Written Responses

Regulatory

Sponsor Question 1:

Does the agency agree that this non-profit charitable sponsor could be granted an exemption by OTAT from eCTD rules and submit their future IND as non e CTD (PDFs with CTD structure) for the first in human study for this ultra-rare disease, with electronic filing requirements after the first in human study?

FDA Response to Question 1:

You may submit a waiver request for exemption from eCTD requirements. After you have requested and received the IND number from FDA, please send an email to esubprep@cber.fda.gov and formally request the waiver. Once your waiver request has been approved by the esubprep staff, you may submit the IND via DCC email at: cberdcc_emailsub@fda.hhs.gov.

Pharmacology/Toxicology

Sponsor Question 2:

Does the Agency agree that the GLP toxicology study in Sprague Dawley rats, coupled with the ex vivo and in vivo data from various studies, supports the intrathecal dosing of Melpida in patients with SPG50?

FDA Response to Question 2:

Based on the limited information provided in Sections 14.3 (pages 12-15/99), 15.3 (pages 36-50/99) and 17.5 (pages 65-99/99) of your briefing package, we cannot yet agree that the preclinical studies described will be sufficient to support the proposed clinical trial. Please address the following comments in your IND submission.

1. The proposed clinical trial will involve administration of Melpida to pediatric subjects. For clinical investigations associated with more than a minor increase over minimal risk involving children, these risks must be justified by a prospect of direct clinical benefit (PDB) to the children (21 CFR 50 § 50.52). Preclinical data used to support PDB at an optimal range of clinical dose levels should be derived from studies conducted in a biologically relevant animal model that demonstrates improvements in a comprehensive battery of clinically meaningful biochemical, pathophysiological and functional parameters in addition to durability of effect. As a result, we have the following comments regarding your ongoing study #2020-06 (page 43/99; Section 15.3.4),

    a. There is insufficient information in your pre-IND briefing package regarding the *Ap4m1-/-* mouse model to determine its suitability for use in establishing proof-of-concept (POC) and PDB for Melpida. Please provide a comprehensive discussion, with accompanying data, regarding the

biological relevancy of the *Ap4m1-/-* mouse model to the proposed patient population, including: i) life-span of the model, ii) onset and progression of the abnormal phenotype (i.e., biochemical, morphological, functional), iii) the similarities and differences in this model and the human disease (e.g., pathophysiology, biochemistry, functional changes, etc.), and iv) the timing of Melpida administration relative to the disease progression in the proposed patient population.

b. In addition to the proposed behavioral outcomes, we recommend that you conduct assessments of a more comprehensive set of disease parameters in this animal model, including survival and relevant biochemical and pathophysiological changes, over time. A comprehensive rationale should be provided for any parameters that are not evaluated.

c. Please provide justification for evaluating only two dose levels of Melpida in this study.

2. Recent published data indicates the potential for AAV-mediated toxicity in the dorsal root ganglion (DRG) and peripheral nerves in non-human primates following intrathecal administration (e.g., Hordeaux et al, 2020). At this time, it is unclear whether rodents are sufficiently sensitive to adequately characterize these toxicities. As a result, given the nature of your clinical product, route of administration, and target population, we recommend that you comprehensively evaluate the potential for vector biodistribution and toxicity in the DRG, spinal cord, and peripheral nerves of NHPs in addition to your ongoing GLP rat study (Study #CRL555008). We have the following additional details regarding the design of this study:

    a. The NHP study should use the intended clinical delivery device, administration procedure and dose levels that bracket the range of proposed clinical dose levels.

    b. Please incorporate a comprehensive battery of safety assessments including both neuropathological and functional assessments (i.e., neurological examinations, electrophysiological assessments, etc.) at multiple timepoints over a study duration that enables characterization of the onset, progression and potential recovery from any toxicities.

    c. Please provide justification if this study is not completed for your initial IND submission and include a comprehensive discussion of the benefit/risk profile for administration of Melpida in your proposed study subjects in the absence of this data.

