Control of Allogeneic Donor Cells and Characterization: Integrating into Cell Therapy Regulatory Strategy

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Allogeneic Donor Cell Control and Characterization: Integrating into Cell Therapy Regulatory Strategy

• Critical Material Attributes of Donor Derived Starting Material: How to Incorporate into Regulatory Strategy
  • Example Strategy: Potency
  • Example Strategy: Process Understanding
  • Case Study: Potency

• Regulatory Considerations for Allogeneic Donor-Derived iPSC Products
  • Example Strategy: Donor Exemption Request

• Allogeneic Donor Selection/Testing for Global Products
  • Case Study
Characterization of donor cells

Characterization of cell banks

Infectious disease testing of donor cells (US)

Type of Donor Material for Allogeneic Products Can Direct Testing Strategy

Autologous

Allogeneic (Limited Lot Size)
  • Cord blood-derived

Allogeneic (Larger Lot Size)
  • Cell bank derived

iPSC-Derived
  • Manufacture multiple products from a single donor cell

Manufacturing potential: potential quantity of products derived from a single donor
Critical Material Attributes of Donor Derived Starting Material: How to Incorporate into Regulatory Strategy
Starting Materials are Complex for Allogeneic Cell Therapies

- **Phase I**: Materials purchased from approved suppliers and accepted based on supplier CoA or NDC number
- **Pivotal & Commercial**: In-house identity testing and additional functional testing likely will be needed

Often a Risk-Based Approach is Used Early in Development

Critical material attributes of these materials are often not well-defined until after clinical data have been generated

Even when not part of the material specification, In-house characterization studies are needed:
- Assess any potentially relevant phenotypic markers for the starting material
- Correlate raw material attributes to product functionality/MOA in vitro
- Corelate raw material attributes to process performance
- Allow eventual identification of correlation to clinical data and identification of CMAs/CQAs

Potential Critical Material Attributes for Allogeneic donor derived material can relate to:

- **Growth/process potential**:
  - Phenotypic markers
  - Genotypic markers
  - Cell differentiation and proliferation
  - How supplied (cryopreserved or fresh)
- **Product CQA (Safety)**:
  - Karyotype
  - Genotypic markers
  - Donor profile
  - Genetic stability
- **Product CQA (Efficacy)**:
  - Innate cell killing
  - Phenotypic markers
  - Genotypic markers
- **Attributes related to ability to be genetically modified**:
  - Transduction efficiency
  - Cell biology

Early Characterization Studies are Important to Allow CQA/CMA Identification from Later Clinical Data

Even when not part of the material specification, In-house characterization studies are needed:
- Assess any potentially relevant phenotypic markers for the starting material
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- Corelate raw material attributes to process performance
- Allow eventual identification of correlation to clinical data and identification of CMAs/CQAs
### Key Considerations Prior to Implementing CMAs

<table>
<thead>
<tr>
<th>Donor-to-Donor Variability</th>
<th>Supply</th>
<th>Development Timing</th>
<th>Comparability</th>
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<tbody>
<tr>
<td><strong>Key Questions</strong></td>
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<tr>
<td>Is donor-to-donor-variability well understood for the manufacturing process prior to CMA implementation?</td>
<td>Is the desired material profile sustainable from a supply standpoint?</td>
<td>Can the CMA be defined prior to pivotal trials so that the trial is designed to support the hypothesis?</td>
<td>Can you appropriately demonstrate comparability of the donor-derived material pre- and post-implementation of the CMAs?</td>
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#### Example Strategies

- **Donor-to-Donor Variability**
  - Manufacture sufficient lots in early phase studies in order to assess potential effects of donor variability on product.
  - Hypothesis-based approach to ensure lots manufactured throughout early phase encompass the range of the desired characteristics.

- **Supply**
  - Work with suppliers on the TPP for donor material. Calculate how restrictive the desired specification could be from a national and global standpoint.

- **Development Timing**
  - Early characterization studies inform a list of potential CMAs.
  - Early clinical studies include sufficient range of batch characteristics to allow identification of potential CMAs/CQAs.
  - Pivotal trial is powered to test potential CMAs/CQAs.

