City of Hope™
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To: CIRM Independent Citizens' Oversight Committee
c/o: Maria Bonneville
Re: CIRM CLIN2-12153 Application, Leo D. Wang, MD, Ph.D., PI

We would like to thank CIRM for the opportunity to apply for CLIN2 funding to support our important and innovative clinical trial, and thank the Grants Working Group for their thoughtful and detailed review of our proposal. We also thank CIRM for the chance to address the ICOC concerning our application, and hope that you will agree that the proposed clinical trial presents a unique and important opportunity to advance pediatric cancer immunotherapy.

Our trial would be the first pediatric cancer-focused trial funded by CIRM. One in 265 children in California will develop cancer by age 20, and CIRM has an opportunity to demonstrate leadership in funding pediatric oncology translational research by supporting our application. Brain tumors are the most common solid tumor of childhood, accounting for about 20% of all pediatric cancer cases but killing more children than any other malignancy. Aggressive pediatric brain tumors have a dismal prognosis, and cure rates for these diseases have not improved much in decades. New and better treatments are desperately needed for these diseases. Fortunately, City of Hope has developed innovative and exciting chimeric antigen receptor (CAR) therapies for brain tumors, and recently were the first to demonstrate that CAR T cell therapy could cause regression of a metastatic brain tumor in an adult patient. We have a long track record of success with CAR T therapy, having successfully manufactured CAR T cells for over 240 patients across 18 institution-initiated trials to date. Our infrastructure and manufacturing capacity are unrivalled anywhere in the state, or indeed in the country. We are therefore uniquely positioned to bring this novel therapy to the pediatric population that so desperately needs it.

Virtually all reviewers agreed that our proposal has **significance and potential for impact** (13/14), **sound rationale** (11/14), and is **feasible** (12/14). Most reviewers also agreed that it was **well-planned and designed** (8/14). We have taken the liberty of responding to the specific reviewer comments below in a point-by-point fashion. We agree that their concerns are important, and indeed have already addressed most of them. However, due to space constraints, we could not provide exhaustive data in the main application. We have provided substantially more supporting evidence here, and hope that you will agree that this satisfactorily addresses reviewer concerns.

Reviewer Comments (confidential):

1) *More preclinical data in a immunocompetent murine model demonstrating effects of lymphodepletion would enhance the rationale.*

We are happy to provide additional preclinical data supporting the addition of lymphodepletion to intraventricular delivery of brain-tumor-targeted CAR T cells, which we could not include in the initial application. We did show Kaplan-Meier curves demonstrating significant increases (~ doubled, p=0.01) in median survival time in mice receiving CAR T therapy after lymphodepletion with radiation or chemotherapy (reproduced as Fig. 1). Additionally, we provide data (Fig. 2, below) showing that lymphodepletion improves tumor control by intraventricularly-delivered CAR T cells; mice receiving lymphodepletion (right) prior to CAR therapy have a lower tumor burden than non-lymphodepleted mice (left) as measured

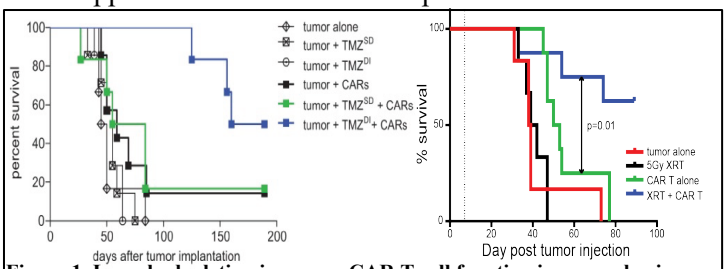


Figure 1. Lymphodepletion improves CAR T cell function in mouse brain tumor models. Kaplan-Meier curves of mice inoculated with glioblastoma and treated with CAR T cells, +/- lymphodepletion. **A)** Mice treated with lymphodepleting (DI, dose intense) temozolomide (TMZ) followed by CAR T cells had superior survival (blue line). SD, standard nonlymphodepleting dose. (From Suryadevara 2018). **B)** Mice treated with 5Gy RT followed by IL13BBζ CAR T cells (blue line) had superior survival to controls.

by photonic flux using luciferase-expressing brain tumor cells. Importantly, irradiation alone does not affect tumor growth (Figure 1B, black line).

