information along with any stability studies initiated (see section V.A.1 below). You should document the time and conditions of storage between cell collection and final harvest. You should record whether there are adequate procedures in place to ensure the stability of the bulk harvest while in storage.

2. Final Harvest

You should document whether the final cell harvest is centrifuged prior to final formulation, and if so, you should describe the wash conditions and media used. You should document whether the cells are cryopreserved after formulation or formulated immediately and given to the patient. If the final harvest is stored, you should describe the storage conditions and the length of storage.

3. Final Formulation

You should document the formulation of the final product in the review. You should record whether any excipients such as growth factors or human serum albumin are included in the final formulation and state their source (see section II.A.2 above). You should document the vendor and final concentration of these proteins. You also should record the cell density or concentration used in the final product. If the final product is delivered to the clinical site frozen, you should include in the review a description of how the product will be shipped and data to show that the product can be thawed with consistent results.

4. Product Manufacturing Concerns That Need to Be Addressed

You should summarize any areas of concern identified during the review of the product manufacturing procedures. You should discuss these concerns with the sponsor and/or communicate in a letter to the sponsor, as described in section X below.

III. PRODUCT TESTING

Product testing for cellular therapies includes, but is not limited to, microbiological testing (including sterility, mycoplasma, and adventitious viral agent testing) to assure safety and assessments of other product characteristics such as identity, purity (including endotoxin), viability, and potency. You should verify that the sponsor will or has performed appropriate testing throughout manufacturing, including manufacture of cell banks, to evaluate the manufacturing process itself and to insure the quality and consistency of the product lots. If the manufacturing process is not controlled, it will be difficult to produce consistent products from lot to lot; this would make it difficult to identify the critical parameters necessary for the desired clinical effect. You should refer to "FDA Guidance Concerning Demonstration of Comparability of Human Biological Products, Including Therapeutic Biotechnology-

Derived Products" (Ref. 13) for additional information. For this section of the IND review, you should describe the specifications used for intermediate and final product release criteria. Specifications are the quality standards (i.e., tests, analytical procedures, and acceptance criteria) that confirm the quality of products and other materials used in the production of a product. Acceptance criteria mean numerical limits, ranges, or other criteria for the tests described. You should assess the appropriateness of acceptance criteria based on any results previously obtained by the sponsor. You also should ensure that the proposed specifications are appropriate to the stage of product development, because release criteria should be refined and tightened as product development progresses toward licensure (see Appendix B). Release tests and specifications should include, but are not limited to, the following:

A. Microbiological Testing

You should verify that the sponsor will perform microbiological testing on cell banks, in-process intermediates, and the final product, as appropriate.

1. Sterility Testing (Bacterial and Fungal Testing)

The following information is provided to instruct you on current practices for sterility testing.

a. Test Method

You should verify that the sponsor will perform sterility testing on the final product. Suitable tests include the test described in 21 CFR 610.12 and the test described in United States Pharmacopoeia (USP) <71> Sterility Testing (Ref. 14). If the sponsor is using another test method, you should assess the adequacy of this alternative test method and confirm that it has been validated to be equivalent to the testing prescribed in 21 CFR 610.12, or inform the sponsor that such validation will be required prior to product licensing pursuant to 21 CFR 610.9. If antibiotics are used in product manufacturing, you should verify that the antibiotics were removed prior to sterility testing. If the antibiotics cannot be removed, you should assess the validity of the sterility assay using the bacteriostasis and fungistasis testing as described in USP <71> Sterility Tests. This assay should be performed to ensure that any residual antibiotic present in the product does not interfere with the results of sterility testing.

b. Test Timing

Sponsors frequently perform in-process sterility testing at critical points during manufacturing. For example, this might be done routinely during extended culture periods and after critical points in manufacturing, such as when cells have

undergone activation or other modification. You should document in the review whether in-process testing is done. You also should assess whether proposed in-process testing is appropriate based on the manufacturing scheme and discuss this with the sponsor as needed. The test method used for in-process sterility testing is at the discretion of the sponsor.

