relevant laws and notifications, such as "Standards for Biological Raw Materials" (Notification No. 210, Japanese Ministry of Health, Labor, and Welfare, 2003; note that a new version will soon be issued). It is particularly important to sufficiently evaluate information related to the inactivation and elimination of viruses and to indicate measures for ensuring retrospective and other studies.

(1) When culturing cells

(i) Indicate the appropriateness of all media components, additives (serum, growth factors, antibiotics, etc.), and reagents, etc. used in the treatment of cells and set specifications if necessary. Give consideration to the route of clinical application, etc. of the final product when setting specifications concerning the appropriateness of each component.

(ii) Take into consideration the following points with respect to media components:

(a) The ingredients and water used in media should be of high quality and high biological purity, and their quality should be controlled at standards equivalent to those used with pharmaceuticals and pharmaceutical raw materials.
(b) Provide information on all ingredients in the media, as well as the rationale for their selection, and, if necessary, the quality control and other procedures. However, widely known and commercially available media products such as DMEM, MCDB, HAM, and RPMI are regarded as a single raw material set.
(c) Conduct sterility tests and performance tests on media that contain all components in order to determine whether they are suitable as target media. Set specifications for any other relevant parameters thought to be controlled in the process and perform proper quality control.

(iii) Heterologous serum or components derived from heterologous or homologous serum shall not be used unless they are essential for processes such as cell activation or cell growth. In particular, for products that may be used repeatedly, investigate, to the extent possible, ways to avoid using these serum components. If the use of serum or other such material is unavoidable, consider the following points and investigate ways to prevent the contamination of serum and other products and the spread of bacteria, fungi, viruses, and abnormal prions, as well as treatment methods for their elimination, to the extent possible, from the final product.

(a) Clarify the origin of the serum or other component.
(b) Make strenuous efforts to minimize the risk of prion infection, such as by strictly avoiding the use of serum from areas or regions with known outbreaks of bovine spongiform encephalopathy (BSE).
(c) Only use these sera after confirming that they are not contaminated with viruses or other pathogens by conducting appropriate tests to prove the absence of specific viruses and mycoplasma that originate in animal species.

(d) Perform appropriate procedures to inactivate and eliminate bacteria, fungi, and viruses to an extent that does not impact the activation and growth of the cells. For example, to avoid the risks associated with latent viral contamination, perform combinations of heat treatment, filtration, irradiation, and/or UV treatment, if needed.

(e) Preserve and store a portion of the serum used in order to monitor for viral infections in cultured cells, monitor for outbreaks of viral diseases in the patient, and measure antigen production in response to a component of the heterologous serum used.

(iv) When using feeder cells, conduct a quality evaluation while referring to "Derivation and Characterisation of Cell Substrates Used for the Production of Biotechnological/Biological Products" (Pharmaceutical Notification No. 873, Ministry of Health, Labor, and Welfare, issued July 14, 2000), "Guidelines on Public Health Infection Issues Accompanying Xenotransplantations" (Notification No. 0709001, Research and Development Division, Health Policy Bureau, Japanese Ministry of Health, Labor, and Welfare, issued July 9, 2002), and "Guidelines on Epithelial Regenerative Therapy Using 3T3J2 Strain or 3T3NIH Strain Cells as Feeder Cells" based on "Guidelines on Public Health Infection Issues Accompanying Xenotransplantations" (Notification No. 0702001, Research and Development Division, Health Policy Bureau, Japanese Ministry of Health, Labor, and Welfare, issued July 2, 2004) in order to prevent the contamination of feeder cells and the spread of bacteria, fungi, viruses, and abnormal prions. Indicate the methods for the inactivation of cell division potential and conditions such as cell density when using the feeder cells. However, if, for example, the feeder cells or equivalent cells are being used in the manufacture of a cell or tissue product that has already been used clinically and whose characteristics and microbiological safety have already been assessed and confirmed, it may be possible to omit the virus tests or parts of other tests by demonstrating the appropriateness of these cells.

(v) Avoid the use of antibiotics as much as possible. However, if the use of antibiotics in the initial stages of processing is deemed indispensable, attempt to decrease their use in subsequent steps as much as possible, and clearly state the appropriateness of their use from perspectives such as the scientific rationale, estimated residual amounts in the final product, and the effects on the patient. If it has been verified that an antibiotic can be adequately eliminated, its use need not be restricted. On the other hand, if a patient has a past history of allergy to the antibiotic used, in principle, this therapeutic method should not be used. If there is no way to avoid the use of antibiotics, administer the final product in question very carefully and obtain informed consent from the patient.

(vi) If growth factors are used, show the appropriate quality control

methods using relevant parameters, such as purity and potency, for which established acceptance criteria and assay methods are employed, in order to guarantee the reproducibility of the cell culture characteristics.

(vii) For media components and other components that are used in processing and that may contaminate the final product, choose components that do not have any harmful biological effects

(2) When combining with noncellular components

(i) Quality and safety of noncellular raw materials

If the final product consists of cells and noncellular components, such as matrix, medical materials, scaffolds, support membranes, fibers, and beads, describe in detail the quality and safety of the noncellular components.

