

Synchrotron Radiation-based CD and OCD at ANKA: UV-CD12 beam line, Experimental Station and Applications

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In life sciences synchrotrons are generally thought of in conjunction with X-ray techniques, but synchrotron radiation circular dichroism (SRCD) as a complementary method plays a valuable role in structural biology and biophysics research. While it is clear that CD data are no substitute for the high resolution information obtained by protein X-ray crystallography, SRCD can provide static and dynamic information on the secondary and tertiary structure of proteins in a more natural environment, such as aqueous buffers or lipid vesicles. Furthermore, it can offer access to structural information of precious proteins for which only small amounts of material are available and of systems such as membrane and fibril forming proteins, or intrinsically disordered proteins that are difficult to be analyzed by high-resolution methods. During the last decade SRCD has been a rapidly growing technique for structure analysis of proteins, carbohydrates, nucleic acids and other chiral biomaterials [1] with currently 12 beam lines in operation worldwide.

The SRCD beam line UV-CD12 at the ANKA synchrotron (Karlsruhe, Germany) covers the VUV to near-UV spectral range and provides a flux of 1×10^{12} ph s⁻¹ (@ 200 nm, 170 mA). Originally, this beam line had been conceived and designed by the Centre for Protein and Membrane Structure and Dynamics (CPMSD), a consortium of U.K. structural biologists. It was constructed at the SRS synchrotron facility of the Daresbury Laboratory and was open for users since 2003. Following the closure of SRS in August 2008, the beam line was transferred to ANKA in 2009, and after installation and commissioning it has become active again in August 2011 for the SRCD community. It is now operated by the IBG-2 as a so-called collaborative research group (CRG) beam line to continue its working life. Since its reopening UV-CD12 has repeatedly attracted 5 KIT internal and 10 external user groups from German and international universities, which have performed conformational studies on di-

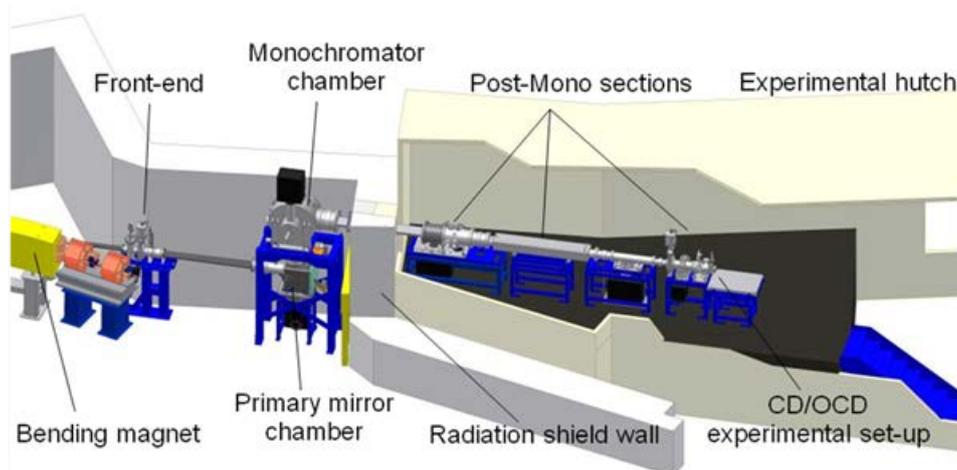


FIGURE 1. 3D sketch of the UV-CD12 beam line at the ANKA synchrotron (Karlsruhe, Germany).

verse biomolecules such as myelin-related proteins (MBP, P2 and CNPase), voltage-gated sodium channels, the heart muscle protein α -tropomyosin, collagen nanofibers or antimicrobial peptides.

Details on the main beam line components of UV-CD12 and its current experimental end-station at ANKA will be presented. Special emphasis will be put on the method of oriented circular dichroism [2] that has been developed and applied for many years in our institute using a bench-top spectropolarimeter, and which is currently implemented as a new experimental station at UV-CD12. Synchrotron radiation-based OCD will be a valuable technique for getting a global view on secondary structure, alignment and aggregation behaviour of peptides in oriented anisotropic lipid membranes, i.e. hydrated lipid bilayers that are macroscopically oriented with respect to the light beam. Examples of SRCD applications in structural biology research, e.g., secondary structure analysis of proteins, which are active in the cell membrane (PDGF β -receptor, E5 oncoprotein) and investigations on their orientation in aligned lipid bilayers determined by OCD, as well as conformational studies of membrane-active antimicrobial peptides (SB056, KIGAKI) in lipid environment and thermal denaturation experiments will be presented.

REFERENCES

1. B. A. Wallace, Protein characterization by synchrotron radiation circular dichroism spectroscopy, *Quarterly Reviews of Biophysics*, **42**, 2009, 317–370.
2. J. Bürck, S. Roth, P. Wadhvani, S. Afonin, E. Strandberg, and A. S. Ulrich, Conformation and membrane orientation of amphiphilic helical peptides by oriented circular dichroism, *Biophysical Journal*, **95**, 2008, 3872–3881.