The results of this test should meet acceptance criteria as part of required final product specifications. If the final product is frozen prior to use, the sponsor should perform testing on the product prior to cryopreservation with results available prior to patient administration. However, if the product undergoes manipulation (e.g., washing, culturing) after thawing, particularly if procedures are performed in an open system, the sponsor might need to repeat sterility testing. If cells must be administered prior to obtaining the results from 14-day sterility testing, you should ensure that the sponsor performs sterility testing on a sample taken 48-72 hours prior to final harvest or after the last re-feeding of the cultures and that the sponsor checks the cultures prior to release of the product. This test should be continued for the full 14 days even after the product has been given to the patients. If the results from a 14-day sterility test are not available prior to patient administration, you should ensure that the sponsor performs a gram stain and a final sterility test on the final formulated product. To assure safety, the sponsor should use the no-growth result from the 48-72 hour sterility test and the negative gram stain for release criteria. You also should document and assess the procedures that the sponsor will use if ongoing sterility results show that the product the patient received was contaminated. Since such contamination would suggest a significant risk for human subjects, such procedures must include notification to FDA and all participating investigators in accordance with 21 CFR 312.32(c).

2. Mycoplasma

You should confirm that mycoplasma testing is being performed on the product when there is the best chance of detecting contamination, such as after pooling of cultures but prior to cell washing. You should document that testing is being conducted on both cells and supernatant. There are several potential sources of mycoplasma contamination; two major sources include animal serum products used in culture and the culture facility environment, particularly with open culture systems. You should document whether there is in-process testing for mycoplasma during extended culture procedures. Due to the limited shelf life of many cellular therapy products, it is frequently not feasible for a sponsor to perform the recommended culture-based assay (Ref. 7) for release testing. In these cases, the use of polymerase chain reaction (PCR)-based mycoplasma assays is acceptable during product development. However, you

should discuss with the sponsor that prior to product licensing, data should be provided to demonstrate that the PCR test has adequate sensitivity and specificity.

3. Adventitious Agent Testing

For more information on adventitious agent testing, refer to ICH guidance Q5A: "Guidance on Viral Safety Evaluation of Biotechnology Products Derived From Cell Lines of Human or Animal Origin" (Ref. 15) and Ref. 7.

a. *In Vitro* Viral Testing

When cell lines are used, you should document that *in vitro* viral testing is conducted on the MCB and end of production cells (one-time test). This assay is carried out by inoculation of the test article into various susceptible indicator cell lines. The choice of cells used depends on the species of origin of the product to be tested. The test should include monolayer cultures of the same species and tissue as that used for production of the product, as well as a human and/or a non-human primate cell line susceptible to human viruses. In addition, the test should include a measure of both cytopathic and hemadsorbing viruses. You should document the cell lines used in the review.

b. *In Vivo* Viral Testing

When cell lines are used, you should document that *in vivo* viral assays were conducted on the MCB. These tests are carried out by inoculation of the test sample into animals such as adult and suckling mice and embryonated hen eggs. In some cases, additional testing of guinea pigs, rabbits, or monkeys may need to be included. These assays measure the test animals for any indication of illness. You should document in the review the animals used by the sponsor. The sponsor should provide an assessment of the results of such testing, which you should summarize in your review.

c. Selected Species-Specific Testing for Adventitious Viruses

You should document what specific adventitious agent testing is done at the different stages of manufacturing (e.g., cell banks, final product) and the test methods used. In addition, you should document whether FDA licensed/approved/cleared test kits were used. Since human cell lines are used as the therapeutic product, there should be documentation of testing for human pathogens. Human viral agents can be tested using a PCR-based test system. Tests for CMV, HIV-1 & 2, HTLV-1 & 2, EBV, HBV, HCV, and other human viral agents should be included, as appropriate.

B. Identity

You should ensure that the sponsor verifies the identity of the MCB and the final product by assays that will identify the product and distinguish it from other products being processed in the same facility. If the final product consists of one or more cell lines, you should ensure that the sponsor documents whether there are tests in place to distinguish between the multiple cell lines used. Identity testing for the MCB should include testing to distinguish between multiple cell lines used to produce a single final product. These tests might include assays for cell surface markers or genetic polymorphisms (see Ref. 1 for additional information). For the final product, identity testing is important to ensure that the contents of the vial are labeled appropriately. For additional information on labeling, refer to section VI. (b), below.

C. Purity

Product purity can be defined as freedom from extraneous material, except that which is unavoidable in the manufacturing process (21 CFR 610.13). Testing for purity includes assays for pyrogenicity/endotoxin (see below), residual proteins or peptides used to stimulate or pulse cells, reagents/components used during manufacture, such as cytokines, growth factors, antibodies, and serum, and unintended cellular phenotypes.