Provide any relevant information concerning the noncellular raw materials, taking into consideration their type and characteristics, form and function in the final product, and an evaluation of their quality, safety, and efficacy from the perspective of the presumed clinical indication. If using materials that are absorbed by the body, perform the necessary tests on the degradation products to assess safety concerns.

With respect to the tests that should be carried out, refer to "Basic Views on Biological Tests Necessary for Regulatory Approval for Manufactured or Imported Medical Devices" (Notification No. 0213001, Pharmaceutical and Food Safety Bureau, Japanese Ministry of Health, Labor, and Welfare, issued February 13, 2003), describe the test results, and justify the use of such raw materials. The use of information obtained from the literature is encouraged.

(ii) Interactions with target cells

Demonstrate the validity of the test methods used and justify the results obtained for the following three items with respect to the interactions between noncellular components and cells in the final product as well as in any intermediate products.

(a) The noncellular components do not have any deleterious effects on the function, growth capability, activity, or stability of the cells in the final product required for the presumed clinical indication or the cells in any intermediate products.
(b) Evaluate to the extent possible any potential interactions between the cells and noncellular components, taking into consideration, for example, the mutation, transformation, and/or dedifferentiation of cells in the final product or cells in intermediate products.
(c) Show there is no loss of the expected properties of the noncellular components in the presumed clinical indication as a result of any interactions between the noncellular components and the cells in the final and intermediate products.

(iii) When using noncellular components to isolate the desired cell

products from the application site

When using noncellular components with the objective of segregating the desired cell products from the application site, confirm their usefulness and safety by referring to (a) through (d) below.

(a) Membrane permeability kinetics and the pharmacological effects of target physiologically active substances derived from the cells in the final product.
(b) Diffusion of nutritional components and excretory products.
(c) Effects of noncellular components on the area near the application site.
(d) When a pharmacological effect of a target physiologically active substance derived from a desired cell product is anticipated and the objective is segregation of the application site and the desired cell product or undifferentiated cells, confirm that the cells do not leak out, which might result from the degradation, etc. of noncellular components.

(3) When cells undergo genetic engineering

When genes are introduced into cells, provide the following details:

(i) For the target gene (specific gene encoding a desired protein or RNA), information related to its structure and origin, the method by which it was obtained, cloning methods, and methods of cell bank preparation, control, renewal, etc.

(ii) Nature of the transgene.

(iii) Structure, biological activity, and properties of the desired protein or RNA derived from the target gene.

(iv) All raw materials, properties, and procedures (transgenic method, origin and properties of the vector, and method of obtaining the vector used for gene introduction) needed to produce the transgenic construct.

(v) Structure and characteristics of the transgenic construct.

(vi) Control and preparation methods for cell and virus banks needed to prepare vectors and transgenic constructs.

For manufacturing methods for transgenic cells, refer to Chapter 2 and other sections of "Guidelines for Ensuring the Quality and Safety of Gene Therapy Pharmaceuticals," which is an appendix in "Concerning Guidelines for Ensuring the Quality and Safety of Gene Therapy Pharmaceuticals" (hereafter referred to as "Gene Therapy Pharmaceutical Guidelines"), published as Notification No. 1062 by the Ministry of Health and Welfare on November 15, 1995. In addition, state the appropriateness of the establishment in accordance with the appendix of the same notification.

On the basis of the law (Law No. 97, 2003) implemented to ensure biodiversity by regulating the use, etc. of genetic recombination organisms, etc., a separate application procedure for evaluation will be required when living organisms, including certain cells, "viruses," and "viroids," are

genetically modified. The following cells are not regarded as living organisms: "human cells, etc." or "cells that have the ability to differentiate, or differentiated cells that are not viable when alone under natural conditions."

Regardless of the guidelines mentioned above, if a gene introduced into cells is used as a reagent in the manufacturing process and does not either chemically or functionally contribute to the final product, it is acceptable to describe, on the basis of current knowledge, how the quality and safety of the gene conform to the intended use.

II. Manufacturing Process

When manufacturing pharmaceuticals and medical devices derived from the processing of autologous human somatic stem cells (i.e., autologous human somatic stem cell-based products), describe in detail the manufacturing method and verify, to the extent possible, the appropriateness of the method, using the items listed below, in order to maintain consistency in the quality of the product.

1. Lot composition and lot control

Indicate whether a lot comprises final products and intermediate products. If a lot comprises both final and intermediate products, establish standardized procedures concerning the composition and control of the lot.

2. Manufacturing method

Provide an outline of the manufacturing method, from the receipt of the cells and tissues to be used as raw materials and through to the isolation of somatic stem cells and the establishment of the final product. Describe the technical details of the process and necessary process control and product quality control.

(1) Test upon receipt

Establish a battery of tests as well as acceptance criteria with which to assess the appropriateness of the cells and tissues that will serve as the raw materials, taking into account the nature of the cells and their intended use. These may include, for example, visual tests, microscopic examination, recovery factor of target cells, cell viability assays, characterization of cells and tissues, microbiological tests, etc. At the stage of initiating clinical trials, provide the actual measured values obtained up until that point with test samples, and propose a provisional set of acceptance criteria based on these values.

