November 8, 2023

Professor Nobuhiro Kosugi
Director
Institute of Materials Structure Science
High Energy Accelerator Research Organization
Tsukuba, Japan

Re: **Evaluation of the Structural Biology Research Center, IMSS, KEK**

Dear Professor Kosugi,

Upon invitation of the IMSS management, I had a privilege to participate in a two-day presentation and discussion of the activities of the Structural Biology Research Center, IMSS. The meeting took place on October 16 and 19, 2023. Based on the presentations by staff members of the SBRC, as well as on the written activity report covering the years 2020-2023, I would like to provide my assessment of the progress achieved by SBRC during the last 3½ years since the previous evaluation.

As already discussed in the previous report, SBRC is principally responsible for the structural biology portfolio of KEK. SBRC plays a double role – it is responsible for the management of the structural biology pipeline for the university-based and company-based scientific community, as well as conducting its own research projects. With the scientific staff of ~45, supported by 5 administrative personnel, SBRC is tasked with providing access to the state-of-the-art protein expression, purification, and characterization facility, operation of five Photon Factory (PF) synchrotron beamlines devoted to protein crystallography (MX), as well as two beamlines for biological small-angle scattering (SAXS). Additionally, SBRC is in charge of the operation of an electron microscopy facility that utilizes two high-performance instruments.

The main change of the scope of operation of SBRC since the last site visit was the vast expansion of its abilities to conduct research using the methods of single-molecule cryo-EM. This was accomplished by construction of a new research building that now houses two instruments, a 200 kV Talos Arctica and 300 kV Titan Krios. The former machine has been in operation since 2018 and has now been moved into its new location, whereas the latter one was installed only in 2022. Nevertheless, access to these instruments for both the SBRC staff and for researchers elsewhere in Japan has already resulted in more than 20 scientific publications during the review period. Some of them were published in top journals, such as Nature, Nature Communications, Molecular Cell, or PNAS. This is quite a remarkable achievement.

Only minor modifications and improvements have been made to the biochemical laboratories and the crystallographic facilities of the SBRC. The protein expression, purification, and characterization laboratories found an additional home in the recently constructed building that also houses the cryo-EM facility. These laboratories are equipped with a variety of instruments

for protein purification and characterization, and I do not see any important gaps in that instrumentation. The cryo-AM facility has also all the tools needed to prepare samples, including some home-made instruments such as a small plunger for rapidly freezing grids in an anaerobic chamber.

Major instruments for crystallographic and SAXS studies have undergone some improvement. A dedicated goniometer for *in-situ* data collection has been installed on beamline BL-17A and a robot for automatically changing plates is being installed. Software capabilities for evaluating diffraction in crystallization plates, as well as for room temperature crystallography, are being upgraded. These are important developments for keeping the facility on a competitive level.

Bio-SAXS facilities have also undergone significant improvements, both in the area of hardware, as well as in extending the capabilities of software. It is now possible to conduct time-resolved SAXS studies using a prototype microfluidic cell. Software packages such as SAngler and MOLASS, developed at SBRC, are used both in-house and in other laboratories.

Ever since its establishment over 20 years ago, the SBRC has been playing a dual role by, first, providing unique facilities for researchers from a number of institutions within Japan and also from other countries, as well as by conducting high-quality in-house research. Research efforts by the SBRC staff have been very successful, leading to around 100 publications in Japanese and international journals (mostly the latter ones) during the review period. Many of these publications appeared in the top journals in the field of structural biology, testifying to the high quality of the underlying science.

Support for the research activities of SBRC has been provided mainly by the Ministry of Education, Culture, Sports, Science and Technology (MEXT), largely through the BINDS program. This support allows no-cost utilization of the facilities by academic users in Japan, as well as some foreign visitors. An important component of the funding is support from industrial entities that gain paid access to SBRC facilities. It is important to note that some of the development projects use the latter funding mode. An example is development of new types of crystallization plates with very thin window that would enhance the capabilities for *in-situ* data collection on a PX beamline.

Although SBRC can be, in general, considered as being very successful, some potential future problems have been identified during the review. One of them is staffing that is not fully adequate for the number of facilities that need to be supported. For example, only three scientists are responsible for operation of five PX beamlines – even with the very high level of automation this is not a sustainable model. For comparison, each sector of the APS synchrotron at the Argonne National Laboratory, providing X-rays to two or three beamlines, is supported by between 10 and 12 scientists. With the current switch to cryo-EM as the preferred technique in investigation of macromolecular structures cutting the number of beamlines to 4 or even 3 might still provide sufficient support for the users without undue pressure on the beamline staff.

It needs to be stressed that Photon Factory is one of the oldest operating PX facilities in the world that has not undergone extensive modifications. Such modifications are probably overdue, although the problem affects not only SBRC, but also other uses of this synchrotron. However, proposals for increasing the flux and brilliance of the radiation source are beyond the scope of this review. Another problem that is becoming clear is that some major instruments, such as the anaerobic chamber that is over 20 years old, are becoming difficult to maintain and may need

replacement. It is clear to me that SBRC will need to be able to secure sufficient funding not only for the new projects, but also for maintaining what is already in place.

I would like to add another general comment. It is my understanding that, as an accelerator research organization, KEK operates large-scale facilities and associated experimental equipment and provides machine time to researchers for their own research. Typically, users analyze the obtained data in their own laboratories. However, due to the specific nature of life sciences, providing machine time alone is not sufficient in the field of life sciences. Therefore, large-scale accelerator organizations around the world (for example, DESY in Germany and SLAC in the United States) have established dedicated life science research centers to support users. Although the current IMSS support for the development of the SBRC in accordance with the trend in the world is highly commendable, it is important to recognize that the resources of the IMSS have limitations in meeting current needs. It is my strong recommendation that KEK as the parent organization needs to make more efforts to promote the life sciences.

Despite all of these comments, I still am confident that the future of SBRC is quite bright. I was positively impressed by efforts such as creation of a virtual cryo-EM facility that would allow users unimpeded access to instruments located in a number of sites and not only to the ones operated by SBRC. Efforts to cooperate with the other synchrotron facilities in Japan and elsewhere are also laudable. Although Photon Factory may lose some of its international users due to becoming a bit obsolescent, it is still crucial for assuring progress for structural biology research in Japan. The move to create a unified, virtual cryo-EM facility is a step in the right direction. With strong management and very capable staff I am confident that the future of SBRC is assured.

Sincerely,

Alexander Wlodawer, Ph. D.
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Cc: Professor Toshiya Senda