STUDY OF THE BIOLOGICAL PROPERTIES OF THE GLIOBLASTOMACELL LINE U87, WHICH INDUCIBLY SYNTHESIZES SOX2 AND OCT4

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Glioblastoma multiforme (GBM) represents the most prevalent malignant tumor of the central nervous system (CNS). Despite a multimodal therapeutic approach, consisting of maximal surgical resection, radiotherapy, and concomitant chemotherapy, clinical outcomes remain suboptimal. The prognosis for GBM patients is still poor, with a median overall survival of approximately 15 months and a five-year survival rate of less than 5% [1]. Treatment is complicated by chemoresistance caused by Glioma Cancer Stem Cells (GCSCs) [2]. The GCSC phenotype is regulated by a specific group of transcription factors, traditionally identified as the ”Yamanaka factors”: Sox2, Oct4, c-Myc, and Klf4. However, according to research by Lopez-Bertoni et al. [3], the expression of only two genes—SOX2 and OCT4—is sufficient for stemness induction in glioblastoma cells, suggesting that the full reprogramming typically used to generate induced Pluripotent Stem Cells (iPSCs) is not required in this context.

The objective of this study was to investigate how ectopic expression of the SOX2 and OCT4 genes influences the biological functions of glioblastoma cells.

To investigate the causes of glioblastoma malignancy, experiments were conducted using U87-TSO cells, a modified U87 cell line engineered for the tetracycline-inducible expression of the transcription factors Sox2 and Oct4. The proliferation rates of both tetracycline-treated and control U87-TSO populations were quantified via MTT assay. The MTT assay was also used to evaluate the U87-TSO response to the anti-cancer drug temozolomide (TMZ). The ability to form spheroid structures was also evaluated by culturing the cells under conditions that promote a stem-like phenotype. Additionally, the influence of transcription factors on cellular motility was characterized by performing a scratch wound healing assay. All research findings were evaluated using arithmetic mean and standard deviation. Student’s t-test was used to determine the significance level (p) to evaluate data reliability. Results were considered statistically significant and reliable if $p < 0.05$.

Analysis via MTT assay revealed that the tetracycline-induced expression of SOX2 and OCT4 resulted in a reduction of cellular proliferation, accompanied by significant alterations in cell morphology. While the ectopic expression of these transcription factors did not yield a statistically significant change in the sensitivity of U87-TSO cells to TMZ, it appeared to promote cellular viability. Furthermore, U87-TSO spheroids exhibited prolonged viability compared to those formed from unmodified U87 cells. Results from the scratch assay indicated that the migration of U87-TSO cells is significantly slower compared to the parental U87 cell line.

These studies provide new insights into the stemness phenotype regulators Sox2 and Oct4 and the importance of their gene expression level in the process of glioblastoma pathogenesis.

Keywords: Glioblastoma multiforme (GBM), Stemness induction, Yamanaka factors, Tetracycline-inducible system, U87 cell line

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