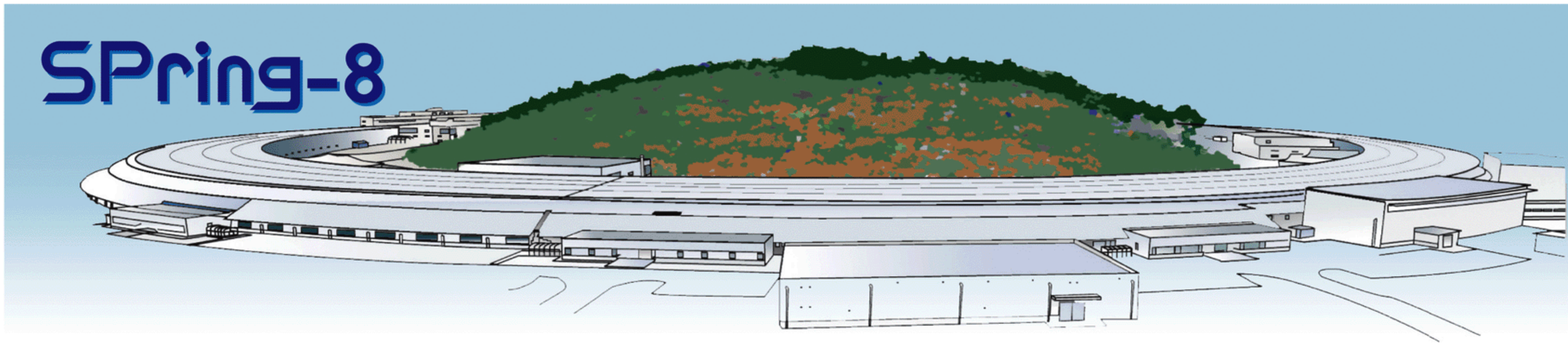

RIKEN Structural Biology II Beamline, BL44B2

This beamline is dedicated to macromolecular crystallography in monochromatic X-ray mode; however, white X-rays could be used until two years ago. White X-rays from the standard "B2"-type bending magnet are monochromatized using a fixed exit double-crystal monochromator (DCM) then focused using a 1-m-long platinum-coated bent-cylinder mirror. The glancing angle (2 - 5 mrad), the radius of curvature (3000 - 7000 m), and the vertical position (±15 mm) of the mirror are adjusted for optimum focusing at the sample position. The photon flux at the sample position is estimated to be 1.1×10^{11} photons/sec at 12.4 keV for a beam size of 0.22×0.20 mm^2 (FWHM).

Dynamical Observation of Single Molecular KcsA Potassium Channel by Diffracted X-ray Tracking (DXT)

Dr. Y. C. Sasaki (JASRI/SPring-8), Professor S. Oiki (University of Fukui), and their research group succeeded in monitoring the dynamical motions of individual single KcsA potassium channels upon gating [1]. In this single molecular detection system using X-rays, which they named Diffracted X-ray Tracking (DXT), they observed the rotating motion of an individual nanocrystal linked to a specific site in individual protein molecules as shown in Fig. 1. In this experiment, a gold nanocrystal was attached to the end of the cytoplasmic domain of individual KcsA molecules. They observed the motions of both the full-length KcsA molecule and its pore portion. For DXT, white X-rays at BL44B2 of SPring-8 were used to record Laue diffraction spots from labeling gold nanocrystals. As a result, at an acidic pH where the channels underwent gating, the individual channels exhibited vigorous twisting motion as shown in Fig. 2. The range of this twisting motion was 10 - 50 degrees, while the bending motion was within 5 degrees. These motions were also observed from the channel fixed in two different orientations: an erect orientation and a sideways-laid orientation. DXT is expected to be used for detecting the single molecular motions of many ion channels and functional proteins with super high accuracy.