- **Comparability**
  - Tightening or changing the specification for the donor material is accompanied by a risk assessment of the change on product quality.
  - Comparability studies are designed to show whether change results in comparable product.
  - Opportunities for discussion with regulators: EOP2/pre-pivotal, pre-comparability, post-comparability, pre-BLA.
CellTrans, Inc. submitted biologics license application (BLA) 125734 seeking to market donislecel, a cellular therapy product composed of allogeneic islets of Langerhans for the treatment of “brittle type I diabetes mellitus (T1D) in adults whose symptoms are not well controlled despite intensive insulin therapy.”

Each lot of donislecel is manufactured from a deceased donor pancreas procured via the Organ Procurement and Transplantation Network (OPTN), and is for the treatment of one patient.

Highlights from meeting:

- Each lot of donislecel is derived from one allogeneic, cadaveric donor pancreas, with each patient receiving up to three lots over the course of their treatment. Inherent variability between donor pancreata contributes to starting material variability. Thus, starting material variability of quality is one of the limitations of the manufacturing process. We ask the Committee to consider the contribution of purity, one of the CQAs, to product safety, efficacy, and manufacturing quality, and discuss whether evaluation of purity and potency is adequate to ensure that the manufacturing process will produce product lots of consistent quality.

- Each dose of donislecel is derived from one deceased donor pancreas. Patients are expected to receive up to three doses to attain the suggested clinical outcomes. Based on data provided from clinical trials, the critical quality attributes are highly variable, and it is not clear how the Applicant achieves lot-to-lot manufacturing consistency in terms of islet purity and potency.

- The Committee discussed that it is difficult to have consistent product lots because each lot is derived from a different donor pancreas. In addition, the committee noted that it would be good to have better quality control of the product to avoid administration of multiple doses. The committee suggested adding rapid quality control assays to aid in controlling lot-to-lot variability.

**CASE STUDY: UNDERSTANDING EFFECT OF DONOR VARIABILITY ON POTENCY**

**Background:**

- CellTrans, Inc. submitted biologics license application (BLA) 125734 seeking to market donislecel, a cellular therapy product composed of allogeneic islets of Langerhans for the treatment of “brittle type I diabetes mellitus (T1D) in adults whose symptoms are not well controlled despite intensive insulin therapy.”

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**Cellular, Tissue, and Gene Therapies Advisory Committee (CTGTAC) Meeting Session on Product Characterization (AM Session) April 15, 2021**

[https://www.fda.gov/media/147524/download]
Regulatory Considerations for Allogeneic Donor-Derived iPSC Products
Donor Eligibility Considerations for iPSC-Derived Products

- iPSC-Derived allogeneic therapies can theoretically use a single donor to manufacture product throughout lifecycle
- Understanding donor-to donor variability may be important in early R&D efforts prior to selection of line for routine manufacture
- Prior to cell line creation, variability of donor cell candidates may be assessed for:
  - Genome editing capability
  - Differentiation capability
  - Genetic mutations
  - Othered desired profile(s)
- Same donor eligibility and screening rules apply for iPSC allo donor as other allogeneic donors
  - Note nationality
    - US donor screening for Variant Creutzfeldt-Jakob Disease (vCJD) risk excludes most Europeans from HCT/P donations.
    - For Japan: United Kingdom, France, Italy, Ireland, Netherlands, Portugal and Spain are prohibited to be used as blood sourcing country and are precautionary not to be used as a tissue sourcing country (Enhancing the Quality and Safety of Medicines and Medical Devices Using Human-Derived Ingredients and Amendments PFSB No. 0629001 PFSB No. 1213002)
iPSC-Derived Products

- Donor eligibility from a regulatory standpoint does not differentiate for iPSC at this time
  - Genetic testing not specifically mentioned for donor derived products by any health authorities
  - However, genetic safety (i.e. genetic stability, oncogenic mutations, etc.) will need to be demonstrated for cell therapy product(s)
  - “Tumourigenicity/genetic stability should be evaluated for stem cell preparations that undergo extensive in vitro manipulation such as prolonged cell culture” EMA/CAT/852602/2018 Guideline on quality, non-clinical and clinical requirements for investigational advanced therapy medicinal products in clinical trials