(2) Inactivation and elimination of bacteria, fungi, viruses, and other microorganisms

In cells and tissues that serve as raw materials, inactivate and eliminate bacteria, fungi, viruses, and other microorganisms, if needed and whenever possible, to the extent possible without affecting the characteristics (cell viability, phenotype, genetic traits, specific functions, etc.) and quality of the cells and tissues serving as raw materials. State the appropriateness of the measures, procedures, and evaluation methods used, if any.

(3) Tissue disintegration, cell separation, isolation of specific cells, etc.

Describe methods for the disintegration of tissues, separation of cells, and isolation of somatic stem cells, as well as methods for washing, etc. cells and tissues, that are performed in the early stages of manufacturing somatic stem cell-based products from collected cells and tissues. When isolating specific somatic cells, establish methods of cell identification.

(4) Preparation of cells that are a principal component and active ingredient in the final product

Describe the methods used to collect human cells and tissues, to isolate somatic stem cells, and to obtain the cells that serve as the active ingredient in the final product. The methods to be described include the induction of differentiation, the isolation and culture of the desired cells, and the media, culture conditions, culture period, yield of the cells, etc. at each step. Describe the appropriateness of each method.

(5) Establishment of cell banks

When a cell bank is established at any stage during the manufacture of autologous human somatic stem cell-based products, describe the rationale for preparing the cell banks; the methods used to prepare the cell banks; the characteristics of the cell banks; and the storage, maintenance, control, and renewal methods, as well as any other processes and tests performed. Justify the appropriateness of each. Refer to "Derivation and Characterisation of Cell Substrates Used for the Production of Biotechnological/Biological Products" (Pharmaceutical Notification No. 873, Ministry of Health, Labor, and Welfare, issued July 14, 2000) and other documents. However, it is acceptable to omit a portion of the study items, if there is rational reason that the cells are of autologous origin.

(6) Measures to prevent erroneous sampling (mix-ups) and cross contamination during the manufacturing process

It is extremely important to prevent erroneous sampling and cross contamination during the manufacturing process when manufacturing autologous human somatic stem cell-based products. Therefore, describe preventative measures in the process.

3. Characterization of cells that are a principal component and active ingredient in the final product

For cells that are a principal component of the final product, analyze their attributes, such as cell purity (to control contamination by non-target cells), cell viability, morphological characteristics, cell growth characteristics, biochemical markers, immunological markers, distinctive substances produced by the cells, karyotype, and other appropriate genotypic and phenotypic markers. In addition, characterize their biological functions, if necessary. Furthermore, to evaluate the

appropriateness of the culture period and the stability of the cells, use appropriate cell characteristic markers to demonstrate that there have been no unintended changes in cells cultured longer than the proposed culture period. It may be acceptable to perform these studies using test samples obtained from donors who are not patients in place of the products that will be prepared for clinical trial.. On the basis of these test results, identify the critical cell characteristics that should be used when applying the product to a patient. Although comprehensive cell characterization is always desirable, it may not always be possible to characterize the cells fully because there are quantitative and technological limits to sample analysis. Thus, it is acceptable to perform the studies to the extent possible. When cell processing, such as growth within the body, is anticipated after application, clearly demonstrate the functions expected by describing the specified criteria with respect to the passage number or number of cell divisions.

4. Form and packaging of the final product

The form and packaging of the final product shall ensure the quality of the final product.

5. Storage and transport of the final product

If intermediate or final products need to be stored and transported, the storage procedure and duration, the containers used for transport, and the transportation procedure (including temperature control, etc.) shall be stated and their appropriateness clearly indicated (refer to Chapter III).

6. Consistency of the manufacturing procedure

When manufacturing autologous human somatic stem cell-based products, assess during the manufacturing process and for each individual product (or each lot, if any) whether differ significantly with respect to the number of cells, cell viability, and cell characteristics (such as relevant markers of phenotype and genotype, functional characteristics, and the percentage content of the desired cells) and from the point of view of the clinical application method and the intended clinical use of the product. It may be acceptable to use test samples obtained from donors who are not patients in place of the products that will be prepared for clinical trial. Evaluation of intermediate products may provide insight into the appropriateness of cells and tissues used as raw materials and the validity of the manufacturing process up until the intermediate product stage and provide an appropriate guidepost en route to the final product. Therefore, it may be reasonable to adopt such an approach, where necessary and appropriate.

When the manufacturing process involves long cryopreservation periods or cell cultivation periods, perform sterilization tests at constant intervals to confirm sterility has been maintained.

7. Changes in the manufacturing process

If the manufacturing method is altered at some point during development and test results obtained using products manufactured before the change in manufacturing method are to be used in the application for clinical trial or regulatory approval, demonstrate that the products manufactured before and after the change to the manufacturing process are comparable.

III. Quality Control of the Final Product

1. Introduction

The overall quality control strategy for products manufactured using human somatic stem cells include specifications (a set of acceptance criteria and analytical procedures) for the final products, quality control of raw materials for each different application to each individual patient, verification of the appropriateness of the manufacturing process, maintenance of consistency, and proper quality control of any intermediate products.

