

The 5th Review Committee of Experimental Animals (B)
Evaluation and Suggestions

(January 22, 2019)

Division / Team Name: Bioresource Engineering Division

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1. Achievements and plans for the Division

- (1) Have the current achievements reached the standards of those made by the major international bioresource centers?
 - The Division is developing various technologies that are necessary for improved efficiency and advancement of Bioresource Infrastructure Projects. Furthermore, the fact that they continue to be an internationally accredited division is highly valued.
 - In all the following novel basic technological development projects: 1) improvement of cryopreservation technology, 2) novel development of micro-insemination (ICSI) technology, 3) development of nuclear transfer cloning techniques, 4) development of novel stem cells and model animals, and 5) developmental biology research, the Division has had an excellent accomplishments and the Division is releasing information on all developmental bioengineering technologies to all over the world.
 - Induction of superovulation from rare female mice through anti-inhibin antibodies and estrous cycle synchronization as well as the improvement of micro-insemination (sperm injection) and nuclear transfer cloning techniques are considered to be internationally accredited and highly ranked.
 - The originality of the newly developed resources is highly regarded, including novel mouse models of allergies generated by nuclear transfer cloning using antigen-specific T cells, genome editing using a wild-derived strain (MSM), and knockout hamster lines etc.

- (2) Have sufficient achievements been made for contributing to society and to the research community within Japan and overseas?
 - The Division is supporting the research community by providing internationally accredited knowledge and techniques in reproductive engineering research and the Division is making a large contribution to the research community within Japan and overseas.
 - The Division has contributed to various areas of the research community through press releases, the current technology on the homepage, lectures in symposia, books, journal editing (chief editor for two journals), research collaborations. More specifically, the Division has participated in projects that are developing new methods for the preservation of gametes and

- embryos.
- In technical training backed by their advanced techniques, including a fundamental course on the cryopreservation of mouse sperm and embryos as well as a practical course on micro-insemination, the Division is meeting the needs of society and the research community. This has been highly valued by participants.
 - The Division has contributed to society by teaching many graduate students and researchers, accepting high school and university student visitors, as well as providing outreach activities such as lectures for graduate students in universities.
- (3) Are current activities and plans based on the results of the 3rd Mid- to Long-Term plan or the achievements in the previous position? Are they in line with the BRC's 4th Mid- to Long-Term Plan (7 years from 2018 to 2024)? Are they appropriate and do they contribute to the development of the center?
- In the BRC's 3rd Mid to Long-Term Plan, the Division made solid and satisfactory achievements which were highly evaluated. The Division's current and future plans are based on these achievements, in line with the BRC's 4th Mid- to Long-term Plan. Their plans target the continuation and development of the following six areas of technological developments/researches: 1) cryopreservation, 2) micro-insemination, 3) nuclear transfer cloning, 4) generation of novel stem cell lines, 5) generation of novel animal models, and 6) developmental biology. The Division's current and future subjects to be addressed have been clarified and incorporated into appropriate plans. It is anticipated that they will contribute to the development of the center in the future.
 - We look forward to the success of the research objective, positioned by the PI as challenging. In particular, the identification of the factors regulating the genome plasticity of the 129 strain and the results from the TS cell research are expected to be highly influential.
 - Through the emergence of the CRISPR/Cas9 system, it is necessary to provide gene modification techniques and to establish research resources in animal species other than mice. The Division has succeeded in the generation of a genome-edited hamster and the Division is expanding studies to take advantage of the hamsters' characteristics, which will undoubtedly contribute to the center's Mid- to Long-term development.
- (4) What are resources to be developed and research/ technological development to be undertaken in addition to those currently planned in the initial 4th Mid- to Long-Term Plan?
- Whilst male germline stem cells are consistently established and supplied, female germline stem cells have not been established so far. Their plan to establish the female germline stem cells from female primordial germ cells (PGC) is challenging, but if this is achieved, it will bring a significant impact.
 - The transfer of established technologies to domestic research institutions is an important

objective for raising their level.

- The development of anti-inhibin monoclonal antibodies will make a large contribution to the BRC and outside researchers. It will also be important for animal welfare. Therefore, it is hoped the Division will complete it by all means.
- In the field of genome editing, it is recommended that the Division will promote the advancement in basic technologies, such as improvements of micromanipulation and electroporation technologies, which may lead to the development of novel and unexpected application of technologies in embryology research.
- It is recommended that technical support and guidance for production of genetically modified hamsters will be provided by the center, as this technique cannot be easily acquired by other researchers.

