using materials that are absorbed by the body, perform the necessary tests on any degradation products.

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. 02013001, Pharmaceutical and Food Safety Bureau, Japanese Ministry of Health, Labor, and Welfare, issued February 13, 2003), and describe the test results and justify the use of such raw materials. It is encouraged to use rationally knowledge and information obtained from the literature as well.

(ii) Interactions with target (desired?) cells

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

(b) Evaluate to the greatest 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 due to any interactions between the noncellular components and the cells in the final and intermediate products.

(iii) When using noncellular components with the objective of segregating the cells from the application site.

When using noncellular components with the objective of segregating the cells from the application site, confirm their efficacy and safety by referring to (a) through (e) below.

(a) When immunological segregation is the objective, describe its level

(b) Membrane permeability kinetics and pharmacological effect of target physiologically active substances derived from cells in the final product.

(c) Diffusion of nutritional components and excretory products

(d) Effects of noncellular components on the area near the application site.

(e) When a pharmacological effect of a target physiologically active substance derived from a desired cell is anticipated and the objective is segregation of the application site and the desired cells or undifferentiated cells, confirm that the cells do not leak out caused by the degradation, etc. of noncellular components.

(3) When cells undergo genetic modifications

When genes are introduced into cells, provide the details concerning the following items.

(i) For the target gene (specific gene

encoding a desired protein or RNA), information related to its structure, origin, method by which it was obtained, cloning methods, and for cell bank of the target gene, methods of preparation, control, and renewal, and so on.
(ii) Nature of the transgene
(iii) Structure, biological activity, and properties of the desired gene products
(iv) All raw materials, properties, and procedures (transgenic method, and origin, properties, and method of obtaining vector used in gene introduction) needed to produce the transgenic construct.
(v) Structure and characteristics of the transgene construct
(vi) Control and preparation methods for cell and virus banks in order to prepare vectors and transgenic constructs.

For the 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 of "Concerning Guidelines for Ensuring the Quality and Safety of Gene Therapy Pharmaceuticals" (hereinafter referred to as "Gene Therapy Pharmaceutical Guidelines"), published as Notification 1062 by the Ministry of Health and Welfare on November 15, 1995. Also, state clearly the appropriateness of the establishment in accordance with the appendix of the same notification.

Be aware that, based on the law (Law No. 97, 2003) concerning ensuring the biodiversity by regulating the use, etc. of genetic recombination organisms, etc., a separate application procedure will be required when living organisms including certain cells, as well "viruses", and "viroids" are genetically modified. The following cells are not regarded as living organism: "human cells, etc." or "cells that have the ability to differentiate, or differentiated cells but are not viable when alone under natural conditions".

Regardless of what is mentioned above, if a gene introduced into cells is used as a reagent in the manufacturing process and does not either chemically or functionally make up part of the final product, it is acceptable to just describe how the quality and safety of the gene conform to the intended use based on the most up-dated knowledge.

II. Manufacturing Process

When manufacturing pharmaceuticals and medical devices derived from processing of human (allogenic) somatic stem cell (i.e. human somatic stem cell-based products), describe in detail the manufacturing method and verify, to the greatest extent possible, the appropriateness of the method using the items listed below in order to maintain consistency of the quality of the product.
1. Lot composition and lot control
Indicate whether or not a lot is made up of final products and intermediate products. If a lot is composed of both final and intermediate products, establish standardized procedures concerning the make up and control of the lot.
2. Manufacturing method

Provide an outline of the manufacturing method from the time of receipt of the cells and tissues through to the isolation of somatic stem cells, and then to the final product, and describe the technical details of the process and necessary process control and product quality control.

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

(2) Inactivation and elimination of bacteria, fungi, viruses, and other microorganisms
For cells and tissues that serve as raw materials, carry out the inactivation and elimination of bacteria, fungi, viruses, and other microorganisms if needed and whenever possible, to such an extent that the procedures do not have any effect on the cell viability, phenotype, genetic traits, specific functions, or other characteristics and quality of the cells and tissues serving as raw materials. State the appropriateness of measures, procedures and evaluation methods taken, if any.

(3) Tissue disintegration, cell separation, isolation of specific cells, etc.
Describe the methods for disintegration of tissue, separation of cells, the isolation of somatic cells, as well as the methods for washing, etc. of these cells and tissues, that is performed in the early stages of manufacture of the somatic stem cell-based products from collected cells and tissues. When isolating somatic stem cells, establish identification methods for the cells.