Specifications will differ for each final product depending upon the type and properties of the desired cells and tissues, manufacturing methods, intended clinical use, method of clinical application, stability, and test methods These differences shall be taken into consideration when setting the acceptance criteria and test procedures. In addition, specifications shall be set and justified from the perspective of achieving the purpose of quality control as a whole, by taking into consideration the mutually complementary relationships between 1) the verification of the suitability of the manufacturing process, 2) the method of maintaining consistency, and 3) quality control of the raw materials and intermediate products. The purpose of the assessment at the initiation of clinical trials is to confirm that the product in question is deemed to pose no significant quality/safety problems for using investigational clinical trials. Therefore, it may be possible to set provisional specifications with allowances for some variation on the basis of values measured in a few test specimens, as long as one can argue for a relationship between the results of clinical tests and the quality attributes after the clinical trial. However, testing for sterility and the presence of mycoplasma is essential. It should be noted that the quality control strategy, including specifications, should be enriched and developed as the clinical trial progresses.

2. Quality control of the final product

Refer to the general quality control parameters and tests shown below, set necessary and appropriate specifications for the final product, and justify the rationale for the specifications set.

Set appropriate specifications for individual products that do not make up a lot and for products that do make up a lot, because normally each individual lot is the unit subject to quality control.

(1) Cell number and cell viability
For cells that are an active ingredient in the final product, determine the cell number and viability in the final product or, if needed, in an appropriate intermediate product. At the beginning of the clinical trial, it is acceptable to set provisional acceptance criteria that are based on values measured in a small number of test samples.

(2) Tests of identity
Confirm that the intended target cells comprise the product by assessing important cell characteristics, such as morphological characteristics, biochemical markers, immunological markers, characteristic products, and other appropriate genotypes or phenotypes.

(3) Tests of purity
To confirm the purity of the cells in a final product, if necessary, set the test parameters, test methods, and acceptance criteria for evaluating and controlling non-target cells, such as undifferentiated cells, cells that exhibit abnormal growth, transformed cells, and contaminating cells, taking into consideration the origin of the target cells and tissues, the culture conditions, other parameters of the manufacturing process, quality control of intermediate products, etc. At the beginning of the clinical trial, it is acceptable to set provisional acceptance criteria that are based on values measured in a small number of test samples.

(4) Tests for cell-derived, undesirable, physiologically active substances
Specify appropriate tests for determining the permissible dose limits of any potential undesirable, physiologically active substances that are derived from the target cells whose presence in the product is presumed to affect the safety of the patient. At the beginning of the clinical trial, it is acceptable to set provisional acceptance criteria that are based on values measured in a small number of test samples.

(5) Tests for process-related impurities
For substances that may be present in the final product as contaminants, residues, or newly generated products or degradation products, etc.; that potentially originate from raw materials, non-cellular components, media ingredients (including feeder cells), chemical reagents, or any other process-related materials; and that may have deleterious effects on quality and safety (for example, albumin derived from fetal calf serum, antibiotics, etc.), it is necessary to 1) prove that the substance is not present in the final product by taking into consideration the results of process evaluations related to the elimination of the substance or the results of in-process substance control or 2) establish appropriate tests with which to control permissible levels of the substance in the final product. When selecting substances to be tested and setting their acceptance criteria, their appropriateness should be explained and justified. At beginning of the clinical trial, it is acceptable to set provisional acceptance criteria that are based on values measured in a small number of test samples.

(6) Sterility tests and tests for the presence of mycoplasma

Sterility should be ensured throughout the entire manufacturing process by evaluating test samples. The sterility (negative for common bacteria and fungi) of the final product should be demonstrated before its use in a patient. Appropriate tests confirming the absence of mycoplasma should also be performed. A validated nucleic acid amplification test can be used. If the results of sterility and other tests on the final product can only be obtained after the product is administered to the patient, methods for dealing with non-sterility detected after administration should be established beforehand. In such an instance, demonstrate by testing that the intermediate products are sterile and that sterility has been strictly controlled in all processes leading to the final product. If a product from the same facility and same process has already been used in patients, its sterility must be confirmed by testing all patients. If complete closure (hermetic seal) of a product that is part of a lot has been assured, tests on representative samples are sufficient. When each different application needs to be tested and if test results can only be obtained after administration to the patient, the decision to administer the product will be based on the most recent data. However, even in such an instance, the final product shall be tested.
It is desirable that every possible effort be made to avoid the use of antibiotics in cell culture systems. However, if antibiotics are used, adopt measures to ensure that they do not influence the sterility tests.

(7) Endotoxin test
Perform the endotoxin test, taking into consideration the impact of the contaminant in the samples. The acceptance criteria do not necessarily depend on the actual measured values. Set acceptance criteria by taking into consideration the safety ranges in the Japanese Pharmacopoeia and/or any other relevant compendia that are based on a single dose of the final product. Endotoxin testing can be established as an in-process control test. However, in such cases, specify the criteria, including the validation results, and justify their appropriateness.