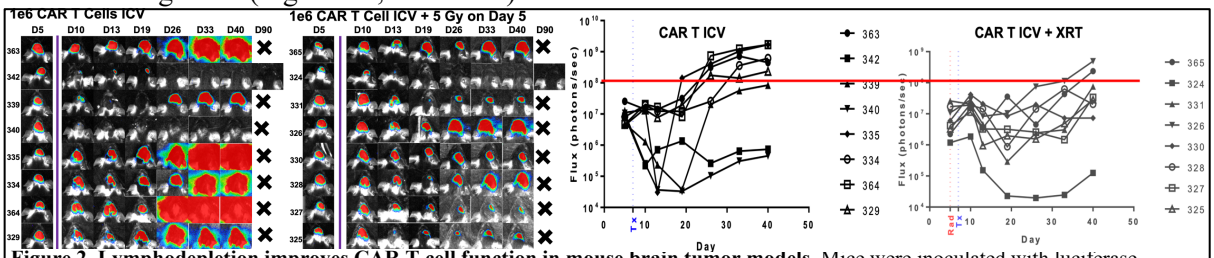


Figure 2. Lymphodepletion improves CAR T cell function in mouse brain tumor models. Mice were inoculated with luciferase-expressing glioblastoma and treated with CAR T cells, +/- lymphodepletion. **A)** Photonic flux imaging of recipient mice shows substantial increase in disease burden in nonlymphodepleted mice (left) as compared to mice receiving lymphodepletion (right). **B)** Quantitation of photon flux demonstrates differential tumor growth over time (compare left and right graphs; red line denotes threshold flux of 10^8 photons/sec for illustrative purposes). Experiment shown is representative of three independent replicates.

2) *Some clarity regarding IL13RA testing; is this a CLIA certified test? How long between results of test and initiation of trial?*

There are no CLIA-certified tests currently available for this antigen. However, we have used our institutional test for years with a high degree of consistency and reproducibility. Patients can be enrolled on the trial as soon as they screen in (functionally speaking, this is as soon as the IL13RA2 testing is complete). Time from trial enrollment to administration of cells is about 4 weeks.

3) *There are some concerns about enrollment at a single institution. PNOC is listed as a consortia to enroll though phase II but not clear how this will be done and how efficacy will be measured across so many different diseases.*

We have already had numerous requests from patients and providers to evaluate candidates for this trial; this week alone, I have evaluated three patients. Against a total accrual of 18 patients over 3 years, we are highly optimistic that this will feasible. However, we are also planning to expand this trial to a second site at CHLA. This expansion is outside the scope of CIRM funding, but we would certainly welcome the opportunity to apply for additional funding to facilitate this process.

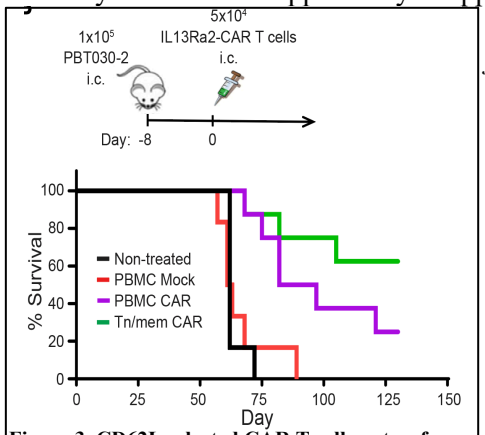


Figure 3. CD62L selected CAR T cells outperform unselected CAR T cells. Treatment of mice inoculated with brain tumor cells with PBMC-generated CAR T cells (purple) improved median survival (median survival 83 days), but treatment with CD62L-selected CAR T cells (green) was superior (median survival undefined). Survival curve shown is representative; experiment was repeated three times.

4) First, does the selection of CD62L-expressing cells from the apheresis of these patients address low(er) potency of CAR-engineered T cells in patients with brain cancers? In other words, do you have potency data to indicate that at least the memory function of these cells outperforms unselected T cells from the same patient.