(4) Preparation of cells that make up principal component of the final product as an active ingredient
Describe the methods used to collect the human cells and tissues, to isolate the somatic stem cells, and finally to obtain the cells that serve as the active ingredient of the final product. The methods to be described include induction of differentiation, isolation, and culturing of the desired cells, and the media, culture conditions, culture period, yield, and so on used at each step. Describe to the extent possible the appropriateness of each method.

(5) Establishment and use of cell line
Establish cell line after having determined to the extent possible the genetic background of the donor. Describe the method of establishment, and its appropriateness to the extent possible.
In order to maintain the stability and consistency of the quality of established cell line, identify important critical quality attributes of the cells (e.g., cell purity, morphological features, phenotypic

specific markers, karyotype, cell growth properties, pluripotency, etc.) and set acceptance criteria for them. Also demonstrate the potent number of passages within which cells can be proliferated with keeping their stability.
Discuss the possibility of tumorigenicity and malignant transformation, using an appropriate animal model, of established cell line.

(6) Establishment of cell banks
When a cell bank is established at any stage during the process of manufacturing human somatic stem cell-based products, describe the details of the rationale for preparing the cell banks, the methods used to prepare the cell banks, characterization of the cell banks, the storage, maintenance, and control methods, renewal methods, as well as any other processes and tests performed, and justify the appropriateness of each. Refer to "Derivation and Characterisation of Cell Substrates Used for Production of Biotechnological/Biological Products" (Pharmaceutical Notification Number 873, Ministry of Health, Labor, and Welfare, July 14, 2000) and other documents.

(7) 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 human somatic stem cell-based products. Therefore, clearly describe preventative measures in the process control.

3. Characterization of cells that make up principal component of a final product as an active ingredient
Analyze various attributes of the cells such as cell purity to control contamination by non-target cells, the cell viability, morphological characteristics, cell growth characteristics, biochemical markers, immunological markers, distinctive substances produced from cells, karyotype, and other appropriate genotypic and phenotypic markers of cells that make up principal component of the final product. Also characterize with respect to biological functions, where necessary. Furthermore, in order to evaluate the appropriateness of the culture period and stability of the cells, use appropriate cell characteristic markers to prove there have been no unintended changes in cells cultured for duration beyond the proposed culture period. When performing these studies, it is acceptable to study and verify beforehand using test samples obtained from donors who are not patients. Based on these test results, it is necessary to 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 perform the study fully since there are quantitative limits to samples as well as technological limits. Then, it is acceptable to just perform the study to the extent possible. When cell processing like growth within the body is anticipated after clinical application, clearly demonstrate the functions expected using the passage number or number

of cell divisions based on specified criteria.

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

5. Storage and transport of final product
If intermediate or final product needs to be stored and transported, 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 manufacturing procedure
When the manufacturing human somatic stem cell-based products, assess beforehand whether or not during the manufacturing process and for each individual product there has been any significant differences between each production (each lot) with respect to the number of cells, cell viability, and cell characteristics (such as relevant markers of phenotype, appropriate markers of genotype, functional characteristics, and the percentage content of desired cells) from the point of view of application methods and intended use of the product. It is acceptable to use test samples obtained from donors who are not patients in place of the real products that will be prepared for clinical trial. Evaluation using intermediate products may provide a good reflection of the appropriateness of cells and tissues used as raw materials and the validity of the manufacturing process up until the point of intermediate products, as well as also being an appropriate guidepost leading up to the final product. Therefore, it may be reasonable to adopt such approach, where necessary and appropriate.
When the cryopreservation period or time of cell cultivation last to a long term during the manufacturing process, perform sterilization tests and so on at constant intervals to confirm that the sterility has been ensured.

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

III. Quality Control of Final Product

1. Introduction
The overall quality control strategy of products manufactured using human somatic stem cells include specification of final products, quality control of raw materials for each different application to each individual patient, verification of the appropriateness of the manufacturing process and maintenance of consistency thereof, as well as the proper quality control of intermediate products.

Since specifications for the final product are to be different depending upon the type and properties of the desired cells and tissues, manufacturing methods, intended clinical use and method of application for each product, stability, and test methods that can be available, these differences that depend on the cell or tissue being handled shall be taken into sufficient consideration when setting acceptance criteria and test procedures. Also, 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 the verification of the suitability of the manufacturing process and the method of maintaining consistency and quality control of the raw materials and intermediate products. The purpose of the assessment for initiating of clinical trials is to confirm that product can be deemed to have 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 the values measured on a few test specimens, as long as one can argue the relationships between the results of clinical tests and such quality attributes after clinical trials. However, testing for sterility and presence of mycoplasma is essential. It should be noted that quality control strategy including specifications shall be enriched and developed along with the progress of clinical trials.

