

MINUTES
Adenovirus Reference Material Working Group Meeting
May 31, 2001
12-1:30 PM, ASGT Meeting, Seattle, WA

Summary

The Adenovirus Reference Material Working Group met May 31, 2001 to review the requests for proposals (RFPs) for the characterization phase: RFP 8.0-Participation in the Assignment of Particle Concentration, RFP 9.0-Participation in the Assignment of Infectious Titer, RFP 10.0-Other Characterization, RFP 11.0-Short-term Stability and Shipping Condition Validation, and RFP 12.0-Long-term Stability. All remaining RFPs will be posted on the website by June 30, 2001. Deadline for submissions will be July 30, 2001. Notifications will be completed by September 5, 2001. The Working Group determined that there was a need for Testing Phase 293 Cell Bank. No RFP will be posted for this activity due to the timeline but a volunteer institution will be identified separately.

The timeline for production was also reviewed with anticipation that the purified Adenovirus Reference Material will be vialled by mid-September. Canji harvested the virus bank material May 31 and a back-up virus bank material was scheduled for harvest June 7, with final processing and vialing to occur June 11 week. Virus bank material will be sent to Introgen based on an interim Certificate of Analysis for process development work and production. The interim Certificate will include Canji's in-house testing (confirming identity and activity), mycoplasma and sterility testing. The virus bank release will be completed prior to vialing of the purified reference material. Introgen anticipated initiating production such that vialing of the purified reference material would take place in early September.

Given that "t₀" testing will need to take place within a few weeks of vialing, the Working Group has targeted its next meeting for August or very early September to review and award the bids for the characterization phase. A flyer (attached) was distributed at the American Society of Gene Therapy Conference (May 31-June 3) to invite groups to participate in the characterization phase of the adenovirus reference material project.

Details

The RFP 8.0 protocol and Excel spreadsheet were discussed. There was consensus that the protocol was acceptable for posting. The spreadsheet will be password protected and the password will not be issued to any participants but will be kept by WBF. All data must be entered and any changes to calculations will be done only by a third party (WBF, FDA).

The RFP 9.0 protocol has not been distributed for general comment. It will be sent shortly. The assay for infectious titer will be a 96-well format CPE-based assay using HEK 293 cells. There will be a corresponding spreadsheet for calculating normalized

adjusted standard (NAS) titers that take into account adenovirus diffusion. The issue of whether or not to provide the cells to laboratories for the testing was raised and discussed again. There was agreement that a testing phase non-CGMP cell bank would be provided and that only sterility, mycoplasma and viability testing would be required.

[Post meeting note: Introgen, Canji and Invitrogen offered to make the bank. It was decided among the three and in discussion with WBF that Invitrogen will make the testing phase 293 cell bank. This bank will consist of at least 50 (100 preferred) x 1 mL vials at approximately 1×10^7 cells/mL in the same freeze medium as the Master Cell Bank. It will be derived from the 293 HEK Master Cell Bank already donated (donation by UAB). Invitrogen will provide a Certificate of Analysis that will also indicate the passage number and number of cells/mL as vialled. See also the minutes of the June 11, 2001 teleconference.]

Further discussion of RFPs 8.0 and 9.0 included whether there should be a maximum number of participating laboratories (minimum of 6 required). No maximum will be set but only qualified labs (based on review of information supplied in the bids) will be considered.

To develop a stability plan, several key parameters were discussed:

- The number of sites participating in stability testing (single vs. multiple)
- Definition of the stability indicating assays
 - Variability
 - Sensitivity
 - Correlation to loss of infectious titer
- Sterility testing/container closure integrity studies
- Storage conditions to be evaluated ($\leq -55^{\circ}\text{C}$, 4°C , Room temperature, other)
- Frequency of testing
- Number of replicates per timepoint
- Definition of T=0
- Shipping validation requirements
- Accelerated studies
 - Multiple temperatures
 - Freeze thaw studies
- Timeframe for availability and shipment of samples for testing