3. With regards to Study #05 (page 44; Section 15.3.1), you indicate that a number of 7-week-old WT C57Bl/6J mice receiving Melpida developed elevated liver enzyme levels and hepatocellular adenomas at 12 months. To address the risk of

AAV vector integration which has been reported to cause tumor formation in mice (e.g., Li et al, 2020), please provide the following:

a. Comprehensive data regarding the incidence of tumors in vehicle control and low and high dose level groups and the time points at which they were observed.

b. Data from analyses conducted to assess the causative nature of tumors from archived tissue such as integration analysis and histopathological assessments of tumor and surrounding normal tissues.

c. A comprehensive discussion, supported by data, regarding the risk of AAV integration and oncogenesis for your clinical product to include any risk factors inherent to your target population that may impact potential tumorgenicity.

d. Based on data collected from Study #05, we recommend that you comprehensively evaluate the potential for AAV-mediated tumor formation post-administration in Study #2020-06 (page 43/99; Section 15.3.4) and consider extending the study duration for a group of animals, as feasible per the life-span of the *Ap4m1-/-* mouse model, to enable further characterization of this risk. This data should be accompanied by a discussion regarding any inherent pathologies in this mouse model that may impact tumor formation and/or study readouts.

4. We have the following general comments regarding your preclinical development program.

a. Regarding the preclinical vector lots used in your pivotal studies:

i. Please ensure the same assay is used to determine the concentrations of your preclinical and clinical vector lots and provide detailed information on the assays and standards that were used.

ii. Please retain adequate material from each preclinical lot so that it can be retested if the assays for future clinical vector lots change. The vector dose levels administered in these preclinical studies should be recalculated based on this analysis.

iii. Please provide a tabulated summary of the similarities and differences between preclinical and clinical vector lots, including vector identity and composition (e.g., capsid, regulatory elements, transgene, etc.), vector titer, proportion of empty to full capsids, presence of aggregates, formulation, production site, and overall manufacturing process. Please note that the dilution buffer,

container, and delivery device should reflect what will be used clinically, as feasible.

b. Please provide data supporting the reproducibility and accuracy of vector delivery using the respective delivery device and administration procedure for each pivotal preclinical study. If vector loss is observed, please provide the actual administered vector dose level in the study report and data tables.

c. Please ensure that the technical personnel tasked with dosing the animals are appropriately trained. All instances of suspected mis-dosing should be documented in the raw data and included in the final study report.

d. For behavioral assessments, please provide a detailed methodology for each test to verify the objective and stringent nature (i.e., masked assessors, appropriate controls, etc.) of the testing procedure and resulting data interpretation

e. Please ensure that you retain comprehensive set of tissues from your definitive studies and archive all unused tissues for possible future analysis, as feasible.

f. For all unscheduled deaths, please perform comprehensive clinical pathology, gross pathology and histopathology on a complete list of tissues, and other analyses, as appropriate, in order to determine the cause of death.

g. Please ensure that all attempts are made to minimize potential study bias, including: i) inclusion of appropriate control groups; ii) randomized assignment of animals to study groups; iii) appropriate staggered dosing of animals across groups; and iv) masked assessment of selected in-life and post-mortem parameters by qualified personnel.

h. Please provide your rationale for the preclinical dose levels evaluated and timing of study assessments and sacrifice timepoints.

i. Please provide a comprehensive justification for the proposed clinical dose levels, dose volumes and route of administration. Please note that these elements should be supported by data from your preclinical studies.

j. Please provide your method of dose level extrapolation from each animal species used to humans. Additionally, please include your rationale, with supporting data, for this method. For example, if CSF volume is used in your dose extrapolation method, please provide a tabulated summary of CSF volumes for all neonatal, juvenile and adult animals used in your preclinical POC and safety studies in addition to the corresponding values

in human pediatric subjects. Please provide the dose levels of Melpida using the appropriate units (e.g., vg/mL of CSF) to allow for comparison across species in your IND.

Clinical

Sponsor Question 3:

Does the Agency agree:

3a: *with the general design and enrollment criteria of the Phase I/II trial, the dose strategy and route of administration as summarized in Section 14.4.2?*

FDA Response to Question 3a:

We have the following comments intended to better protect the safety of subjects and the design of the proposed open-label, single-arm study:

a. For a rare condition such as spastic paraplegia type 50 (SPG50) caused by the AP4M1 gene mutation with heterogenous manifestations, the more efficient clinical development path is to conduct a randomized clinical trial with a concurrent control group and blinding as early as possible, even in the first-in-human trial. A concurrent control group with appropriate blinding:

   i. facilitates interpretation of safety data and provides a comparator for assessments of safety, activity and efficacy. This will also help you to better plan for a more robust late phase trial, including more appropriate sample size estimation.

   ii. may speed development of your product, by potentially enabling results from earlier phase studies to provide supportive evidence of effectiveness in support of a future marketing application.

   iii. maximizes the use of valuable patient resources.