2. Quality control of the final product
Refer to the general quality control parameters and tests shown below and set necessary and appropriate specifications for the final product, and justify the rationale for the specifications set.
Set appropriate acceptance criteria and test procedures for the individual products that do not make up a lot and for the lot consisting of the products that do make up a lot since normally each individual lot is the unit subjected to quality control.

(1) Cell number and cell viability
The number and viability of cells as being active ingredient in the final product or if needed, in an appropriate intermediate product in the manufacturing process should be determined. At beginning of the clinical trial, it is acceptable to set provisional acceptance criteria based on actual measured values from a small number of test samples.

(2) Tests of Identity
Confirm that the cells are the intended target cells by means of important cell characteristic marker selected from among the morphological characteristics, biochemical indicators, immunological markers, characteristic products, and other appropriate genotypes or phenotypes of the intended target cells and tissues.

(3) Tests of Purity
If necessary, set the test parameters, test methods, and acceptance criteria for evaluating and controlling the purity of cells with respect to non-target cells, such as undifferentiated cells, cells exhibiting abnormal growth, transformed cells, and the presence of any

contaminating cells, taking into consideration the origin of the target cells and tissues, the culture conditions and other parameters of the manufacturing process, quality control of intermediate products, and so on. At beginning of the clinical trial, it is acceptable to set provisional acceptance criteria based on actual measured values from a small number of test samples.

(4) Tests for cell-derived undesirable physiologically active substances

Specify appropriate permissible dose limiting tests for any potential undesirable physiologically active substances that are derived from target cells and their significant presence in the product is presumed clearly to impact on the safety of the patient. At beginning of the clinical trial, it is acceptable to set provisional acceptance criteria based on actual measured values from 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 as newly generated products or degradation products, etc., potentially originating from raw materials, non-cellular components, media ingredients (including feeder cells), chemical reagents, or any other process-related materials, and that may have deleterious effect on the quality and safety (for example, albumin derived from fetal calf serum, antibiotics, etc.), it is necessary to either prove that the substance is not present in the final product by taking into consideration the results of process evaluation related to elimination of the substance or the results of in-process control of the substance, or alternatively establish appropriate tests with which to control permissible levels for 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 based on actual measured values from a small number of test samples.

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

The sterility of the final product should be sufficiently assessed to ensure sterility throughout the entire manufacturing process using test samples. The sterility (negative for common bacteria and fungi) of the final product should be demonstrated in tests before use in a patient. Appropriate tests confirming the absence of mycoplasma should also be carried out. A validated nucleic acid amplification method can be used. If the results of the sterility and other tests on the final product can only be obtained after administration to the patient, the methods for dealing with non-sterility after administration should be established beforehand. In such an instance, demonstrate by testing that the intermediate products are sterile, and the sterility should be strictly controlled in all processes up until the final product. If a product from the same facility and same process has already been used in patients, its sterility had to be confirmed by testing in all patients. If

complete closure (hermetically sealed) of the product comprising a lot has been assured, tests using only representative samples are sufficient. When tests need to be conducted for each different application and if the results of sterility and other tests can only be obtained after administration to the patient, whether or not application should be done or not will be determined based on the most recent data. However, even in such an instance, sterility tests and other tests on the final product shall be conducted.

While it is desirable that every possible effort be made so that antibiotics are not used in cell culture systems, if they are used, adopt measures to ensure that the antibiotics do not influence the sterility tests.

(7) Endotoxin test

Carry out the endotoxin test, taking into consideration the impact of the contaminant in the samples. The acceptance criteria do not necessary depend on the actual measured values. It is recommended to set acceptance criteria taking into consideration the safety ranges given 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 criteria, including validation results, and justify their appropriateness.

(8) Virus tests

Conduct tests for titer of possible viruses in the intermediate and the final product, when using cells which are not banked, and are from donors not provided in the window period of infection, and in which HBV, HCV, HIV or HTLV can propagate. If materials of a biological origin are used in the manufacturing process, it may be necessary to consider conducting tests on the final product for viruses originating from those components. However, whenever possible, it is preferable to verify there is no contamination by testing or process evaluation at the stage of the original component.

(9) Efficacy tests

In some instances, it will be necessary to consider efficacy testing that takes into consideration cell type, intended clinical use, or distinctive characteristics of the cells. At beginning of the clinical trial, it is acceptable to set provisional acceptance criteria based on actual measured values from a small number of test samples.

