P17.10.B

DECIPHERING THE INFLAMMATORY TUMOR MICROENVIRONMENT IN RECURRENT, THERAPY-RESISTANT GLIOBLASTOMA

M.-F. Ritz[1], T. Shekarian[1], T. Martins[1], P. Schmassmann[1], J. Roux[2], B. Burger[3], Sabrina Hogan[1], and G. Hutter[1]

[1]Brain Tumor Immunotherapy and Biology Laboratory, [2]Bioinformatics Core Facility, [3]Human Genomics, Department of Biomedicine, University of Basel and University Hospital Basel, Switzerland

Background

Glioblastoma (GBM) is a lethal brain tumor without effective treatment options in which relapses always occur. While some studies have looked into genetic differences between primary and recurrent GBM, no study has specifically analyzed the immune microenvironment with focus on TAMs of recurrent GBM and compared treatment-naïve tumors to patient-matched recurrent tumors. Changes of the immune profile during disease progression and concurring therapy may occur, and only very limited studies have been performed on this topic so far. In order to target the tumor-induced immune evasion mechanisms for clinical research, we characterized the immune profiles at initial and relapse more precisely by transcriptomic, proteomic and spatial transcriptomic analyses.

Objectives

To target tumor-induced immune evasion mechanisms of glioblastoma for clinical research, we aimed at characterizing the immunological changes in patient-matched treatment-naïve and recurrent GBM samples using transcriptomic (17 patient-paired samples) proteomic (6 patient-paired samples) and spatial transcriptomic (7 patient-paired core samples) analysis.

Material and Methods

Patients

Patient-matched treatment-naïve (primary) and recurrent *IDH1* wild type GBM fresh-frozen tissue samples were included after informed consents were obtained. All patients received chemo- and radiotherapy after the first maximal tumor resection. Time to relapse and other clinical data were collected.

Gene expression analysis

Total RNA were extracted from tumor samples and the expression of genes involved in brain immune responses, neuroinflammation and cancer were compared using the nCounter® Neuroinflammation and IO360 PanCancer panels (NanoString).

Proteomic

Total proteins were extracted, digested and subjected to TMT labeling then HPLC and separated by LC-MS/MS.

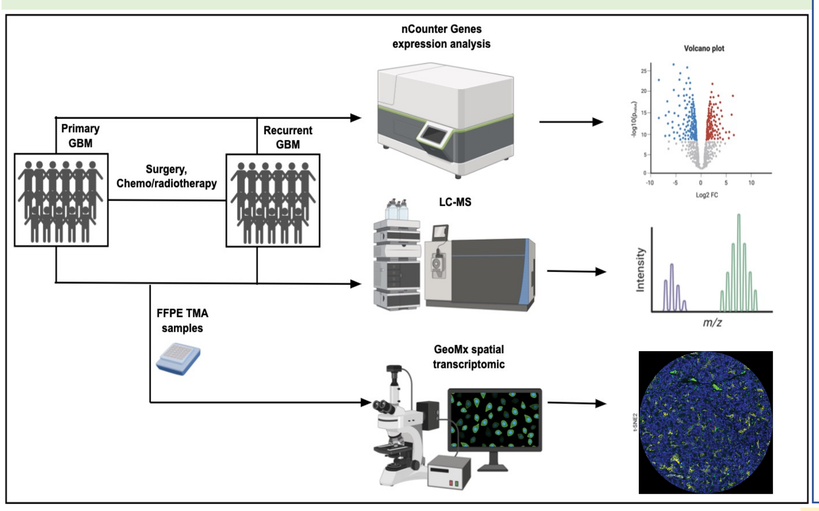
Spatial transcriptomic

Gene expression across patient-matched primary and recurrent tumor cells (GFAP positive) and CD64-positive cells were performed using a tissue microarrays analysed with GeoMX Digital Spatial Profiler (Nanostring). The whole transcriptome for each segment (GFAP or CD64) in the selected ROI was analysed.

Biostatistical analysis

After normalization, all -omic data were analysed using R. Differentially expressed genes/proteins were determined using Limma.

