

## CHILDHOPE – Chimaeric T cells for the treatment of paediatric cancers

RTD-Project 6th FP, STREP, Contract No. 037381  
Start Date: 01/11/2006 – Duration: 48 months  
Coordinating organisation: Centre Léon Bérard

### **Publishable Executive Summary**

#### ***Objectives of the project***

The CHILDHOPE project builds on the excellence of a network of EU-based partners with a broad experience in the field of pediatric hematology and oncology, immunology and cell & gene therapies. The CHILDHOPE project is unique since it brings from bench to bedside (and back) an innovative technology as yet never applied in children with advanced or refractory hematopoietic malignancies. The CHILDHOPE reverse translational research project will focus on:

- Improving and testing efficacy and the safety of anti-leukemia/lymphoma chimaeric T cells in relevant preclinical models in vitro and in vivo in mice.
- Scaling-up this technology to numbers suitable for a clinical application in children with hematopoietic malignancies.
- Based on biological material obtained from our preclinical models and from children treated with these genetically engineered T cells, dissecting the interface between the host's tumor and immune cells and use this knowledge to understand the mechanisms of anti-tumor action, validate novel targets and diagnostic tools specific to children affected with leukemias or lymphomas.

The originality of the CHILDHOPE project is to exploit the immuno-stimulatory properties of EBV-CTLs and retarget them to leukemia/lymphoma cells, which themselves lack many of the co-stimulatory molecules needed to activate CTLs.

Our pioneering project proposes to administer these chimaeric T cells in hard-to-treat pediatric malignancies. In particular, CHILDHOPE focuses on three pediatric tumors: acute B-lineage lymphoblastic leukemia, non-hodgkin B-lineage lymphoma and acute myeloid leukemias.

Another major innovation is that we will investigate modifications to the chimaeric receptor molecule itself, in efforts to augment its capacity to enhance the transduction of signals that increase cytotoxic and memory effector functions while maintaining a high-level of safety. The T-cell proliferation should be strictly antigen-dependent and dwindle after elimination of the antigen.

Finally, the CHILDHOPE project moves the field forward as it brings to clinic this therapeutic modality in a phase I setting. We are confident that the chosen methods of gene transfer (i.e. retroviral vs. electroporation) in conjunction with a suicide gene system will provide a safe albeit sufficiently prolonged anti-leukemia effect in vivo in children with relapsed/refractory leukemia/lymphoma. We anticipate that the anti-leukemia/lymphoma effects - and consequently the survival of the treated patients - will increase with the chimaeric T-cell doses. In parallel, the CHILDHOPE project will provide preclinical data comparing the cytotoxic potency of different T-cell subsets (EBV-CTLs, CIK cells and GDT cells) in an effort to identify the best immune effector for future clinical application.

The final aim is to develop a safe and efficient adoptive immunotherapy for children with advanced or refractory malignancies.

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### ***Expected results***

Our underlying general hypothesis is that expression of chimaeric anti-leukemia/lymphoma receptors on EBV-specific CTLs will allow these cells to retain their known safety and functionality in vivo (ability to expand and regress in response to antigen load, to persist as memory cells, and to retain antiviral activity) in children with hematopoietic malignancies while adding safety, specificity and effector function directed to the residual leukemia cells.

This strategy seems highly feasible since EBV-specific CTL lines have been generated and reinfused in numerous patients using robust and clinically validated methodologies developed by Brenner, one of our key scientific advisors in the CHILDHOPE project. Unmodified CD19 molecule have been manufactured and incorporated into EBV-CTLs, and tested in vitro.

### ***Potential applications***

No animal model can fully dissect the tremendous complexity of the interactions between a human tumor and its host. The CHILDHOPE project is the first comprehensive attempt at administrating anti-tumor chimaeric T cells in children with hematopoietic malignancies. The CHILDHOPE project represents a unique opportunity to address in vivo in a complete human environment some of the most fundamental hypothesis in the field of anti-tumor immunology. Our hypothesis is that targeting CD19 or CD33 will not only kill target cells but will also contribute to the release of yet unknown tumor-associated antigens (TAAs), which in turn may generate further immune activation. This mechanism known as antigen spreading may be of particular interest in tumors known for their propensity to induce antigen-loss variants. As these new TAAs may be either leukemia/lymphoma-associated or have a broader scope of pediatric tumors, they may provide new targets for future immunotherapeutic approaches. While the CHILDHOPE project focuses at this stage on three of the most frequent pediatric tumors (acute B-lineage lymphoblastic leukemias, non-Hodgkin B-lineage lymphoma and acute myeloid leukemias), our innovative technology should pave the way for the generation of T lymphocytes with an antibody-dictated specificity toward other tumor-associated antigen for which a monoclonal antibody exists. This in turn should allow us to redirect immune effectors towards other malignancies, including solid tumors, for which current therapeutic strategies are limited or have failed. This includes - but is not limited to - metastatic disease where T-cell therapies have had some success due to their capacity to migrate and infiltrate distant tumor sites. In fact, it is likely that only combination of therapies that act at key points of tumor escape pathways may help to eradicate tumor cells and also to lessen the dose-intensity of current chemotherapy regimen, a critical element of any therapeutic approach in developing individuals or in elderly patients who have reached maximum tolerable doses of anticancer drugs when their disease reoccur. Hence, our innovative therapeutic approach may provide hope for a cure for young as well as older patients with advanced or refractory tumors.