(10) Potency tests

If the secretion of a specific physiologically-active substance from the cells or tissues accounts for the efficacy or the essential effect of a somatic stem cell-based product, establish test parameters and/or acceptance criteria related to the substance in order to demonstrate the intended effect. Set acceptance criteria for potency, amount produced, and so on for phenotype products or for a desired product secreted from cells when a gene has been introduced. At beginning of the clinical trial, it is acceptable to set provisional acceptance criteria based on actual measured values from a small number of test samples.

(11) Mechanical compatibility tests

For products that require a certain degree of dynamic strength, set acceptance criteria to confirm mechanical compatibility and durability that take into account the site of application. At beginning of the clinical trial, it is acceptable to set provisional acceptance criteria based on actual measured values from a small number of test samples.

Chapter III Stability of Human Somatic Stem Cell-based Products

Taking into full consideration the storage and distribution periods and the storage form, perform suitable stability testing on human somatic stem cell-based products and/or critical intermediate products based on the cell viability, potency, etc. to establish storage methods and expiration date, and justify their appropriateness. In particular, when freezing and thawing are involved in the storage and use of the products, confirm that the freezing and thawing processes do not have any effects on the stability or criteria of the product. Where necessary and possible, it is recommended to conduct stability studies on the 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 being produced.
If a 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 justified their appropriateness.

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

Relevant animal tests and/or in vitro tests may be performed in order to elucidate safety concerns on a human somatic stem cell-based product to a scientifically reasonable and technically possible. The type, characteristics and intended clinical use of the individual final product should be critical elements when considering the scope and protocol of the non-clinical safety study and demonstrating the propriety thereof. Non-cellular constituents and process-related impurities should be evaluated as much as possible by physicochemical analyses but not tests using animals.

Testing specimens derived from humans are very valuable, and it is not always true that meaningful results can be obtained by testing of products of human origin with experimental animals. Thus, where more useful information is expected to be obtained by preparing product models of animal origin and conducting tests with appropriate experimental animals, there may be a scientific rationale for using such a type of test system. In such a case, consider conducting tests using suitable animal models for each target diseases. (Note: For example, monkeys may be suitable for nervous system diseases, while pigs and/or dogs may be suitable for cardiovascular diseases). However, since cell groups that possess identical characteristics to cells that constitute a human somatic stem cell-based product will not necessarily be obtained from

non-human animal species even though the preparation procedures are same as human, and since an animal cell origin product manufactured using identical culture conditions and so on will not necessarily be comparable to a human cell product, careful study beforehand are needed when adopting, conducting, and evaluating such tests. When conducting animal experiments using human somatic stem cell-based products obtained from non-human animal species, explain the feasibility of extrapolation. Depending on the case, consider test systems that employ cells, and clearly explain the appropriateness of the test system when conducting tests using this kind of approach.

Presented below are examples of items and points to consider that should be referred to, if necessary, when confirming the preclinical safety of a product. These are merely examples for illustration purposes and are not suggesting tests with no rational basis be carried out. Conduct necessary and appropriate tests, taking into account the characteristics of the product, intended clinical ~~application?~~ use, and so on, and evaluate and discuss the results in a comprehensive manner.

1. For cells expanded beyond the defined limit for cultivation (by a period of time, population doubling level of the cells, or passage level of the cells) for routine production, clearly demonstrate that transformations other than the target transformation have not occurred.
2. It may be necessary to conduct quantitative assays of some special physiologically-active substances produced by the cells and tissues and discuss their effects when given to patients. In some cases, significant amounts of active substances including cytokines and growth factors would be produced by the cells and this may result in undesirable effects on the patients.
3. Examine and discuss the potential effects and consequence from safety aspect of the product on the normal cells and tissues of a patient.
4. Depending on the type of product, investigate and discuss the possibility of formation of ectopic tissue by the cells in the product and potential safety consequence thereof when the product is given to the patient.
5. Investigate and discuss the possibility and safety of undesirable immunological reactions due to the product and/or expression product of a transgene, and safety consequence thereof.
6. Discuss in a comprehensive manner the possibility of tumor formation including benign tumors, and/or malignant transformation in terms of the type and characteristics of the product, route of administration, target diseases, appropriateness of the tests systems, and so on. If necessary, conduct studies using a suitable animal model. If tumorigenicity or malignant transformation is a possibility, justify the appropriateness of its use and the rationale, taking into consideration the relationship

with the anticipated efficacy (Note: The most important aspect of a tumorigenicity study is to accurately assess the tumorigenicity of a final product that will be used in patients. However, it is conceivable that the tumorigenicity will need to be evaluated using cells from the intermediate product because that cells comprising the final product cannot be used for various reasons, such as the inability to obtain a sufficient number of cells. Furthermore, various conditions such as cell dispersion and cell adhesion to the scaffolding, cell density, and administration site in tumorigenicity tests using animal models are not necessarily the same as for the final product. There are also differences in sensitivity depending on the species, strain, and immunological state of the animal. The tumorigenicity of the final product should be evaluated taking into consideration these circumstances in a comprehensive manner. The risks to the patient arising from tumorigenicity of the final product should be rationally evaluated based on the balance between any risks and the benefits to the patient by treating the disease).