We provide data (Fig. 3, left) demonstrating that CD62L-enriched (Tn/mem, green line) CAR T cells outperform unselected CAR T cells manufactured from peripheral blood mononuclear cells (PBMCs, purple line) in preclinical models.

5) Second, what is the manufacturing feasibility of the proposed process, knowing a) that the selection marker, CD62L, is not well preserved by cryopreservation, and b) will you be able to get sufficient cell numbers to start manufacturing and meet dose?

We have used this exact process successfully to generate over 165 clinical CAR T products thus far, and over 300 clinical products using similar strategies that rely on CD62L enrichment (see Table I below). Because CD62L selection is performed on fresh PBMCs, loss of

Table I. CAR T products manufactured at City of Hope since 2011

Year	CD19 NHL	CD19 NHL	CD19 NHL	CD19 BCL	CD19 ALL	IL13 GBM	CD123 AML	HER2 GBM	HER2 BM	CS1 MM	PSCA Prostate	IL13 Nivo GBM	CLTX GBM	IL13 Mel	Total
2011	1														
2012	7														
2013	3	2													
2014		8	7	4	2										
2015			13/2	8	2/5	9	1								
2016			9	1	6	20	7								
2017			1		10	29/2	9								
2018				10	11	6/25	12	6	1						
2019				12	8	1/8	14	8/6	3/11	2	6	3			
2020				3	10	1	3	13	4	2	5	4	3	1	
Tcm	11	10	20	12	4	65		14	4						136
Tnmem			12	26	45	36		19	15	4		7	3	1	168
PBMC							46				11				57
TOTAL	11	10	32	38	45	101	46	33	19	4	11	7	3	1	361

expression due to freeze/thaw cycles is not a concern. Currently, our manufacturing process requires patients to undergo leukapheresis at City of Hope, so that manufacturing can begin immediately.

Key Questions and Comments:

• Planning and Design (6 No votes):

1) *Inclusion of patients to the age of 25 may not necessarily reflect a true pediatric population and could skew recruitment toward a more adult population.*

We include patients up to 25 because many patients will have seen multiple lines of therapy (and thus might have been diagnosed at 18 but still alive at 21), and we did not want to exclude them. The intent is to study pediatric brain tumors, and we are selecting patients with pediatric histology tumors. We are happy to decrease the age limit to 21.

2) *I did not see proof of cell manufacturing data from blood or apheresis product, so it's unclear they can make the product. I did not see the scheme to make lentiviral vectors, so it is unclear what titer and specifications are attained.*

As stated above (Table I), this is a well-established manufacturing platform that has been in use at City of Hope for many years.

3) *Reviewers' comments about potential bio-distribution, tumor heterogeneity, heterogeneity of target expression and potential inclusion criteria for trial need to be addressed.*

To the extent that reviewer comments referred to here are restated elsewhere in this review, they are addressed there. We would be happy to address any other concerns if CIRM shares them with us.

4) *The clinical rationale is not clear for the pediatric population.*

Aggressive pediatric brain tumors often have a dismal prognosis, and better therapies are badly needed. Testing novel therapies in pediatric populations is both a moral and a clinical imperative, and insisting that new treatments be tested in adults first does an enormous disservice to children with these diseases. Adoptive cellular immunotherapies are known to be effective in children, and we expect that this particular combination of cellular immunotherapy and lymphodepletion deserves to be tested in children.

• Rationale (3 No votes):

1) *They have treated over 60 adult patients. The product has demonstrated safety and some efficacy in adult patients. Yet, the applicants propose to test this product in pediatric patients. There is no clear rationale for moving to pediatric patients with malignant glioma. What is unique about pediatric patients and why do we think this will work in this context compared to the previous clinical experience? I suggest the sponsors start with their lymphodepletion + CAR T combination in the adults and then transition to pediatric patients. Alternatively, I suggest the authors aggregate and compile a more robust pre-clinical package to justify the use in pediatric patients.*

