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Recent Trends in Regenerative Medicine — Regenerative Medicine Utilizing Stem Cells

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1.1 Introduction

In the second Science and Technology Basic Plan, life science was selected as one of the fields having high priority. In the plan, regenerative medicine was referred to as one of the areas in which various challenges should be strategically pursued with high priority in order to tackle national and social problems. In addition, there have been heated arguments about bioethical issues arising in regenerative medicine and regenerative therapy at the Council for Science and Technology Policy, Cabinet Office, etc. In this article, "regenerative medicine" is defined as a term referring to both research and therapy ("regenerative therapy") with the use of regenerated tissues.

1.2 Enlarged possibility of applying regenerative medicine

Various regenerative therapies have been conducted so far, including transplantation of regenerated skin and bone marrow as well as transplantation of regenerated dopamine-producing cells into the brains of patients with Parkinson's disease. However, there is a crying need for every type of tissue to be transplanted.

Use of stem cells is one of the solutions for this problem. Stem cells are the cells that have the ability both to self-replicate and to differentiate into progenitor cells, which then differentiate or develop into many different "mature" cell types that have characteristic shapes and specialized functions. Among the stem cells, somatic stem cells, which are thought to have the ability to differentiate into cells of specific tissues or organs, as well as embryonic stem cells, which have the ability to differentiate into any types of cells in tissues and organs, are expected to create cells, tissues or organs for transplantation and contribute to health care in the future. Moreover, possibility has been suggested that stem cells, in combination with cloning technology, will be applied to the creation of tailored organs for transplantation not causing rejection reactions.

Among the two types of stem cells, embryonic stem cells are harvested by extracting the inner cell mass (a mass of cells that will develop into the baby's tissues and organs) from the center of a human embryo in the first week of life, resulting in the destruction of the embryo. Therefore, deliberate arguments have been made about the bioethical aspects of the use of embryonic stem cells for experimental or therapeutic purposes.

At first, as shown in Table 1, this article introduces; i) the progress in research using somatic stem cells and embryonic stem cells from the viewpoint of regenerative medicine (Section 1.3). Then, from the viewpoint of regenerative therapy, this article describes; ii) the impact of regenerative therapy on health care cost (Section 1.4); and iii) significance of the establishment of foundations

Table 1: Contents of this feature report

Contents of this article
i) Progress in research using somatic stem cells and embryonic stem cells (Section 1.3)
ii) Impact of regenerative therapy on health care cost (Section 1.4)
iii) Significance of the establishment of foundations on which to conduct regenerative therapy (Section 1.5)
iv) Bioethical issues concerning research using human embryonic stem cells (Section 1.6)
v) Relationship between the knowledge possessed by the people in the scientific community and the knowledge which can be shared in society (Section 1.7)

on which to conduct regenerative therapy (Section 1.5); followed by, iv) bioethical issues concerning research using human embryonic stem cells (Section 1.6); and finally, in Section 1.7, v) the relationship between the "knowledge possessed by the people in the scientific community" and the "knowledge which can be shared in society," which often has great significance especially in studies in the field of regenerative medicine.

1.3 Progress in research in the field of regenerative medicine

Even in this postgenome era, no viable cells can be artificially created from scratch. In any event, some diseases require treatment using viable cells. For example, even in this era of postgenome research, blood transfusion and bone marrow transplantation cannot be replaced by therapy without using viable cells.

While the applicability of cell transplantation as regenerative therapy for the treatment of deficiency (loss) of cells or for tissue damage is currently expanding, cells for use in such therapy is desperately lacking. If intended cells can be prepared in vitro, such shortage will be alleviated. Under these circumstances, new findings and technologies have come into the world, which seem to promise to expand the applicability of regenerated cell transplantation. Specifically speaking, the following studies are being pursued.