(8) Virus tests
If the absence of HBV, HCV, HIV, and HTLV cannot be demonstrated at the patient level and these viruses could proliferate in the cells, use titer tests to detect viruses, and confirm that administration of the stem cell-based products does not adversely affect the patient. This does not apply if the absence of viruses has been demonstrated by testing the cell bank or intermediate products. If components of a biological origin are used in the manufacturing process, it may be necessary to tests the final product for viruses originating from those components. However, whenever possible, it is preferable to verify there is no contamination by testing at the original component stage or by process evaluation.

(9) Efficacy tests
In some instances, it will be necessary to consider efficacy testing that takes into account the cell type, intended clinical use, or distinctive characteristics of the cells. At the beginning of the clinical trial, it is acceptable to set provisional

acceptance criteria that are based on values measured in a small number of test samples.

(10) Potency tests
If a specific physiologically active substance secreted from cells or tissues contributes to the clinical efficacy or effect of a somatic stem cell-based product, establish test parameters and/or acceptance criteria for the substance in order to demonstrate the intended effect. Set acceptance criteria for potency, amount produced, etc. for phenotypic products produced by the desired cells or for an expression product secreted from the cells when a gene has been introduced. At the beginning of the clinical trial, it is acceptable to set provisional acceptance criteria that are based on values measured in a small number of test samples.

(11) Mechanical compatibility tests
For products that require a certain degree of dynamic strength, set acceptance criteria for mechanical compatibility and durability that take into account the site of application. At the beginning of the clinical trial, it is acceptable to set provisional acceptance criteria that are based on values measured in a small number of test samples.

Chapter III Stability of Autologous Human Somatic Stem Cell-based Products

Taking into full consideration the storage and distribution periods and the storage form, test the cell viability, potency, etc. of autologous human somatic stem cell-based products and/or critical intermediate products to establish storage methods and an expiration date. Justify their appropriateness. In particular, when product storage and use involves freezing and thawing, confirm that the freezing and thawing processes do not affect the stability or acceptance criteria of the product. Where necessary and possible, conduct stability studies on products whose manufacturing period or storage period exceeds normal periods in order to confirm, to the extent possible, the limits of stability. This does not apply if a product will be used immediately after its production.

If an autologous human somatic stem cell-based product will be transported, the relevant transportation vessels and transportation procedures (such as thermal management, etc.) shall be set and their appropriateness justified.

Chapter IV Preclinical Safety Testing of Autologous Human Somatic Stem Cell-based Products

To the extent that they are scientifically reasonable and technically possible, relevant animal tests and/or in vitro tests may be performed in order to identify safety concerns associated with an autologous human somatic stem cell-based product. For non-cellular constituents and process-related impurities, safety concerns should be addressed as much as possible by physicochemical analyses, not animal testing.

Testing human specimens is very valuable, and testing products of human origin in experimental animals does not always yield meaningful

results. Thus, there may be a scientific rationale for preparing products of animal origin and testing them in appropriate experimental animals, if such a test system is expected to generate useful results. In such a case, consider using an animal model that is suitable for the target disease. (For example, monkeys may be suitable for studies of nervous system diseases, and pigs and/or dogs may be suitable for studies of cardiovascular diseases.) However, because cells with characteristics identical to those of cells that constitute an autologous human somatic stem cell-based product cannot necessarily be obtained from non-human animal species, even if the preparation procedures are the same, and because a product of animal cell origin manufactured by identical processes will not necessarily be comparable to a human cell product, conduct a feasibility study before adopting, conducting, and evaluating such tests. When performing animal experiments using somatic stem cell-based products obtained from non-human animal species, explain how extrapolation to humans is appropriate. Depending on the case, consider test systems that employ cells, and clearly explain the appropriateness of the test system.

Presented below are items and points to consider and refer to when confirming the preclinical safety of a product. These are examples provided for illustration purposes; they are not intended to prescribe tests for which there is no rational basis. Taking into consideration that the cells are autologous-derived, the characteristics of the product, the intended clinical use, etc., conduct necessary and appropriate tests, and evaluate and discuss the results in a comprehensive manner.

1. For cells expanded beyond the limit set for routine cultivation (defined by a period of time, the population doubling level, or the passage level), demonstrate that transformations other than those intended have not occurred.
2. It may be necessary to quantify special physiologically active substances produced by the cells and tissues and discuss their effects when administered to patients. In some cases, significant amounts of active substances including cytokines and growth factors would be produced by the cells, potentially resulting in undesirable effects on the patients.
3. From the aspect of product safety, examine and discuss the potential effects of the product on a patient's normal cells and tissues and the consequences.
4. Depending on the type of product, investigate and discuss the possibility of ectopic tissue formation by the cells in the product and the potential safety consequences thereof when the product is administered to the patient.
5. Investigate and discuss the possibility of undesirable immunological reactions caused by the product and/or the expression product of a transgene and the consequences thereof.
6. Discuss in a comprehensive manner the possibility of tumor formation, including benign tumors and/or malignant transformations, taking into consideration the type and characteristics of the product, number of cells and route of administration,

