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Implementing an integral quantification of the immune contexture in brain tumors with diagnostic, prognostic and therapeutic implications

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Background: Immune cell infiltration is recognized worldwide as a key biomarker in oncologic diseases, therefore is of great importance to establish a consensus for its recording. Galon *et al.*, proposed in 2014 the immunoscore for colon cancer, which considers the subpopulation density of CD3 and CD8 lymphocyte infiltration at the tumor center and margin. This technique has proved to be useful to predict progression free survival (PFS) and overall survival (OS). Unfortunately, there is no current standard method to determine the immune cell infiltrate in the rest of solid tumors. In this project, we aimed at identifying prognosis and decision-making markers at the immune level, analyzing the tumoral microenvironment at the tumor center and its margin in order to develop a systematic immune score for neurotumors.

Material & Methods: We studied retrospectively, three types of astrocytic tumors: Glioblastomas, anaplastic astrocytomas and diffuse astrocytomas (n=15 of each, 45 in total). After formalin tissue fixation and hematoxylin-eosin staining, we selected 1 representative slide of center and margin from each tumor using the highest optical density area. Afterwards, we objectively quantified cellular components of innate and adaptive immune response, tumoral stroma, necrotic and apoptotic cells at 10 HPF. Finally, we proposed an immune score specific for neurotumors. We performed immunohistochemical analyses of CD4, CD8, FOXP3, STAT3, T-BET for lymphocyte differentiation and M1, M2 for macrophage activation differentiation.

Results: From the 45 tumors included, we found a clear correlation between tumoral immune infiltrate and prognosis, OS and PFS. Patients with high lymphocyte tumoral infiltrate (around 150 cell per HPF) correlated with better clinical response to oncology treatment and longer survival (p=0.005). Likewise, a negative correlation was found between the infiltration of plasmatic cells, neutrophils and macrophages with 50 cell per HPF) in all the tumors studied (p=0.001).

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