1. Residual Contaminants

You should document testing for purity of a cell therapy product including assays for residual peptides, proteins, and reagents used during manufacture, such as cytokines, growth factors, antibodies, and serum. This should also include a measurement of contaminating cell phenotypes or cell debris. For further information, you should refer to ICH Q3 on "Impurities" (Ref. 16).

2. Pyrogenicity/Endotoxin

Endotoxin testing using the Limulus Amebocyte Lysate (LAL) assay method is typically done as an alternative to pyrogenicity testing (see 21 CFR 610.13(b)) for early-phase trials. If the sponsor is using the LAL endotoxin method, you should inform the sponsor that, for licensure, the LAL endotoxin test must be shown, as explained in 21 CFR 610.9, to be equivalent to that of the pyrogenicity test described in 21 CFR 610.13(b). For any parenteral drug, except those administered intrathecally, FDA guidance recommends that the upper limit for endotoxin be 5 EU/kg body weight/dose. Intrathecally administered drugs have a lower limit of 0.2 EU/kg body weight/dose. However, specifications should be based on the sponsor's available data. For further

information, refer to the guideline on "Validation of the Limulus Amebocyte Lysate Test as an End-Product Endotoxin Test for Human and Animal Parenteral Drugs, Biological Products, and Medical Devices" (Ref. 17). You should document in your review the specification for endotoxin testing and verify that testing is on the final product and that results are available prior to release.

D. Potency

A suitable potency assay should be a measure of the relative biological function of the product. You should document and assess all assays used to measure potency. These assays should be quantitative, but in addition, they may include a qualitative biological assay. By the end of Phase 2, the sponsor should have in place a potency assay, consisting of *in vivo* or *in vitro* tests, that measure an appropriate biological activity. This assay should be validated by licensure.

E. Other

1. General Safety

Cellular therapy products are exempt from general safety testing under 21 CFR 610.11(g)(1).

2. Viability

You should ensure that minimum release criteria for viability has been established. For somatic cellular therapies, the minimum acceptable viability specification is generally set at 70 percent. If this level cannot be achieved, you should inform the sponsor that data should be submitted demonstrating that dead cells and cell debris do not affect the safe administration of the drug and/or the therapeutic effect, to support the lower viability specification. For further information, see Ref. 1.

3. Cell Number/Dose

As part of the product testing and release, you should ensure that there are specifications for the minimum number of viable and functional cells. You also should document whether a maximum number/dose of cells to be administered has been established and on what basis.

IV. FINAL PRODUCT RELEASE CRITERIA TESTING

The final product is defined as the final formulated product used for patient administration. The IND review should include a tabulation of the sponsor's proposed specifications (tests, test methods, and acceptance criteria), including test sensitivity and specificity, where appropriate, for the final product. Tests should include assays to ensure the safety, purity, potency, and identity of the product (see section III above). You should confirm that final product release criteria testing is performed on each lot of product manufactured. In some situations, each dose could be considered a single lot depending on the manufacturing process. The results from final product release criteria testing should be available prior to administration. You should clearly indicate in the review additional final product tests whose results will not be available prior to release, together with their specifications, and include a description of the reporting notification process if the acceptance criteria are not met.

V. PRODUCT STABILITY

The objective of stability testing during early phases of the clinical trial is to establish that the product is sufficiently stable for the time period required by the study (21 CFR 312.23(a)(7)(ii)). For later phases of clinical investigation, you should inform the sponsor of the need to expand upon this initial stability information and to begin collecting information needed to develop a final formulation and dating period. You should document and assess the product development plan in the IND review to determine how much stability data is needed for the current phase of investigation. You should assess the stability indicating assays, which may be different from final product release criteria test methods, for adequacy as indicators of product stability. You should document what stability measures were used to support the Phase 1 study. For further information, refer to ICH Q5C: "Quality of Biotechnological Products: Stability Testing of Biotechnological/Biological Products," (Ref. 18), ICH Guideline Q1A(R): "Stability Testing of New Drugs and Products" (revised guideline) (Ref. 19), ICH Guideline Q1E: "Evaluation of Stability Data" (Ref. 20), and when finalized, the draft guidance on "Stability Testing of Drug Substances and Drug Products" (Ref. 21).