mode of application (e.g.,cell sheet, cell suspension etc.), cell engraftment site, target diseases, appropriateness of the tests systems, etc. If necessary, conduct studies using a suitable animal model. If tumorigenicity or malignant transformation is a possibility, provide justification and rationale for the use of the product in question, taking into consideration the anticipated efficacy. (Note: For tumorigenicity studies, it is most important to assess accurately the tumorigenicity of the final product that will be used in patients. However, tumorigenicity may have to be evaluated using cells from the intermediate product if, for various reasons, such as insufficient cell number, the cells in the final product cannot be used. Furthermore, when conducting tumorigenicity tests in animal models, variables such as cell dispersion, cell adhesion to scaffolding, cell density, and administration site may not be the same as for the final product. The species, strain, and immunological state of the animal may also affect its sensitivity. The tumorigenicity of the final product should be evaluated in a comprehensive manner taking these factors into account.) The risks to the patient arising from tumorigenicity of the final product should be evaluated by weighing the risks of treatment against the benefits of treating the disease.

7. If an exogenous gene is introduced into certain cells during the manufacturing process, conduct tests in accordance with "Gene Therapy Pharmaceutical Guidelines," published as Notification No. 1062 by the Ministry of Health and Welfare on November 15, 1995. In particular, if viral vectors are used, conduct quantitative tests to determine if any replication-competent viruses are present, and justify the appropriateness of the test method employed. Describe the safety of transgene and its products on the basis of their characteristics. For cells, discuss the possibility of changes in cell growth and tumor formation, including benign tumors and malignant transformations. When using a vector that can insert into a chromosome, consider the necessity of evaluating abnormal proliferative characteristics and/or tumorigenicity and implementing long-term follow-up.

8. Consider conducting rationally designed general toxicology tests if the product, including an animal-derived product, is easy to obtain and if doing so will generate useful information regarding its clinical application When conducting general toxicology tests, refer to "Guidelines for Toxicology Studies on Pharmaceuticals," which is an appendix in the document entitled "Guidelines on Toxicology Studies Required for Regulatory Approval for the Manufacture or Import of Pharmaceuticals" (Drug Evaluation Notification 1:24, Ministry of Health and Welfare, issued September 11, 1988).

Chapter V Studies Supporting the Potency or Efficacy of Autologous Human Somatic Stem Cell-based Products

1. A well-designed study with experimental animals and/or cells should be performed to demonstrate, to a scientifically reasonable and

technically possible extent, the functional expression, the sustainability of effect, and/or the anticipated clinical efficacy (proof of concept) of an autologous human somatic stem cell-based product.
2. For transgenic cells, demonstrate the expression efficiency, sustainability of expression, and biological activity of the desired products from the transgene. Discuss the anticipated clinical efficacy (proof of concept) of the autologous human somatic stem cell-based product in question.
3. Where appropriate products derived from the processing of animal somatic stem cells and/or disease model animals are available, use them to study the potential therapeutic efficacy of the product.
4. At the beginning of the clinical trial, detailed experimental studies will not necessarily be required if scientific literature and/or other information supports the prediction that the potency or efficacy of the product in question will be markedly superior to that of a different therapeutic method.

Chapter VI Pharmacokinetics of Autologous Human Somatic Stem Cell-based Products

1. Pharmacokinetic studies of the internal behavior of transgene expression products or cells/tissues that constitute the final product, which may include assessment of their absorption and distribution in experimental animals, should be performed to an extent that is technically possible and scientifically reasonable. Thereby, it is expected to estimate the survival of cells/tissues administered to patients and the duration of their effect, and determine if the intended efficacy is sufficiently achieved. (Note: Testing methods may include histological studies, Alu-PCR, MRI, PET, and SPECT, and bioimaging.)
2. For autologous human somatic stem cell-based products, clarify, through animal studies, the rationale for the administration method. In particular, extrapolate from animal experiments the systemic distribution of cells after systemic administration and discuss the distribution from the point of view of clinical usefulness. (Note: Although it is unclear exactly where cells adhere with each administration route, local administration is presumed to be preferable to systemic administration. However, if the benefits to patients can be explained, it may be acceptable to use systemic administration. In any case, an administration method that minimizes the distribution of a somatic stem cell-based product to organs other than the target organ would be a rational choice. Even if cells do localize to a site other than the intended transplantation site, the administration method may be used if no adverse effects on patients result. Arrhythmia caused by osteogenesis of mesenchymal stem cells that ectopically localize to the heart is an example of an adverse effect that could result from ectopic differentiation.)
3. When the cells or tissues are directly applied or targeted to a specific site (tissue, etc.) where they are expected to act, clarify the localization and discuss the effect of the localization on the efficacy and safety of the product.

Chapter VII Referring to Clinical Trials

The main purpose of the present guideline is to outline points to consider for evaluating the quality and safety of autologous human somatic stem cell-based products, either at the time of application for marketing authorization or at the beginning of an investigational clinical trial. In the latter case, it is necessary to evaluate, while taking into consideration the clinical usefulness, whether there are any quality and/or safety problems that might impede the initiation of human clinical trials. Thus, quality and non-clinical safety evaluations for determining to initiate the investigational clinical trials of the product in question should refer to the points outlined below. Any presumed risk factors associated with product quality and safety should be eliminated, as much as possible, using up-to-date scientific and technological methods, and the scientific appropriateness should be clearly described. Any remaining risks should be weighed against the risks associated with not performing the trials in patients that suffer from diseases that are serious and life-threatening, that involve marked functional impairment or a marked decrease of quality of life (QOL) resulting from the loss of physical function or form, or for which existing therapies have limitations and do not provide cures. Furthermore, it is critical to entrust to the patient the right to make a decision after providing all of the information available, including all information on identified/unidentified risks and anticipated benefits.

