SUPLEXA, a multimodal autologous cellular therapy, shows immunomodulatory behavior in cancer patients consistent with improved anti-tumor immune function

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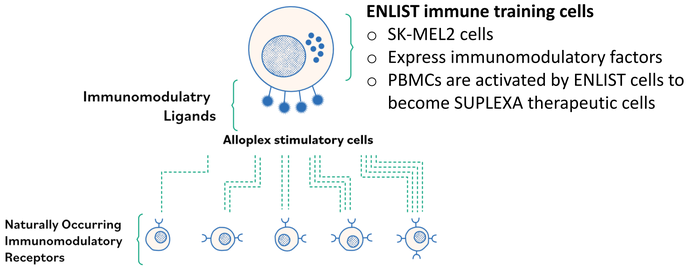
Background

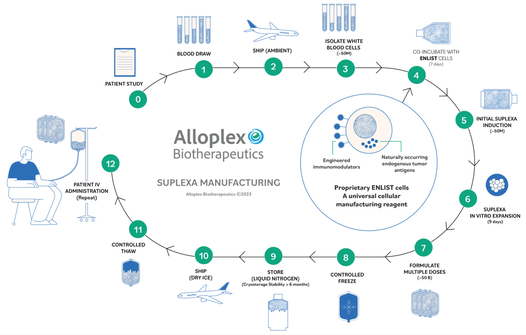
SUPLEXA therapeutic cells are PBMC-derived activated white blood cells, comprised predominantly of lymphocytes, notably devoid of B cells, myeloid, cells, and Tregs. SUPLEXA cells are non-engineered autologous immunotherapeutic cells that are differentiated by an *in vitro* "**immune cell training**" process mediated by engineered tumor cells called **ENLIST cells** that express an array of immunomodulatory adjuvants that convert PBMCs into SUPLEXA cells.



What are SUPLEXA cells? SUPLEXA cells are an autologous cellular immunotherapy for cancer. They have 4 basic immune properties:

1) **Migratory -** Express many chemokine receptors and adhesion molecules.
2) **Cytolytic -** Express high levels of granzymes and perforins
3) **Antigen Presenting Capacity –** Express MHC class II and T cell costimulators, CD80, CD86, and CD40.
4) **Immunomodulatory –** Modulate cancer patient's bone marrow output with rapid alterations to the peripheral myeloid cell populations after adoptive immunotherapy

SUPLEXA manufacturing



Longitudinal Blood Immunophenotyping Studies

Blood Mass Cytometry (CyTOF) Analysis. Blood samples were collected at baseline and multiple time points after SUPLEXA treatments. PBMCs were prepared and cryopreserved for phenotyping by CyTOF. CyTOF panels containing 48 different marker antibodies were designed to identify and profile multiple immune cell types. CyTOF staining data were analyzed by a workflow to identify longitudinal changes in immune cell phenotypes.

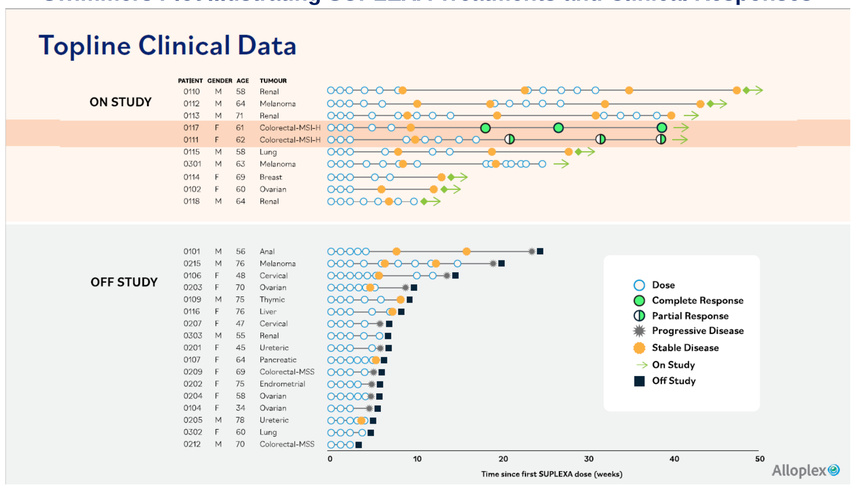
Plasma Biomarker Discovery and Detection by Olink and Luminex Methods. Luminex assays for 40 different cytokines were performed. To discover novel biomarkers, the Olink Discovery panel (3,072 proteins) was used on a subset of patient plasma samples. Data was analyzed by Olink Analyze R package and STRING for network and GO enrichment.

Phase 1 Clinical Outcomes

Clinical Trial Design: Enrolled heavily pre-treated end-stage cancer patients with a variety of metastatic tumor types. SUPLEXA were given to patients at 2.5 billion cell doses at 1 week or 2-week intervals. All patients received at least 3 SUPLEXA doses. Some patients were given multiple doses as indicated on the Swimmers Plot. **Blood samples** were collected at each visit for treatment or follow-up CT scans.