1.3.1 Recent studies on somatic stem cells

(1) Studies aiming to develop therapeutic methods for Parkinson's disease

Parkinson's disease is linked to the death of brain cells that produce dopamine in the substantia nigra. With the purpose of treating Parkinson's disease, fetal brain cells have been transplanted to affected patients to complement the function of dead brain cells. However, because cells to be transplanted should be collected from several fetuses for the treatment of one patient, and because it is technically difficult to extract specific cells exclusively from brain tissues removed from aborted fetuses, fetal brain cell transplantation has not gained popularity as a therapy for Parkinson's disease.

With this being the situation, many universities, companies, etc., are exploring proteins (markers) specifically expressed in progenitor cells for dopamine-producing neurons, which can be transplanted to Parkinson's disease patients for therapeutic purposes, in murine models of Parkinson's disease. It is expected that enough dopamine-producing neurons to be infused to Parkinson's disease patients for therapeutic purposes will become harvestable if marker proteins specific to such neurons are identified and utilized.

(2) Studies targeting the recovery of impaired neural functions

In a study using a rat model of spinal cord (cervical cord) injury (associated with decreased mobility of forelegs), a group led by Professor Hideyuki Okano at Keio University transplanted neural stem cells into the injured part of the spinal cord, and observed that the cells differentiated into neurons, etc., and that the neural network was reconstructed, resulting in the recovery of the mobility of the forelegs (Figure 1).

(3) Studies on plasticity of differentiated cells

It has been shown that hematopoietic stem cells are capable of differentiating into neural, muscle or liver stem cells, and that neural or muscle stem cells have the ability to differentiate into hematopoietic stem cells (Figure 2).

If intended cells become able to be prepared in vitro by utilizing the plasticity of stem cells, i.e., by reprogramming a specific kind of stem cells to

Figure 1: Recovery of neural function after the transplantation of somatic stem cells

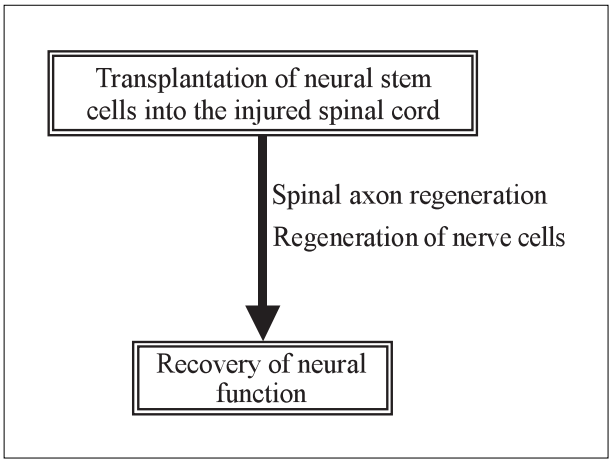
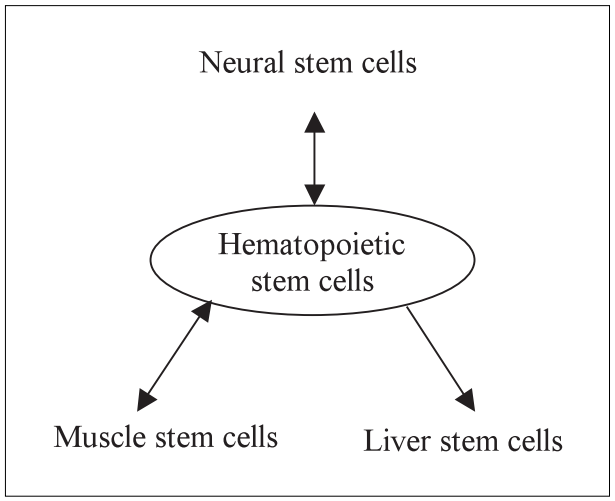


Figure 2: Plasticity of hematopoietic stem cells



Source: Authors' compilation by making reference to the materials provided by Professor Nishikawa

become another kind of stem cells, somatic stem cells are expected to have greater value in the treatment of diseases.

1.3.2 Recent studies on embryonic stem cells

(1) Establishment of embryonic stem cell lines

In 1998, the University of Wisconsin developed the first human embryonic stem cell line with monetary support from Geron Corp., which had a great impact on regenerative therapy (Figure 3).