2. SWOT analysis

(1) Are the results of the presented SWOT analysis valid?

- They are adequate.
- The Division's analysis on the fact that each staff member is burdened with a little excessive number of themes, which was pointed out previously, and their measures against it are considered to be adequate.
- Although the research objectives have been organized, we think it is true that the PI is currently overworked.

(2) Are the countermeasures for the results of the SWOT analysis appropriate?

- It is appropriate.
- Steady plans are being made based on the achievements that they have made.
- The PI keeps considering introducing the recent acceleration of advanced analyses such as next generation sequencers into the embryology field, as was recommended in the last committee meeting.
- Reducing the PI's duties of academic societies is an effective countermeasure. However, as members of academic societies, we would like to ask him for continued contribution to some extent.

3. International collaboration

(1) Is the international collaboration being actively addressed, and is the Division functioning as a hub of international science and technology?

- The Division is actively carrying out international research collaborations, which leave nothing to be desired more.
- The PI of the Division was committed to the operation of the World Congress on Reproductive Biology 2017 as the program chairperson and was appointed to the chief editor of international journals as well. These activities are recognized as significant contribution.
- The Division is accepting and sending many researchers, and thus, functioning as an international hub for science and technology.
- The Division is playing a pivotal role and is an important laboratory in this country. The PI is important researcher for Japan's germ cell research.

4. PI assessment

- (1) Is the PI fulfilling the role in line with the BRC mission?
 - The PI is developing highly original reproductive bioengineering technologies and providing them within BRC as well as outside of BRC, and his achievements have exceeded our expectations.
 - As the core of the BRC, the PI can be evaluated as fulfilling the role in line with the mission to a high standard in regard to the fundamental technology of bioresource engineering and bioresource-related research and development. We hope he will publish his achievements of international level in this Mid- to Long-Term Plan.
 - As the PI's global position shows, his nuclear transfer cloning technology has a reputation for being highly specialized. We would like to recommend that he should have a vision that focuses on the further technical development of nuclear transfer cloning, its potential development as a bioresource, and the utilization of these techniques for other subspecies-derived mouse strains other than B6. We look forward to the future applications of the memory T cell clone mouse.

- (2) Do the PI's achievements in research and development (R&D) satisfy international standards in light of the following three aspects? (i) Results output and impact, (ii) Contribution to specific missions of each laboratory regarding research support and collaborative exchange programs within RIKEN, (iii) Pioneering new fields of research, acquisition, and commercialization of intellectual property rights, social education for science, the fusion of different fields, and social contribution
 - (i)
 - The impact of the stable cloning technology is highly regarded internationally, and the technology is satisfactorily contributing to the strengthening of the collaboration within RIKEN.

- In recent high-impact paper, the Division used epigenetic techniques to clone neural cells that previously could not be cloned (Mizutani et al. Biol Reprod 2015;92:81). The Division is consistently improving cloning technology, including in the aforementioned paper, and is internationally outstanding research group for cloning technology.
 - The impact brought by anti-inhibin antibody utilization meets the international standards.
 - (ii)
 - This Division functions as a hub for domestic germ cell research, and the PI is irreplaceable with other researcher because of his unique contribution.
 - On the PI's support to the research conducted in other facilities and his involvement in the All RIKEN Project, we would like to see definitive descriptions on the collaborations and the output.
 - (iii)
 - They meet the international standards.
 - Remarkable results have been produced by his recent genome editing research on hamsters. These results could not have been anticipated from the mouse studies. As he reported that this technology has led to interdisciplinary collaboration with researchers in hibernation, further joint research with different researchers can be expected.
 - The PI has planned proliferation of female germ cells, which demonstrates his strong motivation to pioneer new fields of research. It can be expected that the outcome may directly lead to the acquisition of intellectual property rights as well as social enlightenment and contribution.
 - The commercialization of anti-inhibin antibodies can be expected.
 - It is difficult to evaluate the PI's achievements in the fusion of different fields.
- (3) Is the PI appropriately tackling the management and operation of the Division? In addition, does the PI make efforts for training and development of young talent?
- The PI is committed to the management and operation (personnel allocation and role allotment) of his Division appropriately.
 - The PI is committed to the development of young talent. Young researchers in his Division have published their results in original papers. They have won competitive funding and awards, attained positions and are playing active roles. Thus, favorable results of young talent development have been attained.
 - As bioresource engineering techniques are important, we hope that they are succeeded to many young researchers. Also, we hope that the PI will further commit to the acceptance and guidance of graduate students.

End