7. If an exogenous gene is introduced into certain cells in the manufacturing process, conduct tests in accordance with "Gene Therapy Pharmaceutical Guidelines", published as Notification 1062 by the Ministry of Health and Welfare on November 15, 1995.
   In particular, if virus vectors are used, test quantitatively to determine the potential presence of any propagating viruses such as replication-competent retrovirus or replication-competent adenovirus and justify the appropriateness of the test method employed. Describe the safety of the transgene and its products based on their characteristics. For cells, discuss the possibility of changes in cell growth, tumor formation including benign tumor and malignant transformation. Whenever the vector which may be inserted in a chromosome is used, consider the necessity of evaluating possible occurrences of abnormal proliferative characteristic and/or tumorigenicity due to insertion mutation in the cells as well as of implementing long-term follow-up for clinical applications.
8. Consider conducting rationally designed general toxicology tests if it is easy to obtain the product, including an animal-derived model product, and if useful information on its clinical application is obtainable.
   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, September 11, 1988).

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

1. A well designed study with experimental animals and/or cells should be performed in order to demonstrate the functional expression, sustainability of effect, and/or the anticipated clinical efficacy (Proof-of-Concept) of a human somatic stem cell-based product to a scientifically reasonable and technically possible extent.
2. For transgenic cells, demonstrate the expression efficiency, sustainability of expression and biological activity of desired products from the transgene and discuss about the feasibility of anticipated clinical efficacy (Proof-of-Concept) of the human somatic stem cell-based product in question
3. Where appropriate models of products derived from 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 beginning of the clinical trial, detailed experimental studies will not necessarily be required if it can be justified by means of scientific literatures and/or other well-known information available that the potency or efficacy of therapy using the product in question is expected to be markedly superior compared to that using a different therapeutic method.

Chapter VI Pharmacokinetics of Human Somatic Stem Cell-based Products

1. Studies on pharmacokinetics relating to internal behavior of cells/tissues that constitute the final products or expression products of transgenes, which may include absorption and distribution in experimental animals should be performed to a technically possible and scientifically reasonable extent. Thereby, it is expected to presume the survival duration and duration of effect of cells/tissues in products that have been applied to patients and clarify if the intended efficacy is achieved to a sufficient extent. (Note: Testing methods may include histological studies, Alu-PCR, MRI, PET, SPECT, and bioimaging).
2. Clarify, through animal studies, the rationale for the administration method for the human somatic stem cell-based products. 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 the cells adhere for each administration route, it is assumed that local administration may be preferable to systemic administration. However, even with systemic

administration, if the benefits to patients undergoing administration can be explained in a rational manner, it may be acceptable to use systemic administration. For example, an administration method that minimizes distribution of somatic stem cell-based product to organs other than target organ would be the rational. Even if the cells do locate to a different site, it might be used as an administration method if there are no adverse effects on patients. A disadvantage due to ectopic differentiation may be, for example, arrhythmia caused by osteogenesis of some kinds of cells which ectopically locate to the heart).

3. When the cells or tissues are directly applied or alternatively targeted to a specified site (tissue, etc.) where they can be expected to exert their actions, 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 address points to consider for evaluating the quality and safety of human (allogenic) somatic stem cell-based products at the time of application for marketing authorization as well as at beginning of investigational clinical trial. In the latter case, it is necessary to evaluate, taking into consideration the clinical usefulness, if there is any quality or safety problems that might pose an obstacle to initiating human clinical trials (First-in-Man). This leads to the necessity of the evaluation by referring to the points outlined below for intended clinical trials in question. At that time, first any presumed known risk factors associated with the product quality and safety should be eliminated as much as possible using up-to-date science and technology, and the scientific appropriateness should be clearly described. The remaining unidentified risks should be weighed against the risks associated with not performing the trials in patients suffering from diseases that are serious and life-threatening, involve marked functional impairment, or a marked loss of quality of life (QOL) due to the loss of a certain degree of physical function or form, and for diseases in which existing therapies have limitations and do not provide cures. Furthermore, it is also critical to entrust the right to make a decision to the patient after making all of this information available, including all identified/unidentified risks and anticipated benefits to the patient.