Studies on embryonic stem cells have been conducted mainly on mice and Primates (Rhesus monkeys and marmosets). The first embryonic stem cell line was established in 1981, 1995 and 1998 for mice, Rhesus monkeys and marmosets, respectively.

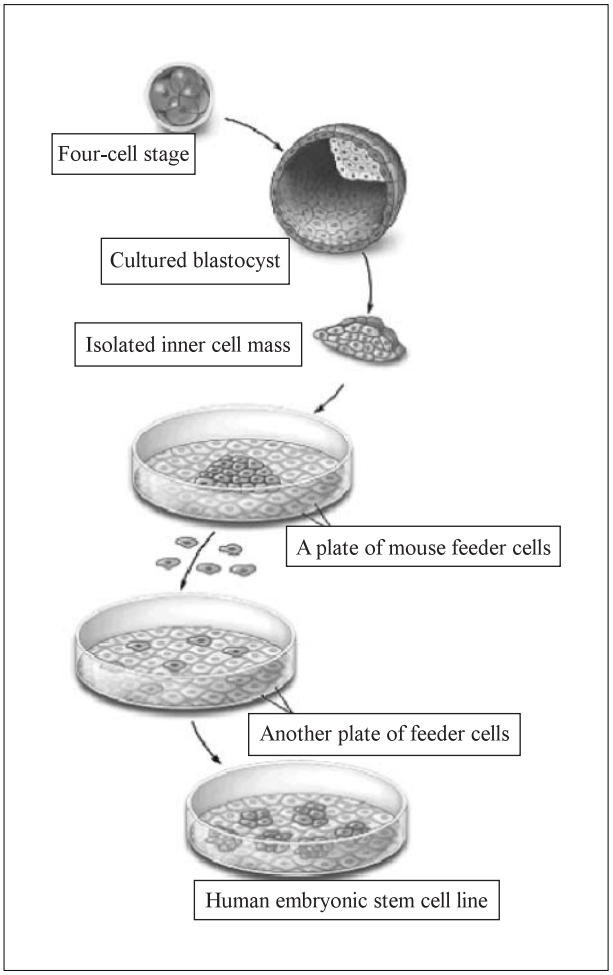
Table 2: Number of established lines of human embryonic stem cells

Goeteborg University (Sweden)	19
CyThera, Inc. (United States)	9
Reliance Life Sciences (India)	7
Monash University (Australia)	6
Karolinska Institute (Sweden)	5
Wisconsin Alumni Research Foundation (United States)	5
BresaGen, Inc. (United States)	4
Technion-Israel Institute of Technology (Israel)	4
National Center for Biological Sciences (India)	3
University of California (United States)	2
Total	64

Number of human embryonic stem cell lines reported to the National Institutes of Health (NIH)

Source: Authors' compilation by making reference to the materials provided by Professor Nishikawa

Figure 3: Establishment of human embryonic stem cells



Source: Authors' compilation by making reference to a figure in the June 2001 issue of the NIH report "Stem Cells."

As mentioned above, the first human embryonic stem cell line was established in 1998. To date, 64 lines of embryonic stem cells have been established in the world (Table 2).

(3) Challenges to be addressed in studies using embryonic stem cells

While it has been shown that embryonic stem cells have pluripotency, i.e., capability to differentiate into various kinds of cells in vitro including nerve cells, muscle cells, blood cells and insulin-secreting cells, the mechanism of differentiation control remains under investigation. The challenges to be addressed in studies using embryonic stem cells include the exploration of factors that induce the differentiation of embryonic stem cells into cells for intended functions, development of techniques for extracting a specific kind of cells from a

mixture of various kinds of differentiated cells, as well as the development of technology for inducing the efficacious proliferation of cells in vitro.

