Alliance for Regenerative Medicine

July 29, 2024

Dockets Management Staff (HFA-305)
Food and Drug Administration
5630 Fishers Lane, Rm. 1061
Rockville, MD 20852

Re: Docket No. FDA-2024-D-1243 for *Safety Testing of Human Allogeneic Cells Expanded for Use in Cell-Based Medical Products*

Dear Sir/Madam:

The Alliance for Regenerative Medicine (ARM) appreciates the opportunity to comment on the draft guidance document, *Safety Testing of Human Allogeneic Cells Expanded for Use in Cell-Based Medical Products.* As clinical trials of cell and gene therapies (CGTs) using allogeneic cells are increasing, this topic is timely for ARM members.

ARM is the leading international advocacy organization championing the benefits of engineered cell therapies and genetic medicines for patients, healthcare systems, and society. As a community, ARM builds the future of medicine by convening the sector, facilitating influential exchanges on policies and practices, and advancing the narrative with data and analysis.

We actively engage key stakeholders to enable the development of advanced therapies and to modernize healthcare systems so that patients benefit from durable, potentially curative treatments. As the global voice of the sector, we represent more than 400 members across 25 countries, including emerging and established biotechnology companies, academic and medical research institutions, and patient organizations.

General Comments

To strengthen this guidance when it is finalized, ARM suggests including reference to other relevant FDA guidance documents on cell bank characterization and qualification and to ICH Q5A and Q5D guidelines (e.g., for testing requirements of master cell banks) to assist in convergence of requirements globally.

ARM recommends the FDA provide additional guidance on the definitions of “extensive” expansion and “many” vs. “limited” individuals, either through numerical ranges for these criteria or factors to consider. In addition to MCB expansion potential and target number of patients to be treated per batch, a potential factor to consider in determining how extensively a cell bank
should be tested for safety is the degree to which the bank is genetically manipulated. For example, if a product is continually processed (e.g., establishment of an MCB), but undergoes several genetic modifications, the risk for adventitious agent contamination may increase and the Sponsor might consider more extensive testing (Section V). If there are other factors that sponsors should consider, ARM suggests the Agency provide examples.

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We would also suggest providing additional guidance surrounding testing for human pathogens and implications for acceptability of donor material such as leukapheresates. ARM also recommends the Agency consider the relationship between acceptable IDM testing of the donation and the benefit of repeating this testing on either the cell bank or on the drug product, especially when the operations associated and the time between the acceptable testing of the donation and creation of the bank or manufacture of the product has little risk of introducing the pathogen into the product. Guidance on the utility of the *in vitro* virus test to be suitable in these situations may be helpful.

We suggest expanding the scope of the guidance to include the different cell sources for allogeneic programs covered (healthy donor PBMC, iPSC, HSC, other) and how they are viewed (continuous cell lines, limited expansion, extensive expansion). Some cell types may have additional complexities regarding safety assessments, the impact of their differentiated state, and the process used to arrest differentiation.

We provide specific line-by-line comments in the table below. Thank you for your consideration of these comments.

Sincerely,

Michael Lehmicke
Senior Vice President, Science and Industry Affairs

Specific Comments

IV. Considerations for Cell Safety Testing			
Lines/Section	*Draft Guidance Text*	*Comment/Recommendation*	*Suggested text*
Lines 113 – 115	"The nature and extent of cell safety testing needed to provide adequate assurance of product safety will generally depend on the expansion potential of the cells and the number of individuals the cell-based medical product is capable of treating."	ARM recommends adding a reference here to Table 1, which provides useful details about the requirements based on the expansion potential and number of individuals a product is able to treat.	"The nature and extent of cell safety testing needed to provide adequate assurance of product safety will generally depend on the expansion potential of the cells and the number of individuals the cell-based medical product is capable of treating (see Table 1)."

Continuous Cell Lines			
Lines 123 – 124	"Continuous Cell Lines: Cellular products may be produced from continuous cell lines, including induced pluripotent stem cells, embryonic stem cells, ..."	Stem cells including iPSC and embryonic stem cells are listed here. Since these cells can differentiate with passage number and this changes functionality, it would be useful to distinguish between stem cells as continuous cell lines vs. cancer cell lines or transformed cell lines which are truly immortal.	
Primary Cells			
Lines 132 – 144	"Primary Cells Capable of Extensive Expansion in Culture"	It would be useful to provide a definition of the term "extensive expansion," by indicating whether the distinction is the number of doublings, time in culture, or number of growth characteristics of the cells, and indicating what the endpoint is—cell death or a particular phenotype.	