Stability Protocol Tests

Stability testing protocols may be appropriate for both in-process material and the final cellular product. If submitted, a sponsor's proposed stability protocol should include a measure of product sterility, identity, purity, quality, and potency. For each test proposed by the sponsor, you should document in the review the test method, sampling time points (there should be a

zero-time point), testing temperature, and other appropriate information, including the adequacy of the assays used to indicate product stability. The stability program should measure the above parameters for the duration of storage required by the clinical protocol or planned further development. You should include preliminary data in the review if submitted.

1. In-Process Stability Testing

If the cells are cryopreserved, you should document the existence of a stability protocol to ensure that the product is stable during the period of cryopreservation, measuring the parameters described above, as appropriate. A comparison is often made of analyses carried out pre-freeze and post-thaw. You also should document any stability testing performed on the product during the holding steps, such as cryopreservation of cells. You should assess whether the time period that the sponsor has established is appropriate.

2. Final Product Stability Testing

You should document and assess any data provided which demonstrate that the product is stable between the time of product formulation and patient infusion, to establish an expiration dating period. You should verify that the sponsor is conducting the testing at the appropriate temperatures and at time points consistent with predicted storage times. You should inform the sponsor of the need to develop validation studies during Phase 3, using conditions that stress the system. If the product is shipped from the manufacturing site to the clinical site, you should ensure that the sponsor documents the time and shipping conditions (i.e., packaging, temperature). You should also assess whether the stability protocol is adequate to demonstrate that product integrity, sterility, and potency are maintained under the proposed shipping conditions. If necessary, you should notify the sponsor that validation studies should be initiated by Phase 3 and completed prior to submission of a biologics license application (BLA).

VI. OTHER ISSUES

A. Product Tracking

For autologous or patient-specific products, the sponsor should have in place a plan to track the therapeutic product from collection to administration of the product and procedures to ensure that the product is segregated from other products in incubators, hoods, and cryopreservation units. You should describe and assess the adequacy of the sponsor's product tracking and segregation system in your review.

B. Labeling

You should document whether there is precise labeling that ensures that the product reaches the proper clinical site if more than one site is involved in the study. In addition, there should be documentation included in the review that describes product labeling throughout the manufacturing process. You should verify that any proposed labeling contains the date of product manufacture, storage conditions, expiration date and possibly time, product name, and patient identifiers. For autologous cell therapies, two unique patient identifiers should be used to minimize the potential for any mix-ups. In addition, as described in 21 CFR 312.6, the label for an investigational product must contain the following statement: "Caution: New Drug – Limited by Federal law to investigational use." For autologous cell therapies, if the donor was not screened or tested for adventitious agents, or if no testing was performed on the cellular product, it is recommend that labeling should carry the warning "Not Tested for Biohazards." For more information refer to Ref. 6, when finalized. To be licensed, the labeling of the final product container and package must conform to the requirements in 21 CFR 610.60-65.

C. Container/Closure

You should include in the IND review a description of the types of container and closure being used. You also should record whether the container used is compatible with the product. For more information, see Ref. 2 and, when finalized, Ref. 3.

D. Environmental Impact

Under 21 CFR 312.23(a)(7)(iv)(*e*), the sponsor must submit either a claim for categorical exclusion under 21 CFR 25.30 or 25.31, or an environmental assessment under 21 CFR 25.40. Such categorical exclusion is ordinarily granted, absent extraordinary circumstances indicating that the specific proposed action may significantly affect the quality of the human environment. Extraordinary circumstances are described in 40 CFR 1508.27 and may include actions that create a potential for serious harm to the environment and actions that adversely affect a species or the critical habitat of a species determined to be endangered, threatened, or entitled to special protection (21 CFR 25.21). You should document in your review your assessment of any extraordinary circumstances. See the guidance on "Environmental Assessment of Human Drug and Biologics Applications" (Ref. 22) for additional information.

E. Validation and Qualification of the Manufacturing Process and Facility

The manufacturing process for somatic cell therapy products entails the use of reagents and source materials of differing complexity, variability and risk for introduction of adventitious agents. Qualification of reagents and source materials, as well as ensuring appropriate controls are in place for monitoring manufacturing consistency and product quality are key elements in ensuring patients receive a safe, consistent, and potent product. Consequently, prior to production of clinical lots and initiation of clinical studies, procedures must be in place to ensure proper manufacturing oversight as outlined in 21 CFR 211.22 in the current good manufacturing practice (cGMP) regulations. This includes programs for product manufacturing quality assurance (QA) and quality control (QC), and the identity of responsible individuals and their duties. In your review, you should describe and assess the adequacy of the sponsor's quality program, including procedures for preventing, detecting, and correcting deficiencies that may compromise product integrity or function or may lead to the possible transmission of adventitious infectious agents.