With regard to the achievements in recent studies on mouse embryonic stem cells, it has been shown that a certain transcription factor (Oct-3/4) is involved in the maintenance of undifferentiated state of embryonic stem cells. In addition, a group led by Professor Yoshiki Sasai at the Graduate School of Medicine at Kyoto University has developed a technique called the SDIA (stromal cell-derived inducing activity) method and succeeded in frequently inducing and culturing the development of mouse embryonic stem cells into dopamine-producing nerve cells in vitro, which can be used to treat Parkinson's disease.

1.4 Impact of regenerative therapy on health care cost

General concerns are rising that widespread introduction of advanced medicine into clinical practice including regenerative therapy might lead to an increase in health care cost, but there are some cases where introduction of regenerative therapy does not seem to increase health care cost.

Dr. Ron Mackay at the National Institutes of Health (NIH) in the United States has developed a technique for inducing the differentiation of embryonic stem cells into pancreatic cells, which could be successfully administered to diabetes mellitus model mice to treat the disease (This technique presents some problems in that the process of differentiation of embryonic stem cells into pancreatic cells cannot be completely controlled, and requires further study).

Patients with type I diabetes mellitus, whose pancreas (β-cells in the pancreas) cannot synthesize or secrete insulin, develop the disease at the age of less than 15 years old almost without exception, and should receive the administration of insulin throughout their lifetime after the onset of the disease. If type I diabetes mellitus can be completely treated with only a single shot of cells, health care cost will be cut. As this example illustrates, application of regenerative medicine to actual clinical practice may not always result in an increase in health care cost.

From this time forward, discussions will take place in various quarters about the impact of the introduction of regenerative therapy on health care cost.

1.5 Significance of the establishment of foundations on which to conduct regenerative therapy

1.5.1 Pittsburgh in the United States as an example

Pittsburgh is one of the cities that succeeded in industrialization though the introduction of regenerative medicine. About a half of the cases of liver transplantation in the United States is conducted in Pittsburgh. In Pittsburgh, health care, education, etc., are provided under the leadership of the organ transplantation centers, and service industries have been actively developed in a wide variety of fields including organ transplantation (Table 3).

Factors which have contributed to the success of Pittsburgh as a tissue engineering industry city include; i) powerful motivation has been shared among Pittsburgh citizens to resurrect the city's faltering economy in the wake of the decline in the steel industry as well as, ii) the city's established intellectual infrastructure (For example, Dr. Starzl, a world-renowned

Table 3: Tissue engineering industry in Pittsburgh

	Total value for regenerative therapy-related companies in Pittsburgh
Number of related companies	26
Total market capitalization or valuation (estimated)*	4.3 billion dollars
Annual gross sales (estimated)	0.774 billion dollars

Source: Authors' compilation by making reference to the materials on the investigation conducted by the Pittsburgh Tissue Engineering Initiative in 2000, http://www.pittsburgh-tissue.net/industry/pdf/industry.pdf.

transplantation surgeon is a citizen of Pittsburgh, and many Japanese transplantation surgeons have visited him to receive training in transplantation techniques).

1.5.2 Trends in Japan —creation of foundations in the Osaka area

In Osaka and Kobe areas, there are universities, institutions and companies that have first-rate experts in the fields of research on physiologically active substances, research in embryology, research in regenerative medicine, transplantation medicine, research on cloning, research on tissue engineering, etc. Therefore, a project for health-care industrialization of the areas, under the initiative of Kobe City, has been proposed. This project has been contracted out as one of the district-oriented collaborative research projects funded by the Japan Science and Technology Corporation (JST), which is due to be completed in 5 years starting from 2000, and is aiming to build up comprehensive technological foundations for research on, and application of, regenerative medicine.

At present, collaborative efforts are being put forth by the Institute of Biomedical Research and Innovation (IBRI) of the Foundation for Biomedical Research and Innovation, which holds the core facility of the Kobe medical industry development project, the Center for Developmental Biology of the Institute of Physical and Chemical Research, the Tissue Engineering Center of the National Institute of Advanced Industrial Science and Technology, the Institute for Frontier Medical Sciences of Kyoto University, the Exploratory Medical Center at the Kyoto University Hospital, the Center for Future-oriented Medicine of Osaka University, and other related hospitals, etc. Under the current circumstances, the challenge to be addressed from now on is to let the collaboration system continue to function efficiently.