V. Testing Recommendations for Highly Expanded Cells			
Lines/Section	*Draft Guidance Text*	*Comment/Recommendation*	*Suggested text*
Section: Testing Recommendations for Highly Expanded Cells	None (addition requested)	ARM recommends referencing ICH Q5D where appropriate. We also recommend adding to this section content on testing of the drug substance (DS) and drug product (DP), including in-process testing during expansion and differentiation.	
Lines 175 – 177	"This section contains recommendations for testing cell banks of highly expanded primary cells and cell banks made from continuous cell lines, including pluripotent	ARM recommends providing a definition of "highly expanded" primary cells, as described in the general comments.	

	stem cells, cancer cells, and transformed cells."		
Lines 186 – 189	"Instead, the cell bank of genetically modified stem cells would generally be considered the MCB, and the cell safety testing described below should be performed on those genetically modified cells since there is potential for adventitious agent contamination during the genetic modification."	ARM requests FDA additionally provide recommendations on safety testing of the cells prior to testing of the MCB (including in-process and raw material testing during handling and genetic modification of the cells). If FDA is not seeking safety testing prior to testing of the MCB, or is only seeking this testing under certain circumstances, then we request the guidance elaborate on this as well.	
A. Master Cell Bank			
Lines 209 – 211	"Alternative assays may be used to detect mycoplasma, but such an assay should be shown to have sensitivity that is comparable to the compendial method."	True comparability is difficult to demonstrate and requires the culture method to be established in the lab. ARM therefore recommends rephrasing to indicate that a comparability study is not required but a sensitivity of 10 CFU/mL is required.	
Lines 213 – 218	"Human pathogen testing using polymerase chain reaction (PCR) may include testing for human immunodeficiency virus (HIV) -1 &2, human T cell lymphotropic virus (HTLV) 1 & -2, hepatitis viruses B and C, 216 cytomegalovirus (CMV), Epstein-Barr	ARM suggests providing more details around the selection of appropriate human pathogens to be tested, since some pathogens are highly prevalent in patient populations. ARM recommends allowing a tiered testing approach and focusing on screening for active infection status vs. identifying prior/inactive infections. Additional guidance would be helpful on which test method(s) FDA considers more acceptable to better differentiate between active and inactive	

	virus (EBV), human parvovirus B19, human papillomavirus (HPV), human herpes viruses (HHV) -6, -7, and 8, John Cunningham (JC) virus, and BK virus, as appropriate."	infection (e.g., NAT, PCR, antibody test). ARM recommends additional text that testing should take a risk-based approach based on cell source.	
Lines 218 – 220	"FDA should be consulted for application-specific testing recommendations when cells will be used in immunocompromised individuals."	ARM suggests FDA provide what additional testing recommendations are recommended beyond what is already described in the guidance testing recommendations for cells to be used in immunocompromised individuals since many cell therapy treatments require immunosuppression to avoid immune mediated degradation of the cells.	
Lines 242 – 243	"Alternatively, a high throughput sequencing method may be used instead of in vivo adventitious virus testing to detect contaminating viruses."	ARM recommends aligning with ICHQ5a, which allows non-targeted next generation sequencing (NGS) to supplement or replace the in vitro assays (Section 3.2.2. ICHQ5a) or stating exceptions to which NGS/high throughput methods may be used to supplement in vitro tests. ARM recommends adding that this test method would also be able to replace the species-specific virus testing since this (agnostic) test method would be able to detect all the various viruses in one test.	"Alternatively, a high throughput sequencing method may be used instead of in vivo adventitious virus testing, in vitro testing, and species-specific virus testing to detect contaminating viruses."
Lines 251 – 252	"Transmission electron microscopy should be performed to detect virus particles."	Because use of TEM is not quantitative and virus size could differ across a wide range, ARM suggests adding recommendations on the sample size for this test.	
Lines 259 – 263	"Species-specific virus testing should be performed. If human	The guidance states in lines 281 – 286 that "It may be acceptable to reduce or eliminate testing of the	