Cell type characterization was obtained using Darmanis Atlas and a modified Allen Atlas.



Fundings

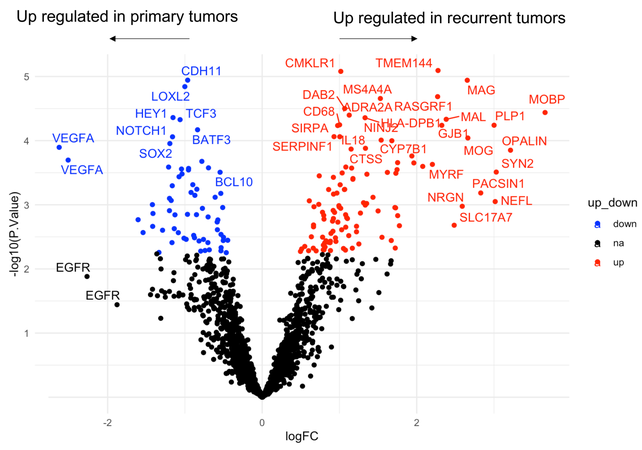
Departement Biomedizin Basel

SwissLife

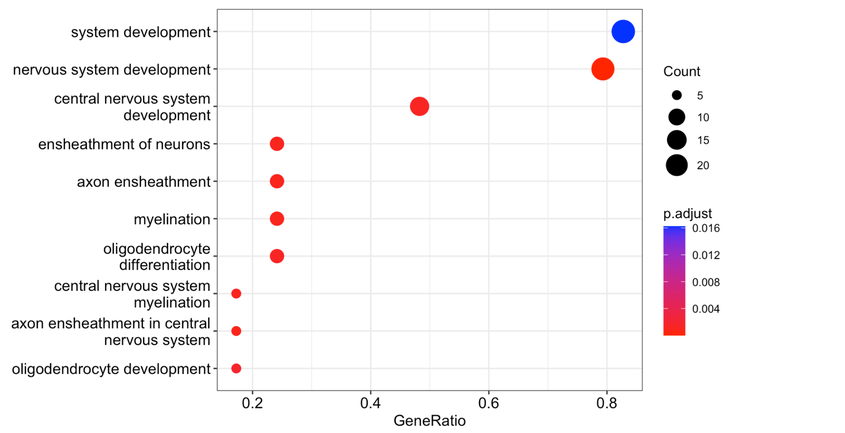
Universitätsspital Basel

Results 1. Transcriptomic

Differentially expressed genes (DEGs)

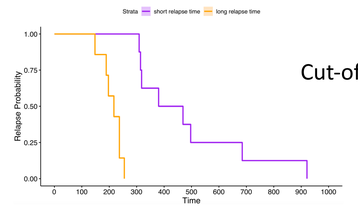


Pathways associated with enriched DEGs

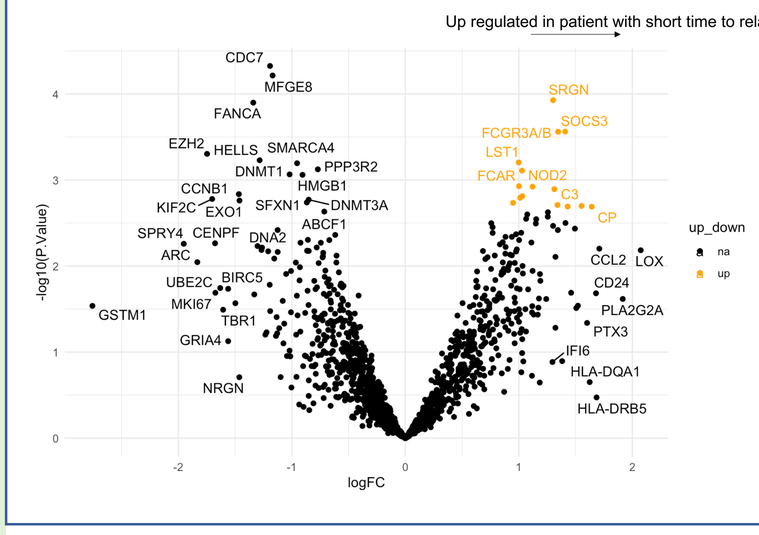


Differentially expressed genes (DEGs) between long and short time to relapse and associated pathways