In addition to the project mentioned above, efforts shown in Table 4 are being made with the aim to establish international foundations for research in life science in the Osaka area.

Table 4: Efforts aiming to establish international foundations for research in life science in the Osaka area

(1)	**Establishment of integrated foundations in the northern area of Osaka and the Kobe area.** Enhancement of the functions as foundations for research, intensive enforcement of measures to support industrialization, etc.
(2)	**Facilitation of cooperation among integrated foundations for research in life science under the initiative of the Osaka and Kobe areas.** Development of a system to promote research though collaboration among government, academia and industry, and construction of high-capacity, high-speed information networks, etc.
(3)	**Provision of opportunities for deliberation by the authorities concerned, etc., to intensively facilitate comprehensive support.**

1.6 Bioethical issues concerning research using human embryonic stem cells

As discussed, in countries with advanced medicine including Japan, ardent expectations are placed on the application of embryonic stem cells for health care, while, deliberate arguments have been made about the bioethical aspects of the use of embryonic stem cells for experimental or therapeutic purposes.

Because human embryonic stem cells are derived from the inner cell mass removed from blastocysts, which have developed from fertilized embryos, early embryos are inevitably destroyed to obtain human embryonic stem cells (Figure 3).

Therefore, some researchers feel that an early human embryo intended to be destroyed to obtain embryonic stem cells is a mere cluster of cells, but other researchers believe that a human embryo should be regarded as a human being at the time of fertilization or at a certain stage of embryonic development, and that harvesting of human embryonic stem cells equals intentional destruction of a human being (Figure 4).

The clue to addressing the bioethical problem is given by the construction of an appropriate decision-making system. For example, such decision-making system can be built up by the disclosure of detailed information by researchers to the public as well as the provision of opportunities for discussions by people with different values agreeing to disagree, followed by the formulation of fixed rules. Concerning the disclosure of information by researchers to the public, discussions are held in the following

chapter.

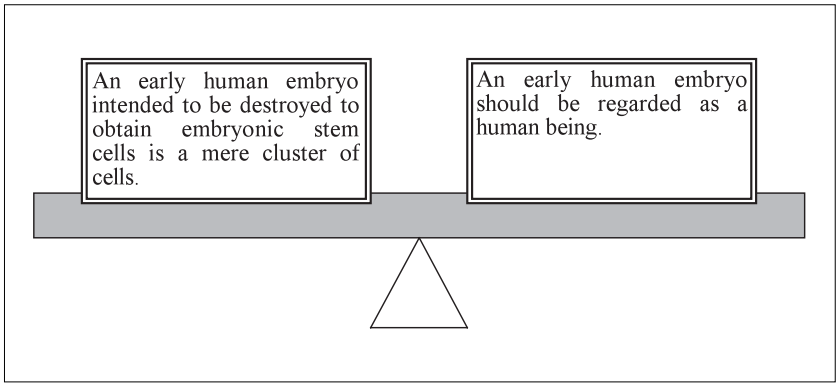
The first public discussion about ethical issues concerning research using human embryonic stem cells was held at the Human Embryo Research Subcommittee meeting under the Bioethics Committee of the Scientific Council. During the discussion, opinions were exchanged about bioethical aspects of studies using human embryos, mainly human embryonic stem cells, and the "Ethical Principles of Research on Human Embryos, Mainly Human Embryonic Stem Cells " were formulated.