	sourced cells contact rodent cells or rodent-derived reagents during manufacturing, then testing for mouse/rat/hamster viruses should be performed. Likewise, if the human sourced cells contact simian or insect cells or reagents, then testing for simian or insect viruses should be performed."	human-sourced cells for animal viruses if the reagent manufacturer performs and documents adventitious agent testing for the animal-derived reagents consistent with 9 CFR 113.53 and 9 CFR 113.47. Reagent testing documentation should be submitted in the IND or BLA submission." It is unclear if this also would be accepted for rodent cell-derived raw materials. ARM recommends that a well-documented rodent testing performed on the rodent-derived reagent could replace the need for performing rodent virus testing of the cell bank.	
Lines 288 – 299	Testing for the presence of residual viral and plasmid reprogramming vectors used in the creation of induced pluripotent cell lines should be performed on either the cell bank, drug substance, or final product	Given the rapid advancement of non-viral or non-plasmid based reprogramming vectors (e.g., mRNA, siRNA, etc.), we suggest removing "viral and plasmid."	Testing for the presence of residual ~~viral and plasmid~~ reprogramming vectors used in the creation of induced pluripotent cell lines should be performed on either the cell bank, drug substance, or final product
Lines 290 – 292	"An acceptance criterion with justification for acceptable levels of residual programming vectors should be established,"	Acceptance criterion" suggests the agency recommends karyological testing as a release test. ARM recommends that a qualified and 'fit for purpose' method would be sufficient for this testing. If FDA does have certain recommended testing criteria, ARM suggests stating them, especially if FDA is seeking validation for this test method. Please also clarify whether testing clearance of re-programming raw materials should be performed on cell bank, DS, DP, which will only be used in Phase III and eventual	

		marketing, or if such testing is also a requirement for earlier phases.	
Lines 301 – 302	"Whole genome sequencing and analysis should be performed on cell banks of continuous cell lines and genome edited cells."	Because WGS will likely identify variants of uncertain significance, ARM recommends that a tiered risk assessment approach should be acceptable, with targeted analysis focusing on known oncogenes, homologous recombination pathways, and genes associated with the disease being treated. ARM also requests clarification if other targeted methods would be acceptable approaches to evaluate genomic integrity.	
Lines 303 – 307	"Cell lines that are cultured extensively often accumulate mutations during cell expansion. Mutations in protooncogenes, such as p53, are of particular concern."	Current language suggests p53 is a proto-oncogene by default, which it is not. P53 becomes a proto-oncogene upon onset of certain mutations while it is not in its wild-type form. We suggest the language change to the right.	Mutations in genes such as TP53, which lead to P53 becoming a proto-oncogene~~s such as p53~~ are of particular concern.
Lines 324 – 328	"Whole genome sequencing as described above is the recommended method of testing genome integrity. Alternatively, if cytogenetic testing is performed, G-banding analysis or other sensitive methods should be used to confirm the cells have a normal karyotype."	ARM recommends clarifying if and under what circumstances karyology may be appropriate.	
Working Cell Bank			

Line 375	None (addition requested)	ARM recommends adding language around risk assessments. Safety testing expectations for MCB and WCB were described in sections A and B, respectively. We suggest adding a separate section C to clarify safety testing (especially viral safety) expectations for DS/DP, especially for the scenarios of rows 1-3 of Table 1, in which product use is for many individuals and cell expansion and/or differentiation are likely necessary steps when using cell banks as starting material to make the final cellular therapy product. We suggest restating or referencing the FDA guidance, Considerations for the Development of Chimeric Antigen Receptor (CAR) T Cell Products, dated Jan 2024: "For allogeneic CAR T cells, where each product lot is meant to treat multiple patients, additional testing beyond what is described in this section may be appropriate. For example, additional adventitious agent testing..."	

V. Testing Recommendations for Cells with Limited Expansion Potential			
Lines/Section	*Draft Guidance Text*	*Comment/Recommendation*	*Suggested text*
Line 389	"Human pathogen testing using PCR, as described in section V of this guidance"	ARM recommends that if the source of the allogeneic product is healthy donor PBMC (or other human source cells collected at a licensed collection center), and the collection has been tested for the IDM panel (those tests listed as well as other relevant tests), the in vitro virus testing should be considered to adequately control the quality of the product, and the human pathogen testing would	

		not need to be repeated on the cell product for primary cells that cannot be expanded extensively.	
Lines 395 – 397	"Genome edited cells that are not extensively expanded in culture should undergo targeted sequencing to assess the frequency of editing at confirmed off-target sites and to ensure the desired on-target editing outcome has occurred."	ARM recommends specifying that sponsors should use a risk-based approach to determine whether this targeted testing is performed as part of characterization or as part of release testing.	