1. Target disease
2. Target subjects and patients who should be excluded as participants.
3. Details of the therapy to be performed in the subjects, including the application of autologous human somatic stem cell-based products and drugs used concomitantly. (Note: If it is anticipated that drugs will be co-administered in order to maintain, enhance, and/or induce the function of administered or transplanted cells, verify the intended activity of the drugs either in vitro or in vivo.)
4. Appropriateness of conducting the clinical trials in light of existing therapeutic methods.
5. Plan for explaining the clinical trial to the patients, including the currently known risks and benefits of the product.

Clinical trials should have an appropriate study design and specified endpoints. They should be designed in light of the desired cells/tissues, target disease, and method of application.

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C.1.4 ヒト（同種）体性幹細胞加工医薬品等の品質及び安全性の確保に関する研究の経緯と視点及びガイドライン英文版

Study on Ensuring the Quality and Safety of Pharmaceuticals and Medical Devices Derived from the Processing of Allogenic Human Somatic Stem Cells

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Background (Chronology and Focus of the Research)

The details of the present study were described in a previous paper[1]. The present paper summarizes points that are closely related to those presented in the earlier paper.

Regenerative medicine using cell-based products derived from the processing of human cells and tissues is keenly anticipated in Japan because of difficulties with securing human organs and tissues in our country. With technology breakthroughs and research advances, people are increasingly hopeful that medical technology using novel cell-based products will develop into therapies.

In Japan, translational research into regenerative medicine is advancing rapidly. In particular, considerable work has been done to develop products that make us of human pluripotent cells, i.e., somatic stem cells such as mesenchymal stem cells, embryonic stem (ES) cells, and induced pluripotent stem (iPS) cells. Thus, there is an urgent need to prepare relevant guidelines for the evaluation of products expected in the near future. Identifying at an early stage of development the technical, medical, and ethical conditions necessary for the utilization of various types of stem cells at an early stage of development is vital for their rapid application in patients.

In fiscal year 2008, the Japanese Ministry of Health, Labor, and Welfare (MHLW) convened a panel of experts entitled "Study Group on Ensuring the Quality and Safety of Pharmaceuticals and Medical Devices Derived from the Processing of Human Stem Cells." The panel was established as an MHLW scientific research project and has been chaired by Dr. Takao Hayakawa since its conception.

The objective of the study group is to promote the sound development of products derived from human stem cells by investigating scientific and technological advances, ethics, regulatory rationales, and international trends regarding human stem cell-derived products and to establish and implement appropriate safety evaluation criteria.

As a result of analyses conducted up to 2009, in accordance with the Pharmaceutical Affairs Law, and with clinical application of products

derived from human somatic stem cells, iPS cells, ES cells, and other cells as the goal, the study group concluded that relevant guidelines should be tailored to specific cell sources and phenotypes (human autologous *vs*. human allogenic; somatic stem cells *vs*. iPS cells *vs*. ES cells *vs*. other cells) to facilitate efficient, effective, and rational research and development(R&D). Points to be considered include but are not limited to: technical details, the manufacturing process, characterization, quality control, and stability evaluation, and the data necessary to guarantee the safety and efficacy of the products.

With this perspective in mind and with a desire for consistency in scientific principles and concepts, two interim reports on draft guidelines for autologous human somatic stem cell-based products and autologous human iPS cell-based products were prepared in 2009 on the basis of MHLW Notification No. 0208003. Three other interim reports of draft guidelines for allogenic human somatic stem cell-based products, allogenic human iPS cell-based products, and human ES cell-based products were also prepared on the basis of MHLW Notification No. 0912006. These five sets of draft guidelines were thoroughly discussed from a variety of viewpoints. They were then widely circulated among interested parties as articles in a relevant scientific journal to allow readers to comment (Hayakawa T., et al.: Regenerative Medicine (Journal of the Japanese Society for Regenerative Medicine), 9, 116–180 (2010), in Japanese). Thereafter, these articles were updated and published as eight articles (Journal of the Japanese Society for Regenerative Medicine), 10, 86–152 (2011)) that served as the basis for the final draft guidelines. After extensive discussions with the study group and public consultation, the Pharmaceutical and Food Safety Bureau of MHLW issued five notifications on September 17, 2012, as described in the previous paper[1)].

In this paper, a continuation of the previous paper, we introduce the basic technological requirements for ensuring the quality and safety of pharmaceuticals and medical devices derived from the processing of allogenic human somatic stem cells. The final products derived from the processing of allogenic human somatic stem cells, which are multipotent and retain the ability to self-replicate, should exhibit cell characteristics different from those of the starting cells as a result of cell processing and may be applied and function at a site (cell environment) different from where the original cells localized. These concerns have been added to Notification No. 0912006, which serves as the basis.