Under the principles, it is specified that establishment of human embryonic stem cell lines should be conducted with great care, and can be performed only within a strictly constructed conceptual framework in consideration of both the benefit of research using human embryonic stem cells and the problems associated with the harvest of those cells, i.e., destruction of the early embryo as early human life. With regard to the use of established embryonic stem cell lines, it is specified that a conceptual framework should be developed because uncontrolled use could lead to the indiscriminate use of human embryonic stem cells, accelerate the destruction of human embryos in vain, and give rise to ethical issues derived from the pluripotency (capability to differentiate any kinds of cells) of such cells. Concerning the clinical study using human embryonic stem cells, it is specified that due consideration should be taken in terms of safety of clinical practices with the use of such cells, and that clinical studies on the introduction of human embryonic stem cells, or cells or tissue differentiated from such cells into an individual human body, should not be conducted until guidelines on clinical application of embryonic stem cells are developed.

In response to the formulation of the "Ethical Principles of Research on Human Embryos, Mainly Human Embryonic Stem Cells," the Ministry of Education, Culture, Sports, Science and Technology prepared the draft of the "Guidelines for the Establishment and Use of Human Embryonic Stem Cells" and invited comments about the guidelines from the public. Thereafter, the Ministry sought the advice of experts participating in the Council for Science and Technology Policy, Cabinet Office, about the draft guidelines in September 25, 2001. Then, following repeated discussions held under the leadership of the Bioethics Review Panel of Experts, which is a subordinate organization of the council, a report on the opinions of experts was submitted to the Ministry of Education, Culture, Sports, Science and Technology.

In this context, the "Guidelines for the Establishment and Use of Human Embryonic Stem Cells" was put into effect by the Ministry of Education, Culture, Sports, Science and Technology in September 25, 2001. These guidelines specify that human embryonic stem cells shall be handled with integrity and care lest the sanctity of human life should be violated. In addition, the guidelines provide that the establishment and use of embryonic stem cell lines shall be restricted to basic research for the time being (Article 2, Section 2). Furthermore, it is also stipulated in the guidelines that practical application of human embryonic stem cells such as the manufacturing of pharmaceutical products derived from such cells for use in clinical practice as well as the

Figure 4: How should the human embryo be interpreted?



Source: Authors' compilation by making reference to the materials provided by Professor Nishikawa

Table 5: Moves to address bioethical issues about the establishment and use of human embryonic stem cells in major industrialized nations

Country name	Date	Moves to address bioethical issues about the establishment and use of human embryonic stem cell lines
Japan	September 2001	According to the "Guidelines for the Establishment and Use of Human Embryonic Stem Cells," the establishment and use should be restricted to basic research for the time being.
Germany	1990	Under the Embryo Protection Law, people are absolutely prohibited from conducting any research using human embryonic stem cells.
United Kingdom	January 2001	Under the Human Fertilisation and Embryology Act, people are allowed to establish human embryonic stem cell lines from cloned human embryos.
United States	August 2001	Under an executive order, public monetary support may be provided to studies using human embryonic stem cells, but establishment of a new human embryonic stem cell line is prohibited.
France	———	Under the Bioethics Law, research on human embryonic stem cells excluding observations of such cells is prohibited. An amendment to the Bioethics Law is planned to be submitted to allow scientists to establish human embryonic stem cell lines from spare embryos.

massive supply of such cells to be used in drug toxicity tests, shall not be conducted for the present time.

Table 5 provides a summary of moves to address bioethical issues about the establishment and use of human embryonic stem cells in major industrialized nations.

1.7 Conclusion

To date, scientists have provided the general public with knowledge derived from scientific evidences they acquired in their research activities. In other words, scientists have

Figure 5: Process of the development of knowledge possessed by the scientific community into that shared among the public