1. Target disease
2. Point of view with respect to the target subjects and patients who should be excluded as subjects
3. Details of the therapy to be performed in the subjects, including the application of human somatic stem cell-based products and drugs used concomitantly (Note: If it is believed drugs to maintain, enhance, and/or induce the function of administered or transplanted cells will be coadministered, verify the activity of the drugs either in

vitro or in vivo).

4. Appropriateness of conducting the clinical trials in light of comparison with existing therapeutic methods.
5. Plan for explaining the clinical trial to the patients, including the risks and benefits of the product from currently available information.

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

C. 12. 5 ヒト（自己）iPS（様）細胞加工医薬品等の品質及び安全性の確保についての英文版

Guidelines on Ensuring the Safety and Quality of Pharmaceuticals and Medical Devices Derived from Processing of Human (Autologous) Induced Pluripotent Stem (-Like) Cells

(September 7, 2012)

Introduction

1. The present guidelines outline the basic technical elements to ensure the quality and safety of pharmaceuticals and medical devices derived from processing of human (autologous) induced pluripotent stem (iPS) cells or human (autologous) iPS-like cells. These products are hereinafter referred to as human (autologous) iPS(-like)cell-based products or merely as "desired cell products".
Human (autologous) iPS(-like) cell-based products are obtained either by artificially inducing the differentiation of various types of iPS(-like) cells generated artificially from human somatic cells and are then used directly or after further processing. There are many different types of manufacturing methods, intermediates, types and characteristics of desired cell products, and methods of clinical applications. On the other hand, the scientific progress and accumulation of experience and knowledge in this field are constantly advancing. Therefore, it is not always appropriate to consider that the contents of the present guidelines are all inclusive and all definitive. Consequently,

when testing and evaluating each individual product, it is necessary to take flexible approaches, on a case-by-case basis, based on rationale that reflects scientific and technological advances at that specific point in time.

2. The main purpose of evaluation of quality and safety of the desired cell products prior to conducting investigational clinical trials (e.g., at the time of "clinical trial consultation") is to confirm whether or not there are any quality and/or safety problems that would obviously be a hindrance to initiating human clinical trials on iPS(-like) cell-based products in question, whether certain quality attributes (QA) of the product are grasped enough to check the relationship between clinical findings and the QA, and whether consistency of the QA are ensured within a definite range. At that time, it is also important at first to eliminate any presumed known risk factors associated with the product quality and safety as much as possible using up-to-date science and technology and to describe the scientific appropriateness of the results of such action. The remaining unidentified risk factors should be weighed against the risks associated with not performing the trials in patients suffering from diseases that are serious and life-threatening, involve marked functional impairment, or a marked loss of quality of life (QOL) due to the loss of a certain degree of physical function or form, and for diseases in which existing therapies have limitations and do not provide cures. Furthermore, it is also important to entrust the right to making a decision to the patient after making all of this information available. In applying for investigational clinical trials, applicants can submit reasonably-prepared provisional data package, which fulfill conditions to meet the purpose as aforementioned, on the premise that data package for ensuring quality and safety at the time of marketing application/registration are enriched and developed along with the progress of clinical trials in line with the guidelines.
Finally, applicant are encouraged to discuss with the Pharmaceuticals and Medical Devices Agency (PMDA) with respect to the type and extent of data that may need for initiating individual clinical trials. There may be numerous variations in individual data package that could not be definitively clarified in the present guidelines due to differences such as in the origin of the product, target disease, target patients, sites of application, methods of application, and methods of processing.

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

Chapter I. General Principles

I. Objective

The present guidelines outline the basic technical elements to ensure the quality and safety of pharmaceuticals and medical devices derived from processing of autologous induced pluripotent stem (iPS) cells or autologous iPS-like cells (excluding allogenic iPS cells and allogenic iPS-like cells). These products are hereinafter referred to as human (autologous) iPS(-like) cell-based cell products or merely as "desired cell products".

II. Definitions

The definitions of the technical terms used in this guideline are as follows:

1. "Human induced pluripotent stem cells (iPS cells)": Cells that are generated from somatic cells through artificial reprogramming by introducing genes or proteins, or by chemical or drug treatment, or cells that are obtained from such cells through cell division, and that possess the ability to differentiate into endoderm, mesoderm, and ectoderm, and furthermore, maintain the ability to self-renew or the similar ability.