   An adequately designed and well-controlled early phase study has the potential, depending on the study results, to provide evidence of effectiveness to support a marketing application. For additional information, please refer to our recently published draft Guidance: Human Gene Therapy for Neurodegenerative Diseases (https://www.fda.gov/media/144886/download). We therefore recommend that you modify your protocol to incorporate the following elements: randomization of study subjects, inclusion of a concurrent sham-procedure control group (e.g., performing a sham lumbar puncture without penetration of the dura and using placebo instead immunosuppression drugs, to help maintain adequate blinding of treatment group assignment), and blinding of subjects and evaluators.

b. You plan to measure baseline neutralizing AAV9 antibody titers at your study screening. However, it is not clear from the meeting package whether you will use this result as a patient selection criterion. Please specify whether patients with pre-existing AAV9 antibodies, including neutralizing antibodies, will be eligible and provide the rationale for either including or excluding such patients.

c. You plan to enroll subjects with "Clinical history or examination features consistent with SPG50 and that include neurologic dysfunction." Please clarify the term "neurologic dysfunction." Please clarify whether you plan to enroll subjects with lower limb spasticity at baseline.

d. You plan to evaluate only one dose level. To increase the likelihood of identifying a safe and efficacious dose, we recommend more substantial dose exploration.

e. You state that "Stopping criteria are based on development of unacceptable toxicity defined as the occurrence two or more Grade 3 (Common Terminology Criteria for Adverse Events (CTCAE) Version 5.0) or higher unanticipated treatment-related toxicities." To limit the number of subjects being exposed to unknown but potentially significant risks, please revise the stopping criteria to occurrence of any Grade 3 or higher unanticipated toxicities independent of attribution.

f. You plan to administer several immunosuppressive drugs, including sirolimus, corticosteroid and tacrolimus before and/or after product administration. To maintain a favorable benefit-risk profile, please provide your justification with sufficient data to support the proposed dose and dosing regimen and treatment duration for each immunosuppressive drug.

g. You plan to perform multiple lumbar punctures (LPs) to obtain cerebrospinal fluid (CSF) during the 2-year follow-up period. To minimize the risks to the subjects, please decrease the number of LPs. Please justify with data that each of the LPs post product administration is essential and unavoidable.

3b: *that since the ratio of central nervous system mass to whole body mass changes with age, the dose will be scaled by age correlated to brain mass and not body weight based. Does OTAT agree with both the rationale for dosing based on age (≥ 4 years old), or by approximate brain size for subjects < 4 years old (Table 6)?*

FDA Response to Question 3b:
The overall dosing strategy seems reasonable. In the IND submission, please provide sufficient data to support your proposed brain volume-based dose in children younger than 4 years of age and a fixed dose for subjects 4 years of age and older.

3c: *that SPG50 is an infant onset progressive disease, associated with severe morbidities and caused by biallelic mutations in AP4M1. The primary outcome for the proposed clinical study is safety, however, exploratory efficacy outcomes will be collected. Does the Agency agree with the proposed safety and exploratory efficacy outcome measures, based on the known cause and natural history of the disease (Section14.4.2)?*

FDA Response to Question 3c:
We have no objection to the proposed safety and exploratory efficacy outcome measures. In addition, we recommend that you also assess Clinical Global Impression of Overall Change by Physician (CGI).

Additional FDA Questions/Comments:

CMC

1. Please note that as the IND sponsor you are responsible for providing all the CMC information necessary to assess product safety for the planned Phase 1/2 trial (either as part of the original submission or via a cross-referenced file). Please refer to the Guidance for Industry: Chemistry, Manufacturing, and Controls (CMC) Information for Human Gene Therapy Investigational New Drug (IND) Applications, January 2020, for our comprehensive recommendations. https://www.fda.gov/regulatory-information/search-fda-guidance-documents/chemistry-manufacturing-and-control-cmc-information-human-gene-therapy-investigational-new-drug

2. Please be aware that if you intend to reference CMC information in your IND that was previously submitted to the Agency under another IND or BB-Master File (MF), then you should clearly specify (preferably in a tabular format) the information to be referenced, including the nature of the information (e.g., reagents, testing, manufacturing, etc,), file name, reference number, eCTD module, and page number where the information can be found. This information should also be clearly stated in the letter of authorization (LoA) provided by the cross-referenced IND sponsor or MF holder.

