The ACTolog® Approach: Multi-target Adoptive Cell Therapy using Endogenous Antigen Specific T cells

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Background

Adoptive cellular therapy (ACT) has demonstrated substantial clinical progresses in hematologic cancers; however, only a small proportion of solid tumor patients have benefited from these advances due to i) lack of suitable immunotherapy targets with high specificity in solid tumors, ii) frequent relapse following immunotherapy to single targets often associated with loss of target expression in the tumor. The ACTolog® concept, utilizing antigen specific T cells against targets identified by the Immatics' proprietary XPRESIDENT® technology, is intended to overcome these limitations by addressing multiple novel relevant tumor antigens per patient. ACTolog® is a personalized, multi-targeted ACT approach in which autologous T-cell products are manufactured against the most relevant tumor target peptides for individual patients whose tumors are positive against at least one target from a predefined target warehouse. IMA101 (ACTolog®) targets up to four antigens per patient, selected from a predefined target warehouse and identified using Immatics' proprietary XPRESIDENT® technology. One key defining feature of the approach is the generation of robust and clinically effective T cells following a proprietary process where autologous T cells are primed in the presence of IL-21, followed by HLA tetramer-guided cell sorting and rapid expansion. Immatics has an exclusive platform to create this product through its in-licensing of the IL-21 mediated approach, which results in higher frequencies of central memory T cells, extended *in vivo* persistence, and a more robust clinical response immunotherapy[1,2]. Products generated by the ACTolog® approach are currently being used in the FIH trial IMA101-101 in collaboration with The University of Texas M.D. Anderson Cancer Center.

Target Identification with XPRESIDENT®

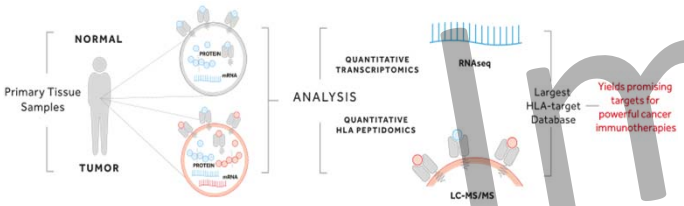
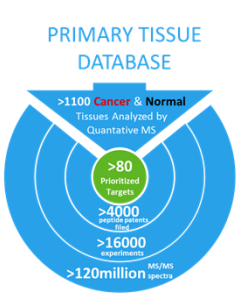


Figure 1. Overview of the XPRESIDENT® platform. HLA-bound, tumor-associated peptides (TUMAPs) are identified and quantified directly from primary human tumor and normal tissue samples by liquid chromatography tandem mass spectrometry (LC-MS/MS). Over-presentation on peptide level is confirmed by over-expression using RNAseq. Furthermore, healthy tissue analyses help to predict potential on- and off-target toxicities.



Cancer Tissues from 20 Major Indications	Control Tissues from 40 Normal Tissues	
AML	Adipose tissue	Ovary
Bladder Cancer	Adrenal Gland	Pancreas
Breast Cancer	Artery	Peripheral nerve
CLL	Bladder	Pituitary gland
Colorectal Cancer	Blood cells	Placenta
Esophagus Cancer	Bone Marrow	Pleura
Gallbladder	Brain	Prostate
Gastric Cancer	Breast	Rectum
Glioma	Cartilage	Salivary gland
Liver Cancer	Cervix	Skin
Melanoma	Colon	Small intestine
Multiple Myeloma	Esophagus	Spleen
Non-Hodgkin lymphoma	Eye	Stomach
NSCLC	Gall bladder	Testis
Ovary	Heart	Thymus
Pancreas Cancer	Kidney	Thyroid
Prostate Cancer	Liver	Trachea
Renal Cell Carcinoma	Lung	Ureter
SCLC	Lymph node	Uterus
Uterus	Muscle	Vein

Figure 2. Key figures of the XPRESIDENT® database. Largest database of HLA peptides and their abundance on different tissues worldwide.

Biomarker Development: IMA_DETECT

Personalization workflow is based on mass spectrometry guided qPCR thresholds. Biopsies from cancer patients are used to measure mRNA expression of warehouse targets using qPCR. Target peptides are considered to be presented by the tumor if expression of corresponding mRNA is above the threshold.

Figure 3. Application of a qPCR based biomarker panel (IMA_DETECT) for target and patient selection.

