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Dear CIRM ICOC Committee,

We are submitting this letter to be part of the public record as a response to the review of our grant proposal: TRAN1-12919 on which I am the lead PI entitled "**Pre-Clinical Development of Gene Corrected Autologous Ariway Stem Cell Therapy to Treat Cystic Fibrosis."**

We are providing a rebuttal to the review of our TRAN grant (TRAN1-12919) developing a genome edited autologous ***stem cell*** based therapy for cystic fibrosis (CF) (we believe that this still may be the only CF focused project in the CIRM portfolio). We were pleased that in every domain except for one, the proposal was considered strongly meritorious. We are proud that our team has discovered that CF is more prevalent in underserved populations than previously recognized and that this underserved population has a mutation spectrum in which newly developed triple modulator therapy will be ineffective (described in the proposal and manuscript submitted). This discovery further increases the significance of our proposal as it is an approach to address this large unmet medical need in an underserved population. We note that CF affects patients regardless of gender, with evidence that it can be more severed in women, and our approach would be applicable to every patient.

We are extremely disappointed, however, in the assessment of the research plan. The review team seemed to bring a biased and pre-conceived notion of what we should do and did not seem to have made an assessment of the rationale and logic of the proposal itself. We are disappointed that we wrote the revision in a way in which the review team seemed to take personal offense that we did not adopt all of their prior suggestions into the revised proposal (since we actually incorporated almost all of their feedback into the revision). An example of the bias is demonstrated in one reviewers' comment that the lung is the most important tissue without acknowledging that we wrote that we agree(!) but that to get to the lung, we needed first test the approach in the sinus for safety reasons. Another example is that a reviewer suggested we have an INTERACT meeting when a key important milestone in both our original and revised proposal was that we have an INTERACT meeting.

We had thought deeply about the issues raised in the prior submission and provided what we thought was a careful response for why the proposed experiments were not the right path for our project. Clearly, from a grantsmanship view, we should have simply outlined experiments that addressed the reviewer's points. Instead, we put deep scientific thought into the plan and concluded that we could not propose experiments and milestones that we did not believe were necessary nor feasible. The review team in their comments did not show any indication that they took our arguments with any serious consideration. In sum, we fundamentally disagree about the utility of the rodent model the review team seems to insist on. In fact, the points raised by the review team in this regard are scientifically fatally flawed and would take us down a developmental path destined for failure. We outline these flaws briefly below:

- The review team states that seeing improvements in nasal potential differences in rodent models was seen in prior gene therapy approaches. These experiments were feasible because the reagents were treating rodent cells in a natural rodent CFTR mutated environment. Our approach, because the reagents are human specific, will not work across species boundaries and so cannot be used in the way that other approaches gene therapy approaches can be used across species. We note that the efficacy in rodent models did not translate to humans and is another example of the now well-known fact that rodent models are an extremely poor predictor of both efficacy and toxicity in humans—in gene therapy and in drug development in general.
- The second fatal flaw is the review team's insistence on in vivo function rather than engraftment as necessary for pre-clinical development. As the CIRM team knows well, in the hematopoietic system for example, genetically engineered human HSPCs are transplanted into immunodeficient mice and it is the engraftment of the cells including differentiation into some (but not all of the lineages) that is considered a metric of success, not whether the human blood cells actually function correctly. In the xenogeneic setting, it is quite reasonable to think that the human cells would not function correctly because of the tremendously different environment both in the blood and the airway. Thus, if we saw successful engraftment and differentiation but not function, it would not be a deal breaker for subsequent testing in a phase I/II clinical trial where function in the autologous, non-xenogeneic setting could be evaluated (along with safety).
- The third fatal flaw is the review team's assumption that wild type human cells would create wild-type CFTR function after transplantation in a xenogeneic setting. There is no evidence that this would occur.
- Finally, the fourth, and most important, fatal flaw, is that the review team's proposed rodent functional experiments are not even technically feasible. If we put the human cells into an immunodeficient but CFTR wild-type respiratory epithelia, the conductance will be from the residual wild-type endogenous rodent cells. If we put the human cells into a CFTR knockout rodent model, the immunocompetent rodent will simply reject the human cells. There is no CFTR mutant immunodeficient animal model to use. It is likely that such a model is not even possible to generate because immunodeficiency may not be compatible with life in a CF background. The review team should explicitly state that they recommend we put the entire project on hold for several years while we attempt to generate this model. Which in the end may not even be of benefit because of fatal flaws 2 and 3 described above. Thus, in a milestone-based grants that CIRM funds, it would be an enormous mistake for our team to propose a set of experiments which has no chance of technical success to satisfy a review team.

In our response to the original submission, we thought we had provided a clear description for why the proposed experiments from the first submission were not the best path for our project. We now recognize that we should have been much more explicit in our rationale for the proposed experiments and the reasons that assessing function of human cells in a rodent model is a fatally flawed approach.

Our approach is highly innovative and we are blazing a path (with machete's sharpened and swinging hard) that nobody has done before in developing a genetically engineered stem cell therapy for the respiratory epithelium and treatment of cystic fibrosis. In contrast to genetically engineered hematopoietic stem cell-based therapies or gene therapies with AAVs or LNPs in which the developmental path is clear and hundreds have walked it. We are aware that there is renewed excitement for re-visiting previously failed gene therapy approaches for CF with the hope that the modern reagents will overcome the reasons for prior failures. It is not surprising that there would be disagreements about our innovative strategy—nobody really knows how to do this. We share the reviewers desire for experiments that could more fully test

efficacy beyond engraftment (which as noted above, however, is all that is asked of HSC based therapies). After extensive discussions and hours upon hours of consideration, however, we do not believe it is possible (or even necessary for the FDA to give a "may proceed" to an IND). We ask CIRM to recognize that our team is absolutely committed to developing this approach so it will be safe and effective in patients and have no interest in taking shortcuts. And we ask CIRM to give our multi-disciplinary team of experts the benefit that nobody in the world has spent more time thinking about how to achieve that goal and that our TRAN proposal is the result of that work and expertise.

Finally, we note that the ultimate arbiter of what is needed to initiate a Phase I/II clinical trial of this innovative stem cell-based therapy for cystic fibrosis is the FDA. In our proposal, we integrated an INTERACT meeting to review the toxicology/tumorigenicity study into our milestones and work flow. We hope that CIRM would agree to fund the project at this point by making a simple change to the INTERACT milestone. In the revised milestone we would use the INTERACT meeting to get feedback from the FDA on both the design of a toxicology/tumorigenicity study *AND* of the utility of demonstrating in vivo functional correction using human cells in a rodent model. In this way, the team can get formal (albeit non-binding) feedback on the necessity of the rodent model from the regulators and the disagreement between our team and the review team would be resolved.

Sincerely,

Matthew Porteus MD, PhD
Sutardja Chuk Professor of Definitive and Curative Medicine