3. Regarding the ddPCR assay that will be used to measure vector genome titer (vector strength) of your product, please note the following:

    a. The assay used to determine vector/product strength (vector genomes/ml) must be qualified prior to Phase I clinical studies. Failure to submit adequate information supporting assay suitability will result in your IND being placed on clinical hold.

    b. Please be aware that the qualification data should be collected for the product under study, AAV9-AP4M1, and should include appropriate product-specific controls.

c. In the IND, please provide a detailed protocol for the qualification study or the SOP used to qualify your assay, including information about the reference standards, controls, and assay optimization.

d. Please provide the study report with data documenting assay qualification, including accuracy, precision (repeatability and intra-assay precision), specificity, range, and linearity. We recommend that the precision of the assay be <15% coefficient of variation (CV).

e. Please describe any deviations that occurred during the qualification study.

f. Please plan to validate the assay prior to the conduct of clinical studies that will assess product efficacy for licensure.

4. Please provide your plan to ensure that the cumulative endotoxin exposure of the pediatric subjects planned in the trial will not exceed the 0.2 EU/kg/hr (USP<85>). In the description of your plan please take into account the following when calculating the potential maximum endotoxin exposure:

   a. The endotoxin levels in the clinical batch of the product

   b. The acceptance criterion/limits set for endotoxin levels in the testing plan for DP lot release

   c. The endotoxin from the delivery devices planned for product administration

   d. The maximum delivery time allowed, and the maximum product volume allowed

   e. The minimum and maximum weight expected of subjects enrolled in the highest dose cohort and the weight range expected of subjects enrolled in all cohorts

   f. Whether a contrast agent will be used at the time of product delivery or soon after within the hour, and the endotoxin levels expected from the contrast agent

   g. Whether a diluent will be used for product formulation in the pharmacy, and the endotoxin levels expected from the diluent

   h. The acceptance criterion/limits set for endotoxin levels in the testing plan for diluent lot release

5. You propose to administer the drug product intrathecally using a Pajunk Atraumatic Sprotte Needle, 60” Marquette Medical IV extension tubing, Braun Discofix 4-way stopcock, 20 mL BD syringe, and an infusion pump. To ensure the devices are being used safely in the context of your proposed clinical study, please provide the information below in your future IND for all delivery devices that will be used to administer the drug product in your proposed clinical study.

    a. Please indicate whether the devices will be supplied by the sponsor or clinical site.
    b. For each device that is FDA-cleared or -approved, please provide the following:
        i. The submission number (e.g., 510(k) or PMA number).
        ii. A comparison of the cleared or approved indications for use and how the devices will be used in the clinical study, as well as a risk assessment for the proposed use in the clinical study.
        iii. Define the essential performance criteria of the device constituents required for the safe use of the device in the context of the IND. Please determine if the defined essential performance criteria are within the cleared or approved indications and specifications and provide performance testing to verify the essential performance criteria if the devices are being used outside of the cleared indication or environment of use.
        iv. If you wish to leverage data from the 510(k) or PMA submission, please provide a letter of authorization for cross reference to that submission.

    c. For each device that is not previously cleared or approved, please provide the following:
        i. A detailed description of the delivery device, including, but not limited to: a description of each component and any accessories that will be used with the device; the manufacturer and trade name; the principle of operation; pictures, diagrams, or engineering drawings; materials of construction; and identification of directly and indirectly (e.g., via fluid path) patient-contacting components.
        ii. Information to establish safety of the delivery device for the proposed clinical use, including but not limited to biocompatibility, sterility, endotoxin, packaging, shelf life, electrical safety (if applicable), electromagnetic compatibility (if applicable), software (if applicable), essential performance requirements (EPRs), and performance testing demonstrating that the device will accurately deliver the drug to the target site within acceptable limits, and identification of how the device may cause harm or may fail to accurately deliver drug during clinical study. Please note that examples of infusion pump EPRs include but are not be limited to delivery accuracy and/or consistency, bolus dose accuracy (if applicable), and delivery status feedback (e.g., visual, audio, or

tactile feedback for delivery start, delivery progress, unintended stoppage, and delivery complete).