- A thorough risk assessment of the process/product should be performed and drive testing strategy
- Extensive testing in donor cells, stocks, banks, in-process, and at product release can de-risk safety concerns with regard to virus propagation, genetic variants/instability
## iPSC-Derived Products: Example Testing Points

<table>
<thead>
<tr>
<th>Test and Examples</th>
<th>Donor Cells</th>
<th>iPSC Stock</th>
<th>Master Cell Bank</th>
<th>Product</th>
</tr>
</thead>
<tbody>
<tr>
<td>Characterization</td>
<td>Potential CMAs RNA-Seq</td>
<td>Potential CMAs Differentiation potential</td>
<td>Phenotypic Genetic characterization</td>
<td>Phenotypic Functional characterization</td>
</tr>
<tr>
<td>Safety</td>
<td>Relevant viruses of interest not tested on donor (HHV 6, 7, 8; B19)</td>
<td>Relevant human viruses not tested on donor (HHV 6, 7, 8; B19) Viruses deriving from a risk assessment of raw materials</td>
<td>Relevant viruses of interest not tested on donor (HHV 6, 7, 8; B19) Genetic testing (Karyotype, FISH, SNP arrays) Off-target analysis Viruses deriving from a risk assessment of raw materials Non-specific viral tests RT activity (RCR, RCL) Clonality/variants</td>
<td>Relevant human viruses that may propagate during expansion culture (HIV, HTLV, HBV, HCV, HHV 6, 7, 8; B19) Viruses deriving from a risk assessment of raw materials Off-target analysis Genetic testing Clonality/variants</td>
</tr>
</tbody>
</table>

**Diagram:**
- **Donor Cells** → Selection Re-programming → Genome editing → Cloning Expansion Selection → Cell banking → Master Cell Bank → DS Manufacture
EXAMPLE STRATEGY: iPSC STOCK DERIVED FROM DONOR MATERIAL THAT DOES NOT MEET US REQUIREMENTS

Challenges:

• Donor does not meet requirements in 21 CFR 1271 (not all testing requirements met; CLIA certified lab not used)

• A globally accepted product is desired for future clinical trials

Example Strategy:

• Submit exemption request per §1271.155(a) to FDA

• Propose alternative testing plan (at various bank and product levels) for virus/infectious diseases of concern
Allogeneic Donor Selection/Testing for Global Products
# Key Regulations and Guidance for Human Cell Donor Eligibility Determination

<table>
<thead>
<tr>
<th>FDA</th>
<th>EMA</th>
<th>PMDA</th>
<th>NMPA</th>
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<tbody>
<tr>
<td></td>
<td></td>
<td>• The Quality and Safety Assurance of Medicines or Medical Devices Manufactured Using Human (Homogenous)-Derived Cells and/or Tissue FSB Notification No.0912006</td>
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## Infectious Disease Testing/Screening Requirements for Allogeneic Cell Donors

<table>
<thead>
<tr>
<th>Infectious Disease</th>
<th>US</th>
<th>Europe</th>
<th>Japan</th>
<th>China</th>
</tr>
</thead>
<tbody>
<tr>
<td>HIV1</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>HIV2</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>HBV</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>HCV</td>
<td>X</td>
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<td>X</td>
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<tr>
<td>TSE/CJD</td>
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<tr>
<td>Treponema pallidum</td>
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</table>
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<table>
<thead>
<tr>
<th>Infectious Disease</th>
<th>US</th>
<th>Europe</th>
<th>National European Authorities*</th>
<th>Japan</th>
<th>China</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chlamydia</td>
<td>X</td>
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<tr>
<td>Neisseria gonorrhoeae</td>
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<tr>
<td>Mycobacterium tuberculosis</td>
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<td>X</td>
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<td>HTLV1/HTLV2</td>
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<td>Epstein-Barr</td>
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<tr>
<td>Parvovirus B19</td>
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<tr>
<td>Trypanosomiasis</td>
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<td>In certain circumstances</td>
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<tr>
<td>HEV</td>
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<td>X</td>
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<tr>
<td>Chikungunya</td>
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<td>In certain circumstances</td>
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<tr>
<td>CMV</td>
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<td></td>
<td>X</td>
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<tr>
<td>Ebola</td>
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<td>In certain circumstances</td>
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<tr>
<td>Vaccinia</td>
<td>X</td>
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<td></td>
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<tr>
<td>Malaria</td>
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<td>In certain circumstances</td>
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<tr>
<td>West Nile virus</td>
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<tr>
<td>Zika</td>
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<td></td>
<td>NO LONGER REQUIRED</td>
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<tr>
<td>SARS-CoV-2</td>
<td></td>
<td></td>
<td>In certain circumstances</td>
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<tr>
<td>Toxoplasma</td>
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<td></td>
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<tr>
<td>EBV</td>
<td></td>
<td></td>
<td>In certain circumstances</td>
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## Donor Testing Window and Procedures for Allogeneic Cell Donors

