Instituto de Medicina Molecular João Lobo Antunes

Master Project Proposal

Title:
Targeting therapeutic resistance in glioblastoma

Synopsis:

Glioblastoma (GB) is the most frequent primary malignant brain tumor in adults[1], accounting for 16% of all central nervous system (CNS) primary tumors[2]. Despite substantial advances in the comprehension of GB biology and the current standard-of-care, including surgery followed by radiation therapy and chemotherapy[3], the median overall survival (OS) is around 15-18 months[4,5] and the 5-year survival is only 5%[6]. This poor prognosis has been mainly associated with tumor invasion of the surrounding healthy brain[7], drug resistance, and the physical and biochemical barrier maintained by the blood-brain barrier (BBB)[8,9]. Tumor invasiveness and resistant cancer cells have been indicated as the main promotors of GB recurrence. Unfortunately, most GB recur after surgery[7] and are usually inoperable. Therefore, there is an unmet medical need for effective therapeutic options for recurrent GB patients.

Previously, our team has performed RNA sequencing in paired GB samples (primary and matched recurrences) to identify genes differentially expressed in recurrent tumors that may play a role in resistance to therapy. Tumor tissues in this discovery cohort were collected in the Department of Neurosurgery at Hospital de Santa Maria, Centro Hospitalar Universitário Lisboa Norte (CHULN), snap frozen and stored in our Brain Tumor Collection at Biobanco-iMM CAML). The list of resistance genes identified by the bioinformatic analysis was narrowed down upon literature review, analysis of publicly available data sets on GB, and after our initial validation of the identified genes by qRT-PCR. The 2 top candidate genes to be further validated were *ANXA4* and *MMP19*. We hypothesize that high expression levels of ANXA4 and MMP19 in GB tumors are associated with increased cancer cell invasion and resistance to therapy, leading to tumor recurrence and a worse patient outcome.

In this master project proposal, the main goal is to evaluate the role of ANXA4 and MMP19 in recurrent GB. To accomplish this, the work plan has 3 objectives:

1) Determine the clinical relevance of candidate genes by quantifying the expression (immunohistochemistry) of ANXA4 and MMP19 using an independent cohort of primary and recurrent GB matched samples and correlate the expression levels with time to relapse (progression-free survival) and overall survival.

2) *In vitro* functional validation of the role of candidate genes in developing temozolomide (TMZ) and/or radiotherapy (RT) resistance using a commercially available GB cell line (U87 cell line) with overexpression of ANXA4 and MMP19, with and without treatment.

3) *In vivo* functional validation of the role of candidate genes in GB progression using intracranial mice models injected with the U87 cell line with overexpression of ANXA4 and MMP19, with and without treatment.

At the end of this project, we expect the results obtained may contribute to validate ANXA4 and MMP19 as important players in GB recurrence. These results can pave the way for future research in the identification of novel targeted therapies, improving the outcome of GB patients.

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Bibliography (facultative):

1. Louis DN, Ohgaki H, Wiestler OD, Cavenee WK, Burger PC, Jouvet A, et al. The 2007 WHO classification of tumours of the central nervous system. Acta Neuropathol (2007)114,2:97-109.
2. Suzuki H, Aoki K, Chiba K, Sato Y, Shiozawa Y, Shiraishi Y, et al. Mutational landscape and clonal architecture in grade II and III gliomas. Nat Genet (2015)47,5:458-68.
3. Baumert BG, Hegi ME, van den Bent MJ, von Deimling A, Gorlia T, Hoang-Xuan K, et al. Temozolomide chemotherapy versus radiotherapy in high-risk low-grade glioma (EORTC 22033-26033): a randomised, open-label, phase 3 intergroup study. Lancet Oncol (2016)17,11:1521-32.
4. Ostrom QT, Gittleman H, Fulop J, Liu M, Blanda R, Kromer C, et al. CBTRUS Statistical Report: Primary Brain and Central Nervous System Tumors Diagnosed in the United States in 2008-2012. Neuro Oncol (2015)17 Suppl 4:iv1-iv62.
5. Taal W, Bromberg JE, van den Bent MJ. Chemotherapy in glioma. CNS Oncol (2015)4,3:179-92.
6. Louis DN, Perry A, Burger P, Ellison DW, Reifenberger G, von Deimling A, et al. International Society Of Neuropathology--Haarlem consensus guidelines for nervous system tumor classification and grading. Brain Pathol (2014)24,5:429-35.
7. Appin CL, Brat DJ. Molecular pathways in gliomagenesis and their relevance to neuropathologic diagnosis. Adv Anat Pathol (2015)22,1:50-8.
8. Weller M, Wick W, Aldape K, Brada M, Berger M, Pfister SM, et al. Glioma. Nat Rev Dis Primers (2015)1:15017.
9. Ohgaki H, Kleihues P. The definition of primary and secondary glioblastoma. Clin Cancer Res (2013)19,4:764-72.