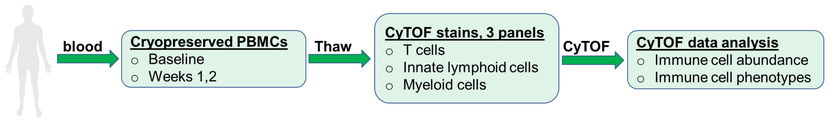
Clinical Efficacy: An ongoing phase 1 clinical trial in late-stage cancer patients has shown that SUPLEXA cells are **1) safe with no adverse events in 28 patients and 2) showed the highest single-agent efficacy in MSI-high colorectal cancer patients.** Beneficial responses were also found in renal cancer and melanoma patients**.**

Swimmers Plot Illustrating SUPLEXA Treatments and Clinical Responses



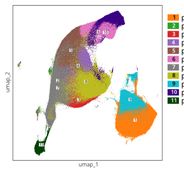
Longitudinal Blood CyTOF Workflow and Data Analysis

Patient PBMC CyTOF Workflow



Longitudinal profiling of 17 patients' PBMCs by CyTOF: Myeloid Cell Populations

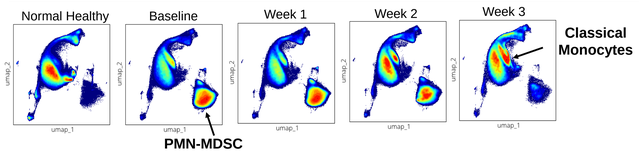
uMAP showing clusters



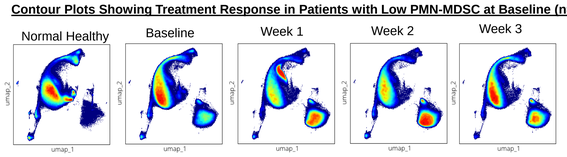
Cluster identification

Cluster#	Cell Type
1	PMN-MDSCs
2	Classical Monocytes
3	M-MDSCs
4	Classical Monocytes
5	Intermediate Monocytes
6	Non-Classical Monocytes
7	Blood Dendritic Cells
8	Blood Dendritic Cells
9	PMN-MDSCs
10	Non-Classical Monocytes
11	Plasmacytoid Dendritic Cells

Contour Plots Showing Treatment Response in Patients with High PMN-MDSC (>20%) at Baseline (n=10 patients)

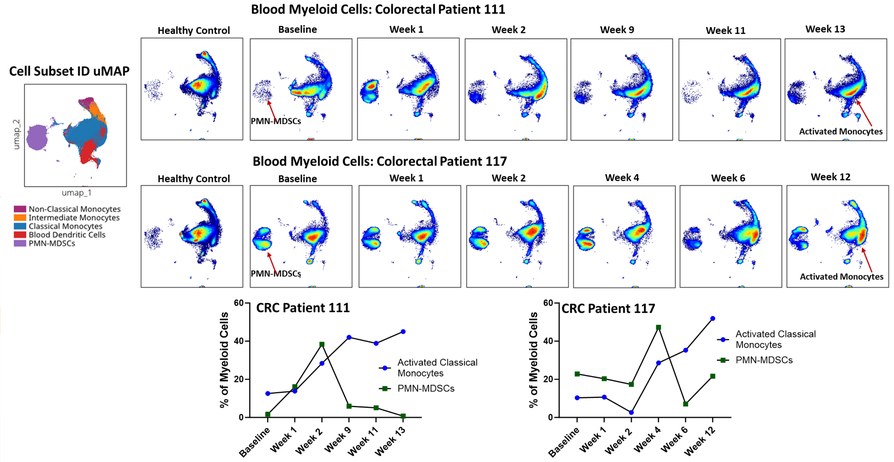


Contour Plots Showing Treatment Response in Patients with Low PMN-MDSC at Baseline (n=7 patients)



Blood CyTOF Analysis of MSI-H CRC Patients

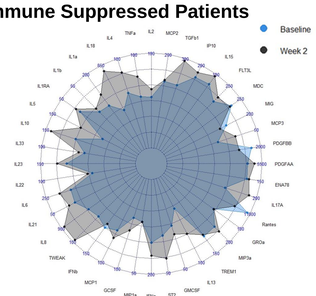
Longitudinal blood samples from MSI-H CRC patients were analyzed for changes in myeloid cell populations by CyTOF



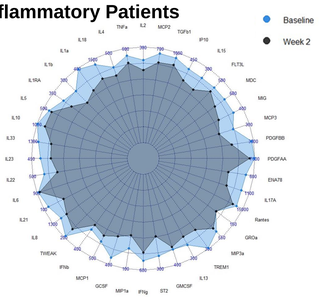
Plasma Proteomics by Luminex and Olink

Luminex Plasma Cytokine Profiles

Immune Suppressed Patients

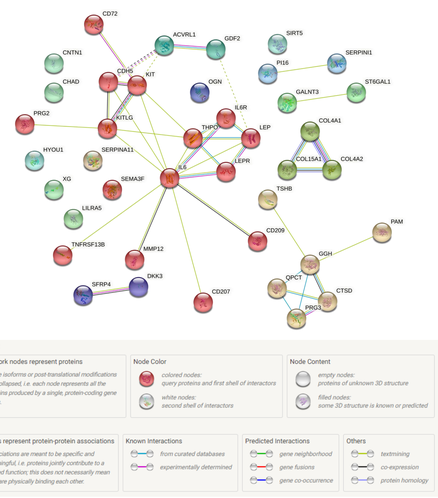


Inflammatory Patients



Olink Discovery Plasma Proteomics

STRING network analysis of longitudinal plasma markers significantly changed by SUPLEXA



Summary and Conclusions

- Phase 1 clinical outcomes indicate; **1) pristine safety** in all 28 end-stage cancer patients treated with SUPLEXA cells, **2) Complete or partial responses** in MSI-H colorectal cancer patients and **stable disease** in renal cell carcinoma and melanoma patients.
- Immunomodulation of **blood activated classical monocytes** and **PMN-MDSCs** in CRC patients showing beneficial clinical responses to SUPLEXA therapeutic cells.
- Circulating **cytokines** and **biomarkers** are significantly modulated by SUPLEXA cell treatments.