You should document the changeover procedures described in the IND and ensure that no cross-contamination occurs among an individual patient's cells and other products produced in the same facility. These procedures should be in place by Phase 1 and should include, but are not limited to: area clearance, cleaning and decontamination reagents and rationale for their selection, and segregation of activities. In addition, you should document that aseptic processing steps have been adequately validated. With most cellular therapies, the manufacturing process should be conducted under aseptic conditions due to the lack of final sterile filtration of the product prior to patient infusion. To validate that the process consistently produces a sterile product, media should be substituted for the product and then taken through all steps in the process. You should obtain consultative reviews from the Division of Manufacturing and Product Quality to assess any data submitted by the sponsor. In addition, you should refer to the "Guideline on Sterile Drug Products Produced by Aseptic Processing" (Ref. 23) for further information. You should inform the sponsor that prior to licensure, the facility and all processes used to manufacture the product must be validated.

F. Biostatistics

In CMC IND reviews, there are many significant design and analysis issues in the areas of assay validation, establishing specifications, evaluation of product potency, and evaluation of product stability. Proper statistical design and analysis of such studies are essential to assure reliable documentation of the safety, purity, and potency of the product. You should obtain consultative reviews for relevant portions of the CMC section from the Division of Biostatistics to ensure the adequacy of proposed experimental designs and analytic plans. If applicable, you should

document in your review recommendations from the Biostatistics consult.

VII. PRECLINICAL STUDIES

You should document information provided by the sponsor to support the scientific rationale underlying the proposal. This section should contain a brief summary of preclinical data that was generated using either *in vivo* animal studies or *in vitro* studies to assess the product's activity and efficacy.

VIII. CLINICAL STUDIES

You should provide a brief description of the following in the CMC review:

A. Protocol Title

B. Patient Population

C. Route of Administration

D. Dose

This should include the dosing regimen and whether there is a dose escalation. You also should document the dosing range and the number of patients enrolled in each dose. You should note whether the dose escalation is intra-patient or inter-patient and what time interval/data evaluations occur between dose increases.

E. Frequency

This should include the frequency of dose injections per treatment cycle and the number of proposed cycles.

F. Genetic and Biochemical Testing

You should assess, in conjunction with the clinical reviewer, whether all genetic and/or product-specific biochemical testing being done on the patient is appropriate and whether the test has been appropriately developed and validated for the stage of clinical investigation. You also should evaluate the sensitivity and specificity of the test methods used to demonstrate biological activity (e.g., immunological assay, PCR) and document this information in your review.

IX. RECOMMENDATION

Based on your review of the IND submission, you should describe any information that is missing or incomplete and any issues that require additional clarification. You also should provide an overall assessment, from the CMC perspective, of whether the trial may proceed. You should document all additional information obtained from the sponsor through telephone conversations or faxes. You should note this documentation in the Recommendation Section of the Product Review Template, throughout the review document, or as an attachment to the review. Upon completion, you should sign and date the review and then obtain concurrence from your supervisor.

X. COMMENTS TO SPONSOR

You should draft comments on unresolved issues that should be addressed either (1) before initiating clinical studies after an investigation has been placed on clinical hold or (2) as product development progresses (i.e., when there is no clinical hold) as discussed below. Refer to SOPP 8201, "Issuance of and Response to Clinical Hold Letters for Investigational New Drug Applications" (Ref. 24), for additional information. You should forward your comments to the RPM for inclusion in a letter to the sponsor, after you have obtained supervisory concurrence on your review.

A. Clinical Hold

These are comments that the sponsor must satisfactorily address prior to allowing clinical studies to proceed after FDA has imposed a clinical hold. These comments must fit the criteria listed in § 312.42(b).

B. Non-Clinical Hold

These are comments that the sponsor should address as product development progresses. In some cases, a sponsor may need to address specific manufacturing issues by a certain point in

clinical development, such as prior to initiation of Phase 3 studies. Your comments should inform the sponsor of any such issues.

Effective Date

Insert signature date

Appendices

Appendix A – Product Review Template (Somatic Cell Therapy)
Appendix B – Review Considerations for Development of Final Product Release Criteria Specifications and Stability Protocols
Appendix C – Relevant Regulatory Documents