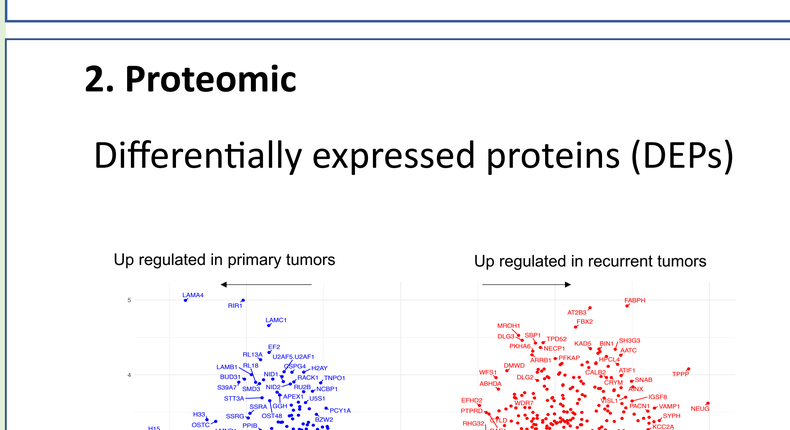
Statification of patients into SHORT and LONG times to relapse



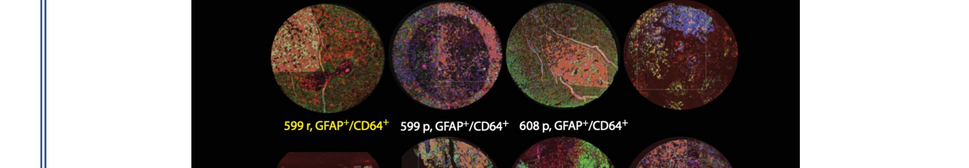
Cut-off: 310 days



Pathways associated with enriched DEPs

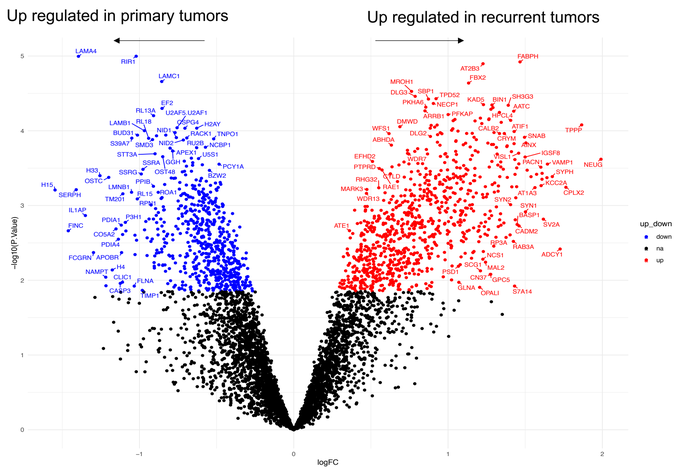


High expression in recurrent tumors strongly correlated with shorter time to relapse for FCGR1A (CD64) and other inflammatory genes