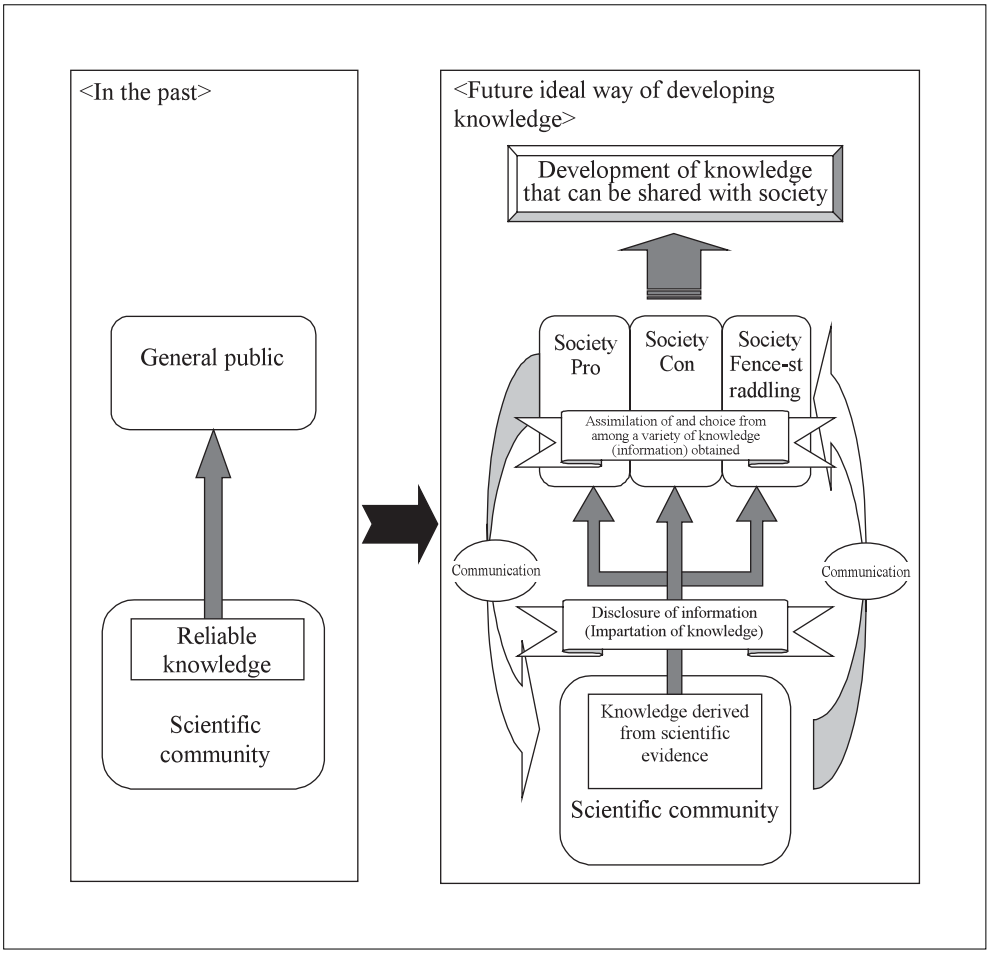


Figure 6: Requirements for the promotion of the development of knowledge within scientific circles into that shared among the public

Candid disclosure of information is required from experts in the field of life science

1. Sense of responsibility as, and candid disclosure of information by, scientists.
   - Establishment of the system of on-site observation tours to laboratories intended for people in the mass media, economic world and judicial world.
   - Improvement in communication between the scientific community and society.
2. Establishment of institutions as providers of life science-related information, which are accessible to everyone.

Source: Authors' compilation by making reference to the materials provided by Professor Nishikawa

conferred a benefit on society in that they have provided information that can be accepted as "reliable knowledge" by the general public, which will eventually bring profit to society by way of practical application.

Future challenges to be confronted by scientists, particularly in researches in the field of regenerative medicine, are to; build up and accumulate knowledge that can be shared among the general public, disclose information (knowledge) derived from their studies with high transparency, and communicate with the general public, which may have various opinions and may favor or oppose a certain subject in question or take a fence-straddling position (Figure 5).

In addition, it is desirable that institutions be established from which information digested into a more easy-to-understand form is continuously disseminated among the general public (Figure 6) (For example, at the Center for Developmental Biology [CDB] of the Institute of Physical and Chemical Research, discussions have been held by the Institutional Review Board (IRB) not only about various bioethical issues but also about how information obtained in research activities at the institute should be disclosed from the viewpoint of outsiders).

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