Manufacturing of ACTolog® Drug Products

Leukapheresis is taken from the patient to isolate PBMCs if at least one target is expressed. CD25 depleted PBMCs and antigen loaded autologous dendritic cells (DC) are used for *in vitro* priming (STIM) for up to four targets in parallel. Antigen-specific T cells are sorted under aseptic conditions using clinical grade pHLA tetramers. After two rounds of rapid expansion (REP) *ex vivo* the drug substance (DS) is filled in infusion bags and stored until patient infusion. In addition to numerous in-process controls, each drug product (DP) is subjected to a final release testing.

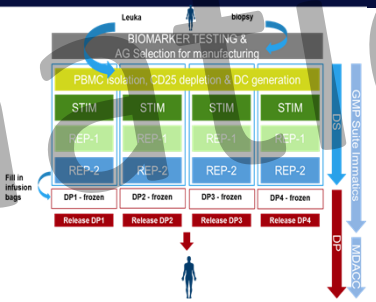
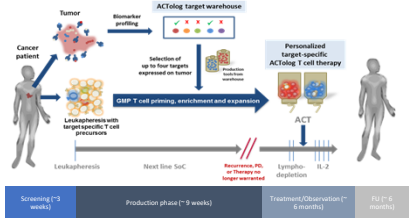


Figure 4. Process flow of patient's drug product manufacturing.

ACTolog®: IMA101-101 Clinical Study Design



ACTolog® is a FIH trial in patients with relapsed or refractory solid cancers. Patients are included based on HLA type and expression of warehouse target(s). Patients undergo leukapheresis to collect mononuclear cells for manufacturing IMA101 cells. IMA101 is infused after lymphodepletion and low-dose IL-2. The study plans to treat 20 patients with ACTolog® IMA101 products.

Figure 5. Clinical study design.

Isolation of Antigen Specific Patient T cells

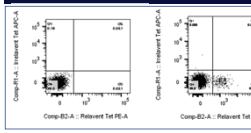


Figure 6a.
Well analysis after DC-antigen stimulation.
Stimulated wells are analyzed using MACSQuant® analyzer and positive wells are pooled before being tetramer-guided sorted for antigen specific T cells. **Left**: Negative well. **Right**: Positive well.

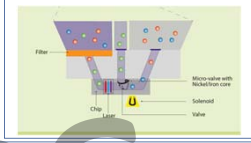


Figure 6b.
MACSQuant® Tyto™ nanosorter system.
Microchip-based cell sorting in a fully closed cartridge system protects the cell sample from contamination.
(adapted from www.miltenyibiotec.com)

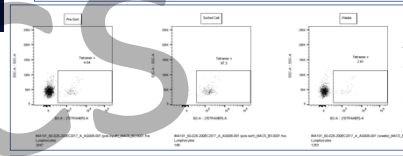


Figure 6c.
Evaluation of cell sorting.
Tetramer staining of pre- and post-sort on the Tyto nanosorter is analyzed using MACSquant® analyzer for sort quality (Enrichment ®= 21x, Yield = 58%).

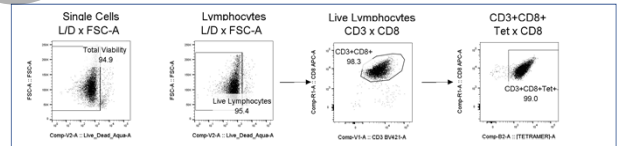


Figure 6d. Analysis of final T-cell products by flow cytometry.
After two rounds of rapid expansion (REP), antigen specific T-cell products are analyzed for lymphocytes content, CD3CD8, and CD3CD8 Tetramer content.

Figure 6. Isolation of patient's antigen specific T cells using a nanosorter system and manufacturing of T-cell products.

Patient IMA101_60_027 Analysis

Targets		Final Product					
	Positive	% Viable	% CD8+	% CD8+Tet+	Final	% Cell	Product
	Wells	Lymphs	Cells	Cells	Cell Yield	Viability	Bags
Ag-008-01	5	94.2	97.3	71.1	1.84E+10	90.5	4
Ag-012-01	4	95.8	98.9	99.0	2.69E+10	91.5	4
Ag-018-01	10	95.6	97.9	98.2	2.86E+10	94.3	4

Table 1. Characteristics of exemplary antigen specific T-cell products manufactured for IMA101 trial patient 027.

References

1. Chapuis, AG, Desmarais, C, Emerson R, Schmit TM, Shibuya K, Lai, I, Wagener F, Chou J, Roberts IM, Coffey DG, Warren E, Robbins H, Greenberg PD, Yee C (2017). *Science Immunology* 2(9)
2. Yee C. *Immunol Rev.* Jan;257(1):250-63. 2014

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