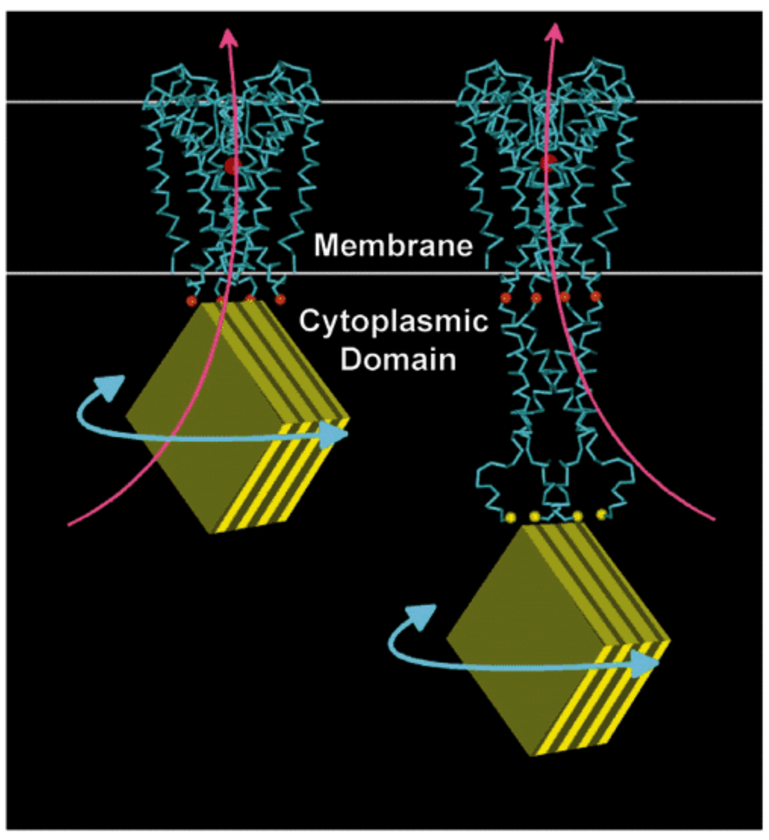

Fig. 1
The arrangement of KcsA molecule and labeled nanocrystal.
The red direction shows the flow of potassium ions.

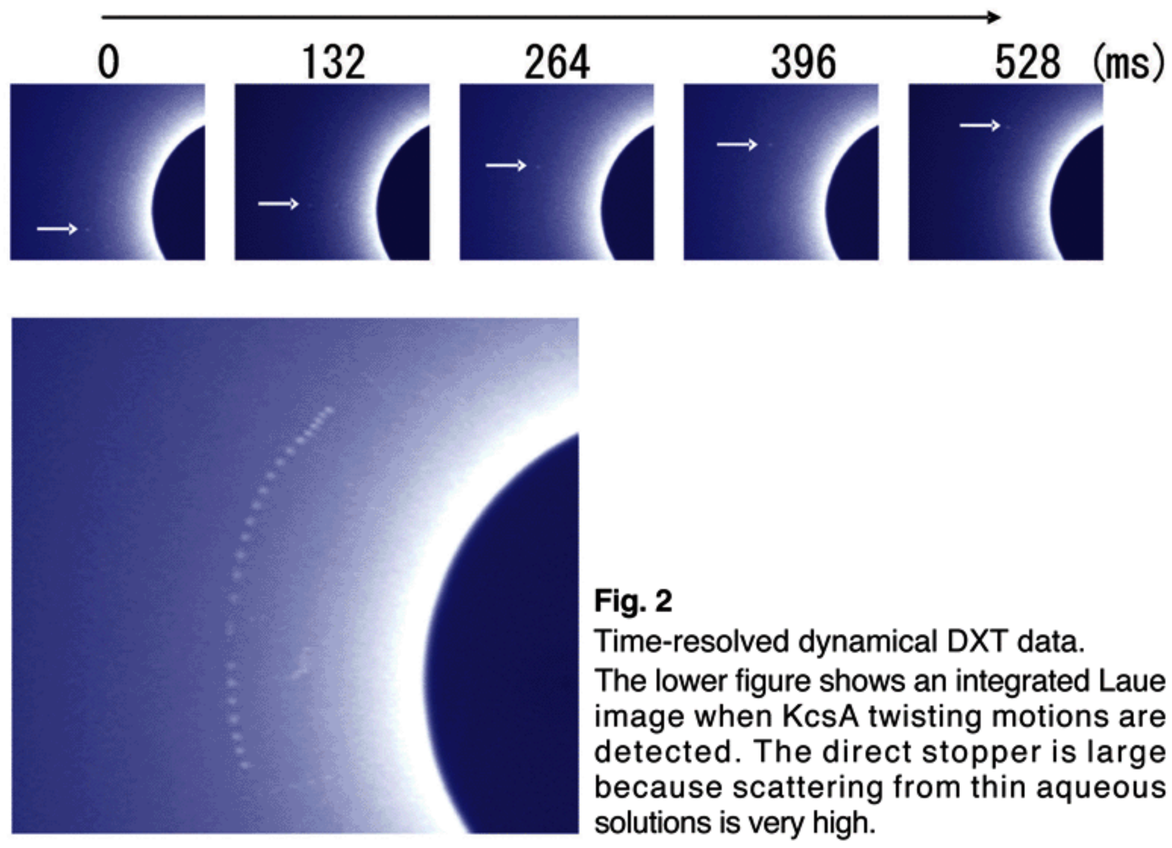

Fig. 2
Time-resolved dynamical DXT data.
The lower figure shows an integrated Laue image when KcsA twisting motions are detected. The direct stopper is large because scattering from thin aqueous solutions is very high.

[1] H. Shimizu, M. Iwamoto, F. Inoue, T. Konno, Y. C. Sasaki, S. Oiki: Global Twisting Motion of Single Molecular KcsA Channel upon Gating, Cell 132, 67-78 (2008)

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