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### Project objectives and major achievements during the third reporting period (M25-M36)

**WP1:** we have obtained new data on innovative ways to connect a CAR activation domain to co-stimulatory signals. We are nearly finished fully characterizing the CD34-QBEND10 epitope work. We also present data with co-expression of CAR with iCasp9 suicide gene in peripheral blood T-cells without diminution of CAR expression or loss of killing by CID. The comparison of different suicide genes in EBV-CTLs is progressing. It appears that in EBV-CTLs, bright expression of iCasp9 may lead to cellular toxicity and death. Work with the different families of SIN vectors is progressing but cloning the promoter sequences into the appropriate place after the 3' LTR is still in progress so we have not presented new data for this section. For SIN vectors, we are selecting the best pseudotype for clinical production because recent new data shows that pseudotyping with measles can lead to a vector capable of transducing resting T-cells. Clinical-grade production of RNA is progressing well with most of the practical barriers overcome.

**WP2:** Additional preclinical in vitro and in vivo data have been generated for the clinical study. We have designed and optimized immune monitoring experiments and updated the respective paragraph in the clinical study protocol. The Investigator Brochure has been updated, and both data and documentation for the IMPD have been provided. We have applied for grant funding with the WLBH foundation to cover the study cost for the German and Austrian centers. Another objective consists of obtaining new Sponsor and to prepare documents for submission to regulatory authorities with first submission to AFSSAPS. In that way, UCL has agreed to act as the Sponsor for the CD19 project and provide the necessary insurance following a successful project risk assessment. Documents required for submission are near completion.

**WP3:** Significant results were obtained to compare chimeric CTL, CIK and  $\gamma\delta$  effectors functions. Significant progress were made in demonstrating the immunogenicity of effector-mediated cell death (CTL and NK), elucidating mechanisms of cell lysis using cell lines and identifying monitoring parameters of possible resistance such as expression of CD19 and integrins. Furthermore, as mentioned in the previous report, we have started to set up alternative assays to evaluate immunogenicity of chimeric CTL induced cell-death, using new Her2/Neu targeting CTL or new CD19 expressing cell lines.

**WP4:** Plans for organising a conference for parent's organisations are under preparation and will be carried out between March-April 2010. The conference will be organised by ICCCP, and several CHILDHOPE members will make their contribution as speakers. The scientific level of the presentations has to be adapted to the parents' level of knowledge.

**WP5:** With a total of 30 communications, of which two third during national, European and international conferences, the scientific community and the health professionals comprising clinicians, hematologists, immunotherapy, transplant experts and gene therapy actors have been well informed. Furthermore, interactions with similar or larger consortium are growing and several partners are looking forward for opportunities to renew this experience, by joining

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larger initiative as well as by applying through a similar but larger scale EU funding instrument.

**WP6:** A research is in progress in which we will evaluate the patients concerns and expectations towards the CHILDHOPE consortium. By sending a questionnaire with the information attached we have at the same time informed the ICCCP partners of the recent progress in the CHILDHOPE consortium and yet again raised their awareness. The aim is to have preliminary results ready in the first quarter of 2010 and have an article ready to be submitted by the end of 2010. The results will be implemented in the information for patients and parents as soon as possible. Furthermore a European meeting is being planned in the beginning of 2010.

### Contractors involved

Participant Number	Participant name	Participant short name	Country
1	Centre Léon Bérard	CLB	FR
3	Fondazione matilde Tettamanti e Menotti de Marchi (Università Milano Bicocca)	FOMT	IT
4	The Norwegian Radium Hospital	NRH	NO
5	Etablissement Français du Sang	EFS	FR
6	University College London	UCL	UK
7	Westfälische Wilhelms-Universität Münster	WWU	DE
8	International Confederation of Childhood Cancer Parent Organisation	ICCCP O	NL
9	ACIES	ACIES	FR

### Coordinator contact details

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### Public website

The website (<http://www.childhope.eu/>) is managed by partner 6, University College of London.  
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