Before interpreting and implementing the present guideline, the following should be considered. The ultimate goal is to provide patients with new therapies that utilize regenerative medicine. The role of the guideline is to present the scientific principles, concepts, ideas, and technical elements that will achieve the specified goal in the most efficient and effective manner possible. Because situations, circumstances, and products will vary, the guideline addresses points of concern in a comprehensive manner. Therefore, it is critical to identify the relevant testing parameters and evaluation methods by taking into consideration the characteristics of the cells in question, the specific clinical objective, the method of application, etc. Those that are applicable should be justified and implemented in an appropriate and flexible manner.

Several points should be kept in mind with regard to the development of medicinal products for regenerative

medicine and throughout the employment of this guideline. The desired products are expected to show a potential as a novel therapeutic method through relevant proof of concept (POC). Relevant data and/or information should establish that there are no critical concerns for product safety that might impede the use of the products in humans for the first time. Thorough observance of the Declaration of Helsinki, including proper informed consent and right of self-determination on the part of the patient, is indispensable.

It should be emphasized again that the primary goal of our endeavor is to offer suitable treatment options as fast as possible to patients suffering from severe diseases that are difficult to treat with conventional medicine. The present guideline should be useful for this purpose. Therefore, it is important to interpret and employ the guideline in a flexible and meaningful way. Stringent observance of the guideline without taking into account the patients and their specific situation, which is like putting the cart before the horse, should be avoided.

It is evident that progress in the application of regenerative medicine is desirable for maintaining and improving peoples' health. The development of innovative and revolutionary medicinal products and therapeutic techniques should benefit our country as well as the international community. Regenerative medicine is a great way to make a peaceful international contribution that will be a legacy to mankind. In this context, the role of government is to promote clinical research and industrialization; regulations and guidelines are adopted such that we advance towards this common goal in a scientific, rational, efficient, and effective manner. All those involved, like players with a common goal in the same arena, should continue to make great efforts to deliver to patients as fast as possible revolutionary, cell-based products and therapeutic techniques.

Guidelines on Ensuring the Quality and Safety of Pharmaceuticals and Medical Devices Derived from the Processing of Allogenic Human Somatic Stem Cells (September 7, 2012)

Introduction

1. The present guidelines outline basic technical elements for ensuring the quality and safety of pharmaceuticals and medical devices derived from the processing of allogenic human somatic stem cells. These products are hereafter referred to as allogenic human somatic stem cell-based products or merely as the "desired cell products."

There are many different types of allogenic human somatic stem cell-based products and methods of clinical application. In addition, scientific progress in this field is constantly advancing and experience and knowledge are constantly accumulating. Therefore, it is not always appropriate to consider the present guidelines all inclusive and definitive. Consequently, when testing and evaluating each individual product, it is necessary to take on a case-by-case basis, a flexible approach based on rationale that reflects the scientific and technological advances at that point in time.

2. The main purpose of evaluating the quality and safety of the desired cell products before conducting investigational clinical trials (e.g., at the time of "clinical trial consultation") is to determine whether there are any quality and/or safety problems that would obviously hinder initiating human clinical trials of the allogenic human somatic stem cell-based products in question, whether certain quality attributes (QA) of the product are understood sufficiently to establish a relationship between the clinical findings and the QA, and whether consistency of the QA can be ensured within a definite range. Simultaneously, it is important to eliminate as much as possible any presumed known risk factors associated with product quality and safety using up-to-date science and technology and to describe the scientific appropriateness of the results of such action. The remaining unidentified risks factors should be weighed against the risks associated with not performing the trials in patients suffer from diseases that are serious and life-threatening, that involve marked functional impairment or a marked decrease in quality of life (QOL) resulting from the loss of a certain degree ofphysical function or form, or for which existing therapies have limitations and do not provide cures. Furthermore, it is important to entrust to the patient the right to making a decision after providing all of the information available. When applying for investigational clinical trials, applicants can submit a provisional non-clinical data package, which is prepared reasonably by taking into account product aspects and patient aspects including a balance between risk of product *vs* risk of patient with/without treatment in question, ~~,~~ for determining to initiate investigational clinical trials, on the premise that the data package submitted at the time of marketing application/registration to ensure quality and safety will be enriched and developed in line with the guidelines as the clinical trial progresses.

Finally, applicants are encouraged to discuss with the Pharmaceuticals and Medical Devices Agency (PMDA) the type and extent of data that may be needed to initiate an individual clinical trial. Because of differences in product origin, target disease, target patients, application sites, application methods, and processing methods, there may be numerous variations between individual data packages that cannot be definitively clarified in the present guidelines.

3. The items, test methods, criteria, and any other technical requirements described in the present guidelines are intended to be considered, selected, applied, and evaluated to serve each intended purpose; they do not necessarily require the most stringent level of interpretation and practice. In accordance with the purpose of the present guidelines, applicants are encouraged to explain and justify how the background, selection, application, and the content and extent of evaluation are appropriate and scientifically rational.

Chapter I General Principles