iii. We recommend that you provide the information in c.ii above in the form of a tabulated risk analysis with references (hyperlinked) to corresponding test reports or other supporting information or test reports provided in your submission or cross referenced elsewhere, as applicable.

iv. If you intend to cross reference a device master file regarding any of this information, please provide a letter of authorization for the master file with the specific location of the information being referenced within the master file.

v. Please also refer to the following guidance documents for additional information:

1. Use of International Standard ISO 10993-1, "Biological evaluation of medical devices - Part 1: Evaluation and testing within a risk management process" (https://www.fda.gov/regulatory-information/search-fda-guidance-documents/use-international-standard-iso-10993-1-biological-evaluation-medical-devices-part-1-evaluation-and).
2. Infusion Pumps Total Product Lifecycle (https://www.fda.gov/regulatory-information/search-fda-guidance-documents/infusion-pumps-total-product-life-cycle).
3. Recommended Content and Format of Non-Clinical Bench Performance Testing Information in Premarket Submissions (https://www.fda.gov/regulatory-information/search-fda-guidance-documents/recommended-content-and-format-non-clinical-bench-performance-testing-information-premarket).

d. We recommend that the clinical study protocol include requirements to capture any delivery device failures/malfunctions.

6. The drug product, Melpida, is intended to treat Spastic Paraplegia Type 50, which is a rare disease. As discussed in the draft FDA guidance document "Human Gene Therapy for Rare Diseases," from January 2020 (https://www.fda.gov/regulatory-information/search-fda-guidance-documents/human-gene-therapy-rare-diseases), an adequately designed and well-controlled early phase study has the potential, depending on the study results, to provide evidence of effectiveness to support a marketing application. As the devices used to administer the drug product are an important factor in demonstrating the safety and effectiveness of your investigational treatment, we strongly recommend that your clinical studies utilize the delivery devices you would intend to be used with the drug product upon licensing. Furthermore, we recommend that you consider your licensing and labeling strategy as it relates to the devices used to administer the drug product early in your product

development to ensure that appropriate devices would be available (i.e. FDA-cleared or approved) to deliver the licensed product. Additional information may be needed depending on your proposed delivery devices and marketing strategy, and you may need to work with a device manufacturer(s) to ensure the feasibility of this approach. We recommend communication with OTAT early in product development regarding your delivery device strategy.

7. Please plan to conduct device compatibility studies to support vector stability in the delivery devices. Please note that failure to submit sufficient information supporting vector stability in the delivery device will result in your IND being placed on clinical hold. In the IND, you should describe the dose preparation (in the pharmacy) and provide the report of the delivery device compatibility study performed to simulate the procedure for dose preparation and product administration at the clinical site. Please note the following recommendations for the conduct of the study:

    a. Please ensure that the product lot used in the compatibility study is manufactured and formulated in a manner comparable to the clinical lot(s). The supporting manufacturing, qualification and testing information for the product lot used in the compatibility study should be submitted in the IND.

    b. Please assess the amount (vector genomes) and activity (infectious units or potency) of the product following exposure to the clinical delivery device.

    c. Please be aware that the study should include tests conducted over the planned dose-range and should take into account the expected time between thaw of the product and infusion.

    d. Please perform device compatibility testing for the product under conditions that mimic the clinical scenario (i.e., hold time, formulation/concentration, temperature, presence of contrast agent, etc.); the study design should consider the worst-case scenario (e.g., low product concentration, maximum hold time).

    e. The device compatibility data should support the post-thaw product handling instructions provided in the Instruction to Pharmacy/Pharmacy Manual document that is supplied with the product to all the clinical sites.

8. You should describe how the drug product will be shipped from the site of manufacturing to clinical sites in the US. You should conduct appropriate shipping study (using representative material) in order to evaluate the impact of shipping on product quality (product purity, sterility and potency). Accordingly, please plan to develop mitigations plans for temperature excursion. Please note that the shipping study should be qualified to support late phase studies and validated by licensure.

9. If a diluent will be manufactured to support the preparation of the final dose in the clinic, please describe the manufacturing and release testing plan for the diluent (i.e., specifications for diluent release). This information should be documented under Section 3.2.P-Diluent, separate from the information for the DP (3.2P-Vector). Also, please ensure that the diluent meets the requirements for subvisible particulates per USP <787>. The IND should also include information on how you will monitor the stability of the diluent during storage and shipping.

