PEPperPRINT
A NEW DIVERSITY

Immunological Services | Regulatory T-Cells

Analyze Treg-mediated immune responses to validate your therapeutic candidates

Regulatory T-cells (Tregs) are essential for controlling the immune response and maintaining self-tolerance. Hence, **Tregs are promising cellular targets for cancer and autoimmune diseases treatment**. While preventing Treg-mediated immunosuppression can help to reinforce antitumor immune responses, forcing an induced Treg (iTregs) phenotype can temper pathologies in autoimmune diseases or transplant rejection.

With our **iTreg phenotyping** service, we can monitor the success of an ongoing clinical trial. Furthermore, via our **iTreg polarization** assay and **iTreg suppression** assay, we can support the development of Treg-targeting drug candidates by validating the modulating effect of your therapeutic candidates on T-cell polarization and function. The iTreg polarization and iTreg suppression assays are established for drug screenings in immune-oncology and autoimmune diseases.

Applications

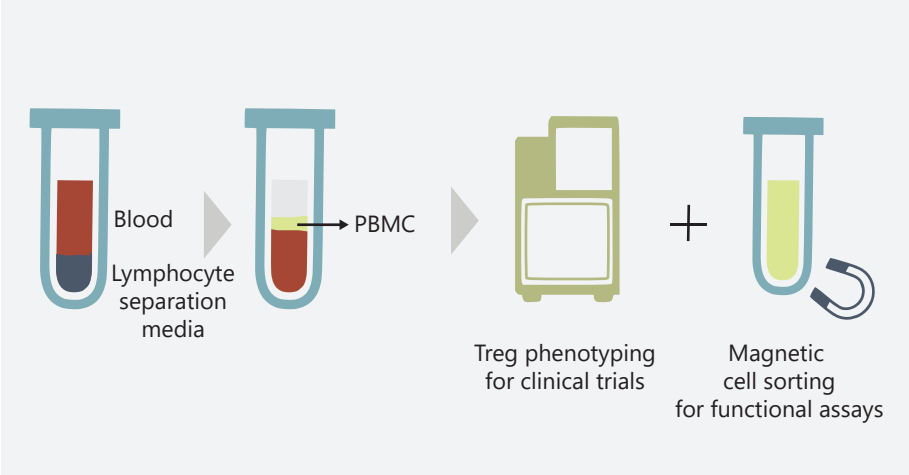
Monitor *in vitro* the amount and character of Tregs during **patients' treatment in clinical trials**.

Test the effectiveness of a drug in enhancing the regulatory phenotype and functional character of iTregs, which could be beneficial for **treating autoimmune diseases or transplant rejection.**

Revise the efficiency of a drug candidate by preventing Treg differentiation and inhibiting iTreg function. This can support cancer treatment by inhibiting Tregs to boost antitumor immune responses**.**

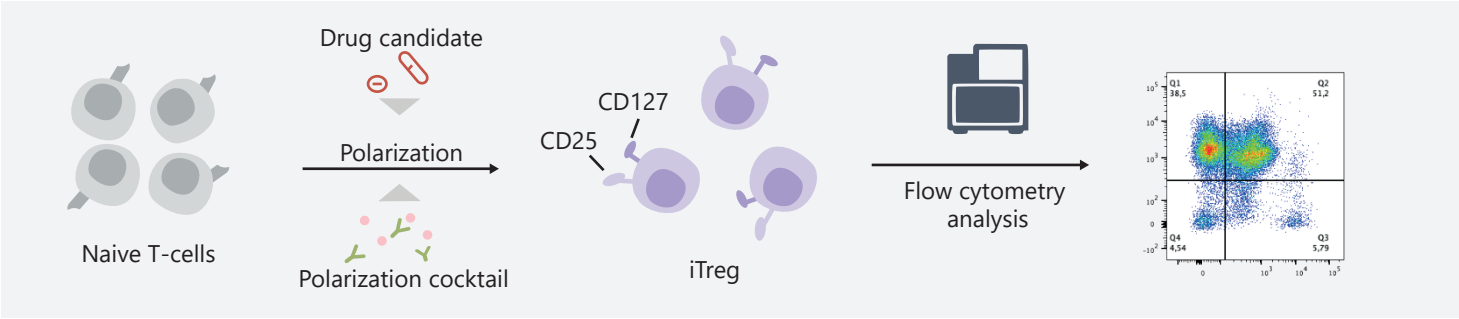
Treg Phenotyping Assay Service

The Treg phenotyping assay is based on antibody-staining of Treg-specific transcription factors, surface markers, and quantitative analyses by flow cytometry. It allows the **characterization of Tregs** among the T-lymphocyte population or peripheral blood mononuclear cell (PBMC) samples. Subsequently, the identified Treg population can be sorted for further functional assays.

