984-1986 Medical School: Université René Descartes, Paris, France**

Oct 1984-Jul 1986    First year of medical studies covering:

- Histology and embryology
- Thermodynamics, chemistry, biochemistry and enzymology
- Fluid dynamics, classic and quantum mechanics
- Statistics

#### **1986-1995 Undergraduate and Graduate Studies: Université Pierre et Marie Curie, Paris, France**

Oct 1986-Jul 1987    Diplôme d'études universitaires générales, science et nature de la vie  
(second year of university diploma for general studies in life sciences, equivalent to bachelor's degree; transferred from first year of medical studies)

- Option: biochemistry-chemistry

Oct 1987-Jul 1988    Licence de biochimie (equivalent to first year of master's degree in biochemistry)

- Certificate: Biochemistry I

- Modules: Bioorganic Chemistry, Fundamentals of Genetics

Oct 1988-Sep 1990 Maitrise de biochimie (equivalent to master's degree in biochemistry)

- Certificate: Biochemistry II
- Modules: Molecular Biophysics, Membrane Biophysics, Biomathematics

Jan 1991-Sep 1991 Diplôme d'études approfondies de biophysique moléculaire

(advanced degree in molecular biophysics, pre-requisite for doctorate program)

- Theory: UV-VIS, IR and Raman spectroscopy, 1D, 2D and solid-state Nuclear Magnetic Resonance, Electron Paramagnetic Resonance, X-ray crystallography, cellular electrophysiology and ion channels, biophysics of nucleic acids and proteins
- Practical work performed at the European Molecular Biology Laboratory, Grenoble Outstation, France, titled:  
"Crystallization of the EF-Tu·EF-Ts complex, polypeptide elongation factors from *Escherichia coli*"

Oct 1991-Dec 1995 Doctorat d'université, spécialité biophysique moléculaire

(doctorate degree in molecular biophysics)

- From the research work at the European Molecular Biology Laboratory, Grenoble Outstation, France, titled:  
"Three-dimensional structure determination of the EF-Tu·EF-Ts complex, polypeptide elongation factors from *Escherichia coli*, by X-ray diffraction"

## Extracurricular Training

26 Jun-6 Jul 1990 Erasmus Intensive Programme Plant Biology in Space, Department of Botany, School of Biological Sciences, University of Nottingham, UK.

- Theory and practical work on studies of plant systems in orbiting vehicles.

## Career:

### 1991-1997 European Molecular Biology Laboratory, Grenoble Outstation, France

Sep 1991-Oct 1995 EMBL Pre-doctoral Fellow

- Performed overexpression, isolation, crystallization and X-ray structure determination of the *E. coli* EF-Tu·EF-Ts complex.
- Used in-house X-ray sources and synchrotron beam-lines at LURE and ESRF

## CV (continued)

in France, and DESY in Germany.

- Helped establish the routine use of secure network-mounted mass storages and remote procedure call-based crystallographic data processing practices in a mixed computing environment running Apple OS, Microsoft Windows, VAX/VMS and UNIX.
- Contributed in the porting of crystallographic data processing suite (CCP4) from VAX/VMS FORTRAN to UNIX SYSTEM V FORTRAN.

Jan 1996-Jan 1997 EMBL Post-doctoral Fellow

- Oversaw follow-up studies of EF-Tu·EF-Ts quaternary structure by scanning electron microscopy.
- Taught practical aspects of cryocrystallography in local workshops for students

### **1997-2022 Department of Molecular Biophysics and Biochemistry, Yale University, USA**

Mar 1997-Oct 1999 Human Frontier Science Program Post-doctoral Fellow, laboratory of Prof. Paul B. Sigler

- In a collaboration with the laboratories of Drs. Robert J. Lefkowitz and Marc G. Caron of Duke University, received training to purify heterologously expressed human beta-adrenergic receptor.
- With the assistance of the technical staff, oversaw the establishment of recombinant expression systems for all three components of the heterotrimeric G-protein Gs, and developed protocols for the reconstitution and isolation of Gs.
- Assisted members of the laboratory collect data at the National Synchrotron Light Source and at the Cornell High Electron Synchrotron Source.

Nov 1999-Aug 2000 Howard Hughes Medical Institute Post-doctoral Associate, laboratory of Prof. Paul B. Sigler

- Continued the project of determining the X-ray structure of the beta-adrenergic receptor alone and in complex with arrestin 2 or the heterotrimeric G-protein Gs.
- With the assistance of the technical staff, started parallel work on the heterologous expression, purification and crystallization attempts of bovine rhodopsin, its variants, and complexes with transducin and arrestin.

Sep 2000-Aug 2002 Howard Hughes Medical Institute Post-doctoral Associate, laboratory of Prof. Thomas A. Steitz

- Continued the work on rhodopsin, by switching to isolation from natural

source, and forming the complex with arrestin.

- Managed the shared tissue culture facility (biosafety level 2) in the Bass Center; duties included user support, enforcing biosafety, handling of emergency calls as well as basic repairs and maintenance of equipment.

Sep 2002-Dec 2004    Research Assistant, laboratory of Prof. Thomas A. Steitz

- Continued the rhodopsin complex structural analysis project.
- Provided technical and scientific assistance to members of the laboratory and other researchers.
- Managed the shared tissue culture facility (biosafety level 2) in the Bass Center.

June 2005-Present    Research Assistant, laboratory of Prof. Donald M. Engelman  
(part-time since November 2012)

- Continued the rhodopsin complex structural analysis project.
- Reprogrammed and adapted liquid handling robots for cold-room temperature crystallization use.
- Participated in scientific discussions with members of the laboratory.
- Provided assistance with preparation of samples for biochemical, biophysical and chemical analyses.
- In a collaboration with the laboratory of Professor Peter M. Glazer, attempted crystallization of Gamma Protein-Nucleic Acid triplexes.
- Performed troubleshooting and minor repairs of scientific equipment, computer hardware and software.
- Helped with reorganizing the laboratory to accommodate changes in the floor plan.

#### Awards/Mentions:

1990-1995    European Molecular Biology Laboratory Pre-doctoral Fellowship

1996    European Molecular Biology Laboratory Post-doctoral Fellowship

1996    Featured prominently in the main article about the Grenoble Outstation, and in a special inset, in the EMBL Annual Report 1995-96, pp 43-46.

1997-1999    Human Frontier Science Program Long-term Fellowship

#### Bibliography:

1) Kawashima, T., Berthet-Colominas, C., Wulff, M., Cusack, S., Leberman, R. The

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structure of *Escherichia coli* EF-Tu·EF-Ts complex at 2.5Å resolution. *Nature* **379**, 511-518 (1996) [Erratum in *Nature* **381**, 172 (1996)].

- 2) Kawashima, T., Berthet-Colominas, C., Cusack, S., Leberman, R. Interconversion of crystals of the *Escherichia coli* EF-Tu·EF-Ts complex between high and low diffraction forms. *Acta Crystallographica* **D52**, 799-805 (1996).