Pharmacology/Toxicology

1. Statements regarding the adequacy of any preclinical study to support a particular clinical trial or fulfill a specific regulatory requirement are made based solely on the information provided in your pre-IND meeting package and are considered preliminary. A final determination regarding the adequacy of the studies cannot be made without CBER review of complete materials that should be submitted in the IND.

2. In your IND submission, please provide complete study reports for all preclinical studies used to support the safety and rationale of your proposed clinical trial. These reports should include, but should not be limited to: a) a prospectively written protocol and all protocol amendments or a detailed methodology; b) a detailed description of the study design (e.g., description of the test system used, animal species/animal models, control and test articles administered, dose levels, detailed procedures for test article administration (including delivery device description), and collection of all study protocol parameters, etc.); c) results for all parameters evaluated for each animal on study; and d) your analysis and interpretation of the study data.

3. For each toxicology study performed, please provide documentation showing that the study was conducted in compliance with Good Laboratory Practice (GLP) as per 21 CFR Part 58. If the study was not GLP-compliant, as directed by 21 CFR Part 314.50(d)(2)(v), you should provide a brief statement of the reason for the non-compliance in your IND submission. In addition, please specify in the study report any areas that deviate from the prospectively written protocol and the potential impact of these deviations on study integrity. Each study should: a) be conducted according to a prospectively written protocol, b) performed in as nonbiased a manner as possible, and c) have appropriate record keeping and documentation of all data.

4. We strongly recommend oversight of the conduct of all non-GLP toxicology studies and each resulting final study report by a Quality Assurance (QA) unit/person that is independent of the personnel responsible for the conduct of this study, as per 21 CFR Part 58.35. This QA oversight is important to

assure study conduct according to sound procedures and to ensure the quality and integrity of the resulting data.

5. In Module 4 of your IND, please provide a copy of all key publications cited that support the safety and rationale for administration of your investigational product in the proposed clinical trial. In Module 2 of your IND, please include a comprehensive summary for each publication. The summary should provide the reason for including the publication (i.e., how it directly supports safety/activity of your product) and a discussion regarding the comparability of the product(s) used in the publication to the final clinical product.

6. Please ensure that you have adequately addressed all CBER pre-IND comments and include these responses in the IND submission.

7. Please provide an Investigator Brochure (IB) in the IND submission. For additional recommendations on the preparation and content of your IB, please refer to Section 7 of the document titled, *E6(R2) Good Clinical Practice: Integrated Addendum to ICH E6(R1) - Guidance for Industry* (March 2018), available at: https://www.fda.gov/regulatory-information/search-fda-guidance-documents/e6r2-good-clinical-practice-integrated-addendum-ich-e6r1.

8. For a comprehensive summary regarding the preclinical assessment of cell and gene therapy products, we refer you to: a) the document titled, *Guidance for Industry: Preclinical Assessment of Investigational Cellular and Gene Therapy Products* (November 2013), available at: https://www.fda.gov/regulatory-information/search-fda-guidance-documents/preclinical-assessment-investigational-cellular-and-gene-therapy-products; and b) the *OTAT Learn* Webinar Series, available at: http://www.fda.gov/BiologicsBloodVaccines/NewsEvents/ucm232821.htm.

9. Regarding biodistribution (BD) assessment for gene therapy products, we recommend reading the 2018 International Pharmaceutical Regulators Programme (IPRP) reflection paper titled, *Expectations for Biodistribution (BD) Assessments for Gene Therapy (GT) Products*, available at: https://admin.iprp.global/sites/default/files/2018-09/IPRP_GTWG_ReflectionPaper_BD_Final_2018_0713.pdf.

10. The preclinical program for any investigational product should be individualized with respect to scope, complexity, and overall design, to maximize the contribution and predictive value of the resulting data for clinical safety and therapeutic activity. As recommended in Section III.B.8 of the *Guidance for Industry: Preclinical Assessment of Investigational Cellular and Gene Therapy Products*, we encourage you to explore opportunities for reducing, refining, and replacing animal use in your preclinical program. For example, it may be appropriate to use *in vitro* or *in silico* testing to

complement or replace animal studies. We encourage you to submit proposals and justify any potential alternative approaches.