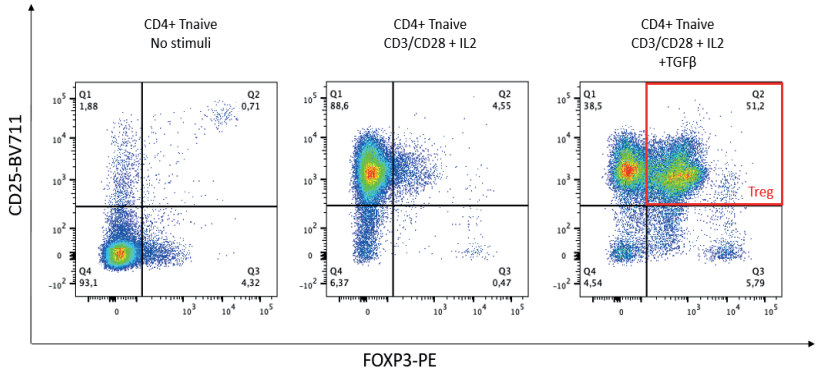


iTreg Polarization Assay Service

The iTreg polarization assay is a rapid and reliable *in vitro* system to quantitatively validate the capability of therapeutic candidates to modulate differentiation of naive T lymphocytes towards an iTreg phenotype, or to verify the candidate compound-mediated prevention of iTreg polarization.



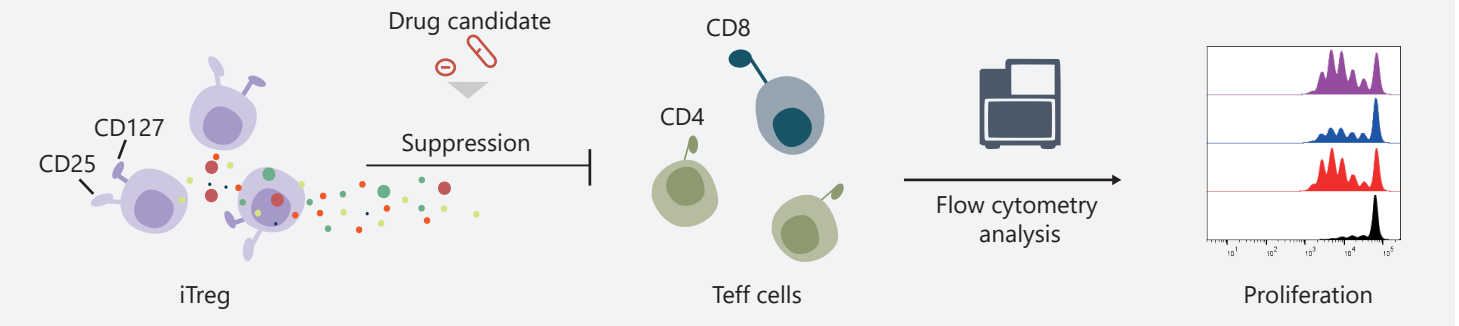
iTreg Polarization Assay. Naïve CD4⁺ T cells isolated from peripheral blood mononuclear cells (PBMC) are activated with a Treg-polarization cytokine cocktail. The drug candidates can be screened for their ability to boost or prevent the differentiation of Tregs via analyzing the expression of Treg-specific markers such as FOXP3, CD127, and CD25 via flow cytometry.



Representative FACS plots of a differentiation assay showing successful generation of iTregs from naive CD4⁺ cells. FOXP3 and CD25 expression in control T-cells and iTregs induced by the differentiation protocol above is shown. The pseudocolor plots show representative FOXP3 and CD25 stainings for one donor, pre-gated on singlet, live CD4⁺ cells.

iTreg Suppression Assay Service

The iTreg suppression assay constitutes a rapid and reliable *in vitro* functional test that validates the iTreg-induced suppression of functional T-cells by analyzing the proliferation of cocultured T lymphocytes. The assay can **monitor the modulating effect of drug candidates on Treg induction** and reveal booster or suppressor functions. This assay complements the iTreg polarization assay to validate the T-cell modulating effect of the drug candidate.



iTreg Suppression Assay. Fluorescently labeled T effector cells are activated and cultured alone or with polarized autologous iTreg cells and treated with the drug candidates. The proliferation of T effector cells and the suppressive effect of iTregs on the proliferation is analyzed by measurement of the decreasing fluorescence signal via flow cytometry. Finally, the modulation of iTreg-mediated suppression through the drug candidate is assessed.