Pediatric and adult patient populations are not comparable, and there is only a limited extent to which studies done in adults are informative in pediatric oncology patients. Indeed, the NIH Policy on Inclusion of Individuals Across the Lifespan as Participants in Research Involving Human Subjects clearly states that "children must be included in…human subjects research" because "children were not [historically] appropriately included in clinical research studies, resulting in insufficient data to establish the effectiveness of treatments in children." There is every reason to advocate for the inclusion of pediatric populations in brain-tumor targeted CAR T cell clinical trials, and excluding them on the basis of the adult experience would make little sense even if the therapy had shown unacceptable toxicity in adult patients. In addition to expanding into pediatric populations, we are also expanding our indications to non-glioblastoma pediatric brain tumors. We and others have shown that up to 50% of pediatric brain tumors express IL13Rα2, and in every preclinical model tested, IL13BBζ CAR T cells eliminate cells that express IL13Rα2^{+}. Thus, we do not believe that there is additional benefit to testing more IL13Rα2^{+} tumor lines in murine models.

2) *Why the transition to pediatric patients, at the same time as the addition of lymphodepletion?*

See above for our justification for expansion into pediatric populations. We have added a 3-patient safety lead-in to test the efficacy of CAR T cells without lymphodepletion in pediatric patients; if CAR T cells alone are safe and well-tolerated, we will add lymphodepletion.

3) *Previous data indicates that IL13 receptors are highly variably expressed in gliomas. It would be important to define inclusion criteria based on expression (e.g., the percent tumor cells that express the receptor, etc.).*

We have adopted our standard inclusion criteria for tissue positivity, which have been used across multiple clinical trials at our institution. This system relies on expert assessment of immunohistochemical staining and assignment of an H-score. Patients whose tumors exhibit an H-score > 60 are eligible for enrollment in the trial.

4) *The variability in tumor types (despite being classified as Grade III or IV) could make drawing potent conclusions from the study difficult.*

This is a Phase I study, and as such is not powered to detect clinical benefit. Rather, it is designed to show that lymphodepletion followed by CAR T cell treatment is safe and well-tolerated in children. If we do observe any clinical or radiographic response, we will propose to open an expansion cohort to focus on the specific disease type in which a potential response has been observed. However, funding for these expansions is outside the scope of the current application.

• **Feasibility (2 No votes):**

1) *It will be difficult to accrue enough patients in a single center trial.*
2) *Not sure if doable at one site.*

These concerns have been addressed above; based on the interest we have received thus far, we project that we will have a waiting list rather than difficulty accruing patients. Additionally, we have plans to expand to Children's Hospital of Los Angeles as a second site in 2021, pending regulatory approval and availability of funding.

• **Significance and Potential for Impact (1 No vote):**

1) *Lack of preclinical data to support the thesis that a lack of naive/central memory T cells is a potential culprit in CAR-redirected patient T cells.*

This is addressed above and in Figure 3.

2) *Pre-clinical data that selected T cells outperform unselected T cells in the target patient population is needed.*

This is addressed above and in Figure 3.

3) *Manufacturing feasibility: Can sufficient selected cells be isolated from apheresis of these patients? There is concern that it will all be CD4 T cells since CD8 T cells are the first to lose their early memory state.*

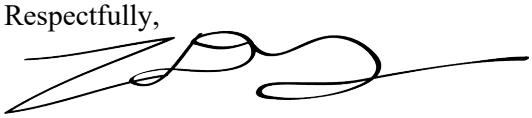
This is addressed above; we have successfully manufactured several hundred products using CD62L-based enrichment strategies at City of Hope.

4) *Correlative studies should include an assessment of the tumor bed prior to (pathology specimens) and post-CAR T cell treatment to a) assess homing and b) tumor microenvironment changes*.

We agree that it would be ideal to assess the tumor bed before and after therapy. Pediatric diagnostic samples are often small, especially of brainstem lesions, but to the extent that we are able to perform comprehensive analyses we propose to do so. Longitudinal tumor bed assessment would require repeat biopsies of patients, which is impractical and may impose undue risk on patients. In situations where repeat sampling is clinically indicated, or in situations where patients succumb to their disease, our consent does allow us to request tissue for more comprehensive analyses.

We thank the Grants Working Group and the Independent Oversight Committee for their support of and advocacy for innovative stem cell translational research in the State of California. We look forward to working with you to develop this important clinical trial, which has the potential to dramatically improve the lives of pediatric cancer patients.

Respectfully,



Leo D. Wang, MD, PhD