These parameters were discussed in the context of the amount of material to be used for stability, the length of stability testing at the proposed storage condition, the possible sources for instability in handling the material in the specific formulation, concentration, vial configuration and shipping conditions. After considerable discussion, there was consensus that the stability assessment of the material should be split into a long term study to be conducted on material stored at $\leq -55^{\circ}\text{C}$ in 6 month intervals for two years and annually thereafter until 5 years, and a short term study focusing on use conditions and shipping condition validation. The short-term study would include evaluating multiple temperatures, multiple freeze thaws, shipping condition validation and would be initiated as quickly as possible. There were motions to select a single site to perform each study

with the clarification that it would be possible for one site to conduct both studies. The motions were approved as 26 Yes, 1 No and 3 Abstentions for the long term study and 27 Yes, 0 No and 3 Abstentions for the short term studies, respectively.

The stability indicating tests that will be required were narrowed down to particle number by the OD260/SDS method issued by the Working Group, infectious titer assay (by the SOP issued by the Working Group), and anion exchange HPLC (with the method to be submitted as part of the proposal). This selection was made based on their relative precision and sensitivity in detecting changes. Bids may include additional testing.

Bids for the short-term stability study should include proposals for number of replicates, specific strategies for accelerated studies and shipping condition validation (based on ATCC shipping configuration and worldwide distances) and any additional methodologies for characterizing stability. The amount of material required to perform the studies, the testing laboratory qualifications and a procedure for AE HPLC (including assay reference standard descriptions and stability information) would also need to be included. For the long-term stability study the proposal would include the number of replicates, any additional testing proposed such as container integrity and timing of such testing, the amount of material required to perform the studies, the testing laboratory qualifications and a detailed procedure for AE HPLC (including assay reference standard descriptions and stability information).

The T=0 timepoint will be defined as the date of manufacture/filtration (in this case the day before final vialing occurs at Introgen). Initial stability indicating tests should be performed within 30 days of the date of manufacture and will be assumed to reflect the true T=0 point. Testing intervals will be scheduled from T=0 (with a +/- 2 week window for testing to be performed).

Other issues discussed at the meeting included what information to include on the vial label. A motion was made that the manufacturer be listed as the Adenovirus Reference Material Working Group (or some version thereof) rather than the corporation's name. The motion was approved as 18 Yes, 1 No, 6 Abstentions.

Finally, it was agreed that the timeframe for shipping samples for short-term and initial long-term stability timepoints would be separate from shipment of the bank to ATCC.

Attendees (including those participating by teleconference):

Estuardo Aguilar-Cordova, Harvard University

Steven Bauer, CBER/FDA

Jeff Beecham, Harvard University

Flavia Borellini, Cell Genesys

Mark Bove, Genetic Therapy Inc.

Andrew Byrnes, CBER/FDA

Jeff Carey, GenVec

Keith Carson, Williamsburg BioProcessing Foundation (WBF)

Eric Cornuvaca, Invitrogen
Maria Croyle, University of Texas at Austin
Mark D'Andrea, Selective Genetics
Charles Golightly, Invitrogen
Tony Green, Puresyn
James Huang, Puresyn
Beth Hutchins, Canji and USP
Bernie Huyghe, Selective Genetics
Claude Larose, Q-Biogene
Tony Meager, NIBSC (UK)
Heike Nesbit, Cell Genesys
Nancy Sajjadi, Consultant
Paul Shabram, Canji
Geoff Sharpe, Cobra Therapeutics
Stephanie Simek, CBER/FDA
Vicky Sluzky, Onyx
Dick Sublett, Introgen
Ruth Turner, Genzyme
Gary Vellekamp, Schering Plough Research Institute
David Venables, Covance

Guests:

Phil Gomez, NIH-Vaccine Research Group
Wolf Klump, Targeted Genetics Corp.
Vivien Mautner, University of Birmingham (UK), GTAC
Len Schiff, Charles River Labs

Submitted by Nancy Sajjadi and Beth Hutchins, June-12, 2001