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C0160 - EFFECT OF ANTI VEGF-A BEVACIZUMAB IN GLIOBLASTOMA CELL PROLIFERATION IN NORMOXIC AND HYPOXIC CONDITIONS

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Resumen

Objectives: To define citotoxicity and impact of bevacizumab on viability and proliferation of glioblastoma in normoxic and hypoxic conditions.

Methods: Three established cell lines derived from malignant glioma (U251, LN229, U87) were cultured at 21% O_2 (normoxia) or 1% O_2 (hypoxia) for 72 hours. VEGF secretion was assessed with VEGF ELISA KIT HUMAN (Life Technologies) in both conditions. Based on this basal secretion we extrapolated the amount of Bevacizumab needed for VEGF neutralization and checked again using the same kit. Following assays were performed using that neutralizing concentration. Tetrazolium dye assay (MTS; Promega) was used to measure bevacizumab cytotoxicity. Cell proliferation was determined by assessing 5-Bromo-2¿-deoxyuridine (BrdU; 100 ?M, Sigma-Aldrich) incorporation. Morphological studies were carried out by hematoxylin staining.

Results: VEGF neutralization. We defined the individualized concentration of Bevacizumab needed to neutralize VEGF-A secreted by U251, LN229 and U87 in normoxia and hypoxia. U251 and LN229. Citotoxicity and impact of bevacizumab on viability and proliferation. Bevacizumab had no significant citotoxicity effect as shown in the MTS assay. Nevertheless, it induced a morphological change in U251 in normoxic conditions and in U87 in hypoxia, but not in LN229, which remained stable in both cases. The inhibition of VEGF-A affected significantly the proliferation of U251 and U87 in normoxia. By contrast, LN229 cell proliferation was only been reduced in hypoxia.

Conclusions: VEGF neutralization and cell proliferation are observed in normoxic conditions in all cases and in some of hipoxic conditions. Our *in vitro* findings suggest that further studies about Bevacizumab effects, depending on its concentration, are required and must be corroborated by *in vivo* models.