2. "Human induced pluripotent stem-like cells (iPS-like cells)": Cells that are generated from somatic cells through artificial dedifferentiation by introducing genes or proteins, or by chemical or drug treatment, or cells that are obtained from such cells through cell division, and that at least possess the ability to differentiate into some type of endoderm, mesoderm, or ectoderm, and furthermore, maintain the ability to self-renew or similar ability.

3. "Processing of cells and tissues": Any processing of a cell or tissue, such as pharmaceutical or chemical treatment, altering a biological characteristic, combining with a noncellular component, manipulation by genetic engineering, and so on, with the objective of propagation and/or differentiation of a cell or tissue, cell activation, and production of a cell line, with the aim of treating a patient, or repairing or regenerating tissue.
The isolation of tissue, disintegration of tissue, separation of cells, isolation of a specific cell, treatment with antibiotics, sterilization by washing or gamma irradiation, freezing, thawing, and such similar procedures regarded as minimal manipulations are not considered to be processing.

4. "Manufacture": Actions undertaken up until the final product (a human (autologous) iPS(-like) cell-based product) is released to market. This includes in addition to processing of cells and tissues, minimal manipulations such as the separation of tissue, disintegration of tissue, separation of cells, isolation of a specific cell, treatment with antibiotics, sterilization by washing or gamma irradiation, freezing, thawing, and other procedures that are performed without any change in the original properties of the cells or tissues.

5. "Phenotype": A morphological or physiological characteristic that is expressed by a certain gene under constant environmental conditions.

6. "Donor": persons who donate their own somatic cells that serve as the

raw material of a human (autologous) iPS(-like) cell-based product. As for a human (autologous) iPS(-like) cell-based product, a patient is difinitely a donor. (Note: A patient is identified as a donor for actual treatment. It is also presumed that cells/tissues obtained from a donor besides a patient are used for a purpose of test production at the development or other stages.)

7. "Transgenic construct": A construct which contains a vector for introducing a target gene (specific gene encoding a desired protein or RNA) into a target cell, the target gene, and the coding sequences of the elements essential for the expression of the target gene.
8. "Protein transductant": A construct which contains a target protein and elements like reagents necessary for introducing the target protein into a target cell.

Chapter II Manufacturing Methods

Describe all important and relevant information concerning the manufacturing method, taking into account the items listed below. This information will contribute to ensuring the quality, safety, and efficacy of the final product, and is important for guaranteeing the consistency of the quality from a manufacturing perspective. It should be noted that assurance of the quality and safety, and their consistency, is achieved by mutual complementary measures throughout the manufacturing method as a whole, and it is most important that the measures are rational and serve the intended purpose. It may be acceptable to omit a portion of the items listed below, after providing the appropriate scientific basis, with respect to the quality tests or controls of the final product or intermediates, or control of the manufacturing process, if the quality and safety, and their constancy, can be assured.

I. Raw Materials and Materials Used in Manufacturing

1. Human somatic cells that serve as raw materials

(1) Characteristics of biological structure and function, and justification of their selection
Explain and justify the reasons for selecting the somatic cells used as raw materials based on the characteristics of their biological structure and function, such as for example, morphological characteristics, growth characteristics, biochemical indicators, immunological indicators, specific substances produced, and other suitably chosen and appropriate genotype or phenotype indicators. It is acceptable to use studies that used test samples prior to initiation of clinical trials.
This should lead to the identification of the main cell characteristic indicators that are to be employed when applying the cells to the patient. It is recognized that such study can only be performed to a reasonably possible extent since there are quantitative limits to samples as well as technological limits.

(2) Considerations with respect to the

donor

From the perspective of ensuring the safety of the patient as well as personnel involved in manufacturing the product or health care workers who treat a patient, establish test parameters related to any type of possible infections that may occur via the somatic cells collected, and justify the appropriateness of the parameters. Particular consideration shall be given to hepatitis B virus (HBV), hepatitis C virus (HCV), human immunodeficiency virus (HIV), and human T-lymphotropic virus (HTLV). Establish eligibility criteria that take into account the genetic characteristics, history of the patient, the condition of their health, and others and justify their appropriateness as donors. If donor genome or gene analysis is undertaken, they shall be done in accordance with "Ethical Guidelines for Human Genome and Gene Analysis Research" published jointly on December 28, 2004 by the Japanese Ministry of Education, Culture, Sports, Science, and Technology, Ministry of Health, Labor and Welfare, and Ministry of Economy, Trade and Industry.

