cytoskeleton and GAP-43 mediates the axonal conus growth. We aimed to investigate the impact of GAP-43 and actin expression on overall survival (OS) as well as crucial epidemiologic, radiologic and neuropathological parameters (TAM) in glioblastoma. MATERIAL AND METHODS: FFPE tissue of adult patients with diffuse and anaplastic gliomas, who underwent first surgery in our center between 2010 and 2019, were selected. GAP-43 and actin expression was analyzed using immunohistochemistry and semi-quantitatively ranked. Clinical, neuropathological as well as follow-up-data were gained from the institutional neuro-oncological database. RESULTS: 118 patients with a median age of 46 years (IQR: 35 - 57) were evaluated. 48 (41%) presented with a diffuse glioma and 70 (59%) revealed anaplasia. 96 (82%) cases were presented with intermediate or strong GAP-43 expression and 78 (67%) with no or light actin expression. Tumors with higher expression of GAP-43 (p=0.024, HR=1.71/rank) and actin (p=0.001, HR=2.28/rank) showed significantly reduced OS. IDH wildtype glioma demonstrated significantly more expression of both proteins: GAP-43 (p=0.009) and actin (p=0.001). The same was confirmed for anaplasia (GAP-43 p=0.028, actin p=0.029), higher proliferation rate (GAP-43 p=0.016, actin p=0.038), contrast-enhancement in MRI (GAP-43 p=0.023, actin p=0.037) and age (GAP-43 p=0.004, actin p=0.001). CONCLUSION: The intercellular distinct communication network in diffuse and anaplastic gliomas formed by actin and GAP-43 is associated with a negative impact on overall survival and unfavorable prognostic features.

P12.06.A. RELATIONSHIP BETWEEN ASCORBATE AND DNA METHYLATION MARKERS IN CLINICAL GLIOMA TUMOURS

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BACKGROUND: Members of the 2-oxoglutarate-dependent dioxygenase family (67%) with no or light actin expression. Tumors with higher expression of GAP-43 (p=0.024, HR=1.71/rank) and actin (p=0.001, HR=2.28/rank) showed significantly reduced OS. IDH wildtype glioma demonstrated significantly more expression of both proteins: GAP-43 (p=0.009) and actin (p=0.001). The same was confirmed for anaplasia (GAP-43 p=0.028, actin p=0.029), higher proliferation rate (GAP-43 p=0.016, actin p=0.038), contrast-enhancement in MRI (GAP-43 p=0.023, actin p=0.037) and age (GAP-43 p=0.004, actin p=0.001). CONCLUSION: The intercellular distinct communication network in diffuse and anaplastic gliomas formed by actin and GAP-43 is associated with a negative impact on overall survival and unfavorable prognostic features.

P12.08.A. UNCOVERING THE GliOBLAstoma TUMOR MICROENVIRONMENT BY HIGH-END MULTIPLEXING WITH IMAGING MASS CYTOMETRY

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BACKGROUND: Glioblastoma is one of the most aggressive cancers, and hypoxia plays an essential role in its tumor microenvironment. Tumor-associated macrophages (TAMs) and microglia, as markers of hypoxia, have been shown to be significant prognostic factors. However, the molecular mechanisms underlying the contribution of TAMs to glioblastoma have not been fully elucidated. The aim of this study was to uncover differences between the hypoxic and normoxic tumor microenvironment of glioblastoma by high-end multiplexing with imaging mass cytometry. MATERIAL AND METHODS: A tissue microarray (TMA) with normoxic and hypoxic areas from 4 IDH-wildtype glioblastomas was prepared based on the hypoxia marker hypoxia-inducible factor 1 alpha (HIF1 alpha). The TMA was stained with 18 metal-tagged antibodies covering TAMs, lymphocytes, immune checkpontes, tumor cells and proliferation. The Hyperion-CYTOF technology was used to ablate the samples and the images were analyzed by MCY viewer, Vissopharm software, and customized R scripts. RESULTS: Single-cell analyses of 160 fields covering around 45,000 cells in the glioblastoma microenvironment revealed multiple cellular phenotypes. It was revealed that proliferating TAMs (CD11b+) and monocytes expressed CD206, whereas proliferating vessels (CD34+, K67+) were more frequent in normoxia. Additionally, proliferating stem-like tumor cells (OLIG-2+, K67+) were more frequent in normoxia regions. CONCLUSION: Our study revealed multiple cellular phenotypes in the glioblastoma microenvironment. The TAMs, endothelial and tumour cell phenotypes revealed may play a critical role in glioblastoma biology however this needs to be elucidated in future studies.

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P12.07.B. GETTING TO THE CORE OF MICROGLIA VERSUS BONE MARROW-DERIVED MACROPHAGES IN GliOBLAstoma

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BACKGROUND: Microglia and bone marrow-derived macrophages (BMDMs) are two ontogenetically distinct myeloid populations present within glioblastoma that can comprise 30-50% of the tumour mass. Historically, these cell types have been conflated and studied as a single population of ‘tumour-associated macrophages’. Recent advances in single-cell omics provide a new approach for delineating these populations. FEPEE tumours have allowed us to study the temporal and spatial evolution of these populations, suggesting that BMDMs may play different roles within the tumour and subsequently differentially affect tumour progression. Despite building evidence for the unique functions of these cells within glioblastoma, the inherent heterogeneity of these populations remains understudied. Indeed, both microglia and BMDMs exist as phenotypically and functionally diverse populations that are polarised in a context-dependent manner. Hence, to understand the differences between microglia and BMDMs within glioblastoma, both ontogeny and spatial location must be considered. MATERIAL AND METHODS: To elucidate the functionalact of microglia and BMDMs across the tumour landscape, a publicly available RNAseq dataset was utilised to classify myeloid cells into four populations based on spatial location and ontogeny. These were tumour core BMDMs and microglia, or tumour periphery BMDMs and microglia. Differential gene analysis was performed to identify significant differentially expressed genes (DEGs) between classified myeloid populations. Tumour core DEGs were then compared against the Ivy Glioblastoma Atlas to define their expression across anatomical tumour regions. Finally, myeloid DEGs were validated at the protein level on human glioblastoma tissue through immunohistochemistry. RESULTS: Microglia and BMDMs showed different spatial distributions across the tumour landscape and displayed distinct functional expression profiles. Microglia held a more chemotactic and immune-inflammatory profile whereas BMDMs held a more pro-tumourigenic profile. However, a comparison of microglia between the tumour core and periphery revealed that tumour microglia upregulate many pro-tumourigenic genes, including multiple genes that have previously been defined as BMDM markers. Moreover, tumour periphery microglia are more enriched for the tumour core to distinct spatial tumour regions such as the vascular or hypoxic niche. Immuno histochemical staining reflected these spatial expression profiles, identifying a distinct population of phagocytic macrophages within the hypoxic regions. CONCLUSION: These results suggest that myeloid cell expression is dynamically influenced by the tumour microenvironment, rather than ontogeny alone.

P12.09.B. EXTRACELLULAR-vesicle DERIVED-MIR-146A INCREASES MELANoma BRAIN METASTASIS PROGRESSION VIA NOOTCH SIGNALLING PATHWAY DISREGULATION

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Abstracts