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<tr>
<th></th>
<th>US</th>
<th>Europe</th>
<th>Japan</th>
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</table>
| **Donor Testing Window** | 7 days  
30 days (bone marrow) | 7 days  
30 days (allogeneic bone-marrow stem cell or peripheral blood stem-cell donor, taking into account that retesting at the time of donation will be informative) | Appropriate timeframe |
| **Repeat Donor Testing** | Repeat donor testing not required*  
*NAT required | Repeat sampling and serology testing is required after 180 days, unless any of the following specific exemption criteria are met. If samples from a living donor undergo serology testing and are also tested by molecular tests (i.e. NAT) for HIV, HBV and HCV, re-testing after a time interval is not required. | Retesting shall be performed in an appropriate timeframe while considering the window periods. In the case where umbilical cord blood is donated as a human cell/tissue material and the cord blood is supplied in a manner which satisfies the standard specified in the MHLW ordinance in accordance with the provision in Article 32 of the “Law on Promotion of Appropriate Donation of Hematopoietic Stem Cells for Transplant” (Law No. 90, 2012), re-testing is not always necessary. The same applies when the level of control of cells other than cord blood is equivalent to the above standard. |
CASE STUDY: CORD BLOOD AS STARTING MATERIAL FOR A GLOBAL PRODUCT
Pre-CTA Stage; 6 National Health Authorities

General Question: Is specification for the cord blood starting material appropriate?

- Cord blood itself should be tested for human viruses
- Appropriate control of donor source supply will be a critical aspect to ensure consistency
- Risk assessment for SARS-CoV-2 and Zika should be performed
- Add Hepatitis E and malaria testing to maternal donor test panel
- CE marked tests should be used
CASE STUDY: CORD BLOOD AS STARTING MATERIAL FOR A GLOBAL PRODUCT

Pre-CTA Stage; 6 National Health Authorities

Challenges:

• Cord blood is licensed for allogeneic stem cell transplant. When used as a starting material for cell therapy manufacture, additional controls may apply to mitigate risk from manipulation/expansion of cells.

• Limitations in implementing additional donor screening.

• Volume limitations on material.

• Limitations on how cord blood can be packaged as typically approved in US under BLA (additional aliquots not always available for additional tests).
CASE STUDY: CORD BLOOD AS STARTING MATERIAL FOR A GLOBAL PRODUCT

Pre-CTA Stage; 6 National Health Authorities

Strategy:

- Test for relevant viruses/infectious diseases in-process and/or at drug product release as opposed to directly in cord blood.
- Work with suppliers for how to implement additional donor tests.
- Develop supplier questionnaire to ensure global compliance of the starting material for global trials.

Umbilical cord blood unit (CBU)

- CBU thaw
- Cell wash
- Enrichment
- Selection
- NK cells
- Modification
- Cell Culture
- Harvest
- Wash
- Formulate
- Fill

1 batch = multiple doses

❖ Testing points in addition to donor testing
Summary

• Regulatory and legal requirements vary by region and country.
• For a global product, regulatory review of donor selection and testing is critical to ensure compliance for your allogeneic starting material.
• Characterization studies and well-designed trials allow for identification of critical material attributes of donor-derived starting materials.
• iPSC derived products do not have more stringent regulatory donor eligibility criteria than any other allogeneic donor material.
  • A thorough risk assessment and strategy should be evaluated for the product
  • Testing at various levels (donor cells, banks, during DS/DP manufacture) is important to address additional safety concerns from allogeneic donor cells not covered by donor testing.
Thank you!
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