(3) Records related to the donor
All records related to the donor shall be complete and kept so that any information necessary with respect to ensuring the safety of somatic cells used as raw materials can be verified. Concrete measures shall be described. For patients and donors of test samples, it is enough to only prepare and keep each specific information corresponding to the intended use of individual cells.

(4) Collection, storage, and transport of cells and tissues

(i) Eligibility of personnel and medical institutions collecting samples

Describe the technical requirements for personnel and medical institutions that collect the cells and tissues.

(ii) Suitability of sampling site and sampling method

Describe the rationale for selecting the cell and tissue sampling sites as well as the sampling method, and clearly state how these sites selected are both scientifically and ethically appropriate. For the cell and tissue sampling methods, indicate the suitability of the equipment and drugs used and the measures adopted to prevent microbial contamination, erroneous sampling (mix-ups), and cross contamination.

(iii) Informed consent from donors
Describe the details of the informed consent of the donor of the cells or tissue.

(iv) Protection of donor privacy
Indicate the measures adopted to ensure protection of the privacy of the donor.

(v) Tests to ensure donor safety
If tests such as those to confirm the state of the sampling site need to be performed in order to ensure the safety of the donor at the time of cell or tissue sampling, describe the details of the tests, as well as any interventions undertaken for test results that indicated a problem existed.

(vi) Storage method and measures to prevent erroneous sampling (mix-ups)
If the somatic cells collected need to be stored for a definite period of time, set the storage conditions and storage period, and justify the appropriateness (validity) for their setting. Describe in detail the measures and procedures to be taken to prevent erroneous sampling (mix-ups).

(vii) Transportation methods
If cells and/or tissues or iPS(-like) cells collected need to be transported, set the containers used for transport and the transportation procedure (including temperature control, etc.) and justify their appropriateness.

(viii) Preparation of records and keeping procedures
Written records for (i) through (vii) above shall be prepared and proper keeping procedures for the records shall be described in detail.

2. Raw materials other than target cells and tissues as well as materials used in manufacturing

Describe raw materials other than target cells and tissues as well as other materials used in the manufacturing process, indicate their appropriateness for their intended use, and if necessary establish their specifications (set of acceptance criteria and analytical procedures). Proper quality control for these materials should be carried out.
When so called 'Biological Products' or 'Specific Biological Products' (refer to Article 2.9 and 2.10 of Pharmaceutical Affairs Law) are used as raw materials, the amounts used should be kept to the minimum amount required and should strictly obey the relevant laws and notifications, such as "Standards for Biological Raw Materials" (Notification Number 210, Japanese Ministry of Health, Labor, and Welfare, 2003). It is particularly important to sufficiently evaluate information related to the inactivation and elimination of viruses, as well as to indicate measures for ensuring retrospective and other studies.
The technical requirements described in this paragraph should be taken into consideration when the process of reprogramming or dedifferentiation from the raw materials into iPS(-like) cells, and of directed differentiation from iPS(-like) cells into the final products in question include any relevant elements/concerns be applied.

(1) When culturing cells
(i) Indicate the appropriateness of all the components of any media, additives (serum, growth factors, antibiotics, etc.) and reagents, etc. used in the treatment of cells, and set specification 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
(d) The ingredients and water used in media should be of high quality and high biological purity, and whose quality is controlled at standards equivalent to those for pharmaceuticals and pharmaceutical raw materials.

(e) Provide information on not only the main ingredients used in media, but all components, 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 one raw material.
(f) Conduct sterile 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 believed to be controlled in 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. For products that may be used repeatedly in particular, investigate as much as possible ways to avoid using these serum components. If the use of serum or other such material is unavoidable, give consideration to the following points, and investigate ways to prevent the contamination and spread of bacteria, fungi, viruses, and abnormal prions from the serum and other products, as well as treatment methods for their elimination, to the greatest extent possible, from the final product.
(f) Clarify the origin of the serum or other component.
(g) 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).
(h) Only use these sera after having confirmed 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.
(i) Conduct appropriate inactivation and elimination procedures for 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.
(j) Preserve and store a portion of the serum used in order to be able to monitor for viral infections in cultured cells, monitor for outbreaks of viral diseases at the patient, and measure antigen production in response to a component of the heterologous serum used.

(iv) When using feeder cells, conduct quality evaluation while referring to "Derivation and Characterisation of Cell Substrates Used for Production of Biotechnological/Biological Products" (Pharmaceutical Notification Number 873, Ministry of Health, Labor, and Welfare, July 14, 2000), "Guidelines on Public Health Infection Issues Accompanying Xenotransplantations" (Notification 0709001, Research and Development