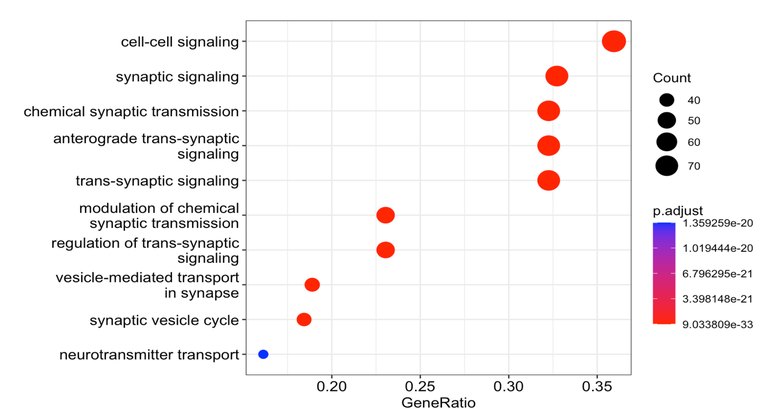


2. Proteomic

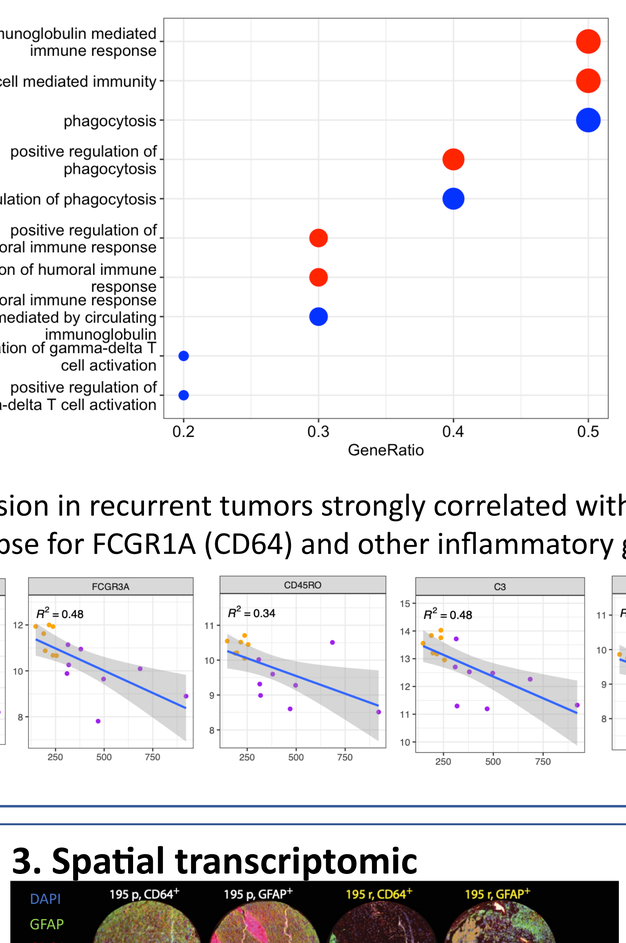
Differentially expressed proteins (DEPs)

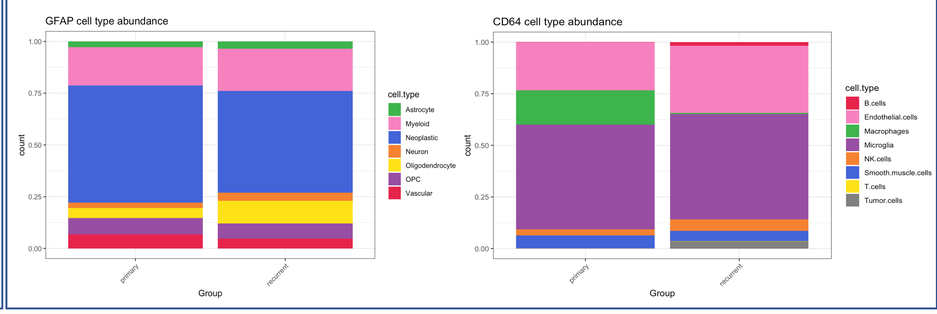


Pathways associated with enriched DEPs



3. Spatial transcriptomic





Conclusions

Transcriptomic and proteomic analyses of patient-paired GBM tissue samples showed that components related to neurogenesis, myelination, synaptogenesis, antigen presentation and phagocytosis are more present in recurrent tumors. High expression of specific genes such as CD64 in relapsing tumors correlated with shorter time to relapse. A change in the cell composition in the tumor immune environment was observed with special transcriptomic, with less macrophages and less microphages in recurrent tumors. These findings illustrate specific proinflammatory phenotypes that may impact GBM recurrence and may represent new therapeutic targets to be used after initial removal of the primary tumors.