IS MICROGLIA ASSOCIATED WITH GLIOBLASTOMA ITS ENEMY OR ALLY?

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INTRODUCTION

Glioblastoma (GBM) is heavily infiltrated with tumor-associated microglia/macrophages (TAM). Recently, the role of TAM in GBM progression has received a great deal of interest. Bearing in mind that the number of peripheral macrophages by the 14th day is negligible, in our study TAM were referred to as microglia.

Here we evaluated histopathological characterization of TAM, the kinetics of their infiltration and their impact on U87 orthotopic GBM, a commonly used model in preclinical research.

MATERIALS & METHODS

After placing *Wistar* rats on stereotaxic frame (Stoelting®, USA), U87 cell suspension was injected into putamen. The animals were sacrificed 4, 7 and 14 days post-inoculation (d4, d7, and d14, respectively). Following the tissue processing, immunohistochemical staining with morphometric analyses were perfomed using anti-Ki67, anti-human nucleoli, anti-Iba1, and anti-CD34 antibodies.

CONCLUSION

Clarifying that microglia in this experimental GBM model showed similar morphological pattern and pro-invasive features as microglia in human GBM, these findings highlight the use of microglia in U87 experimental GBM as a potential target for manipulating GBM growth and a new strategy to fight with.

RESULTS

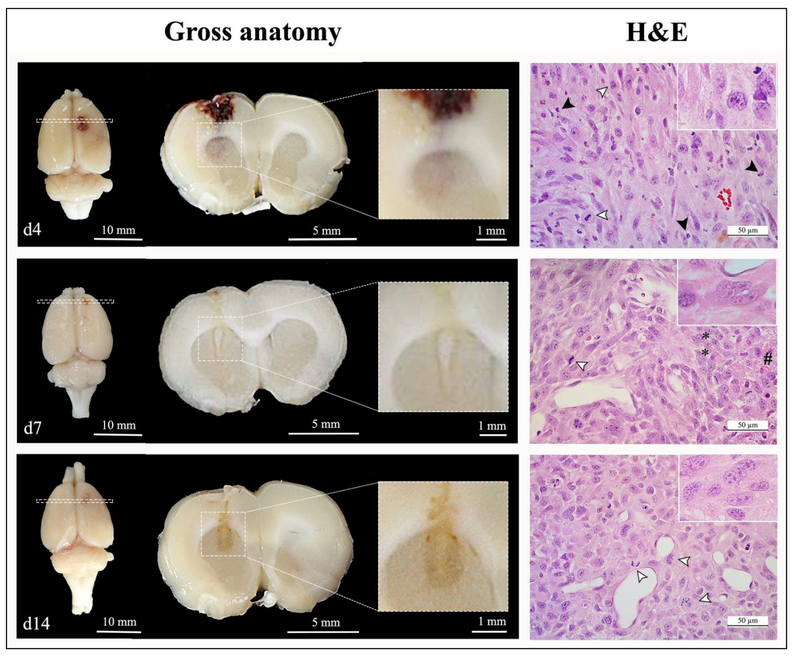

Figure 1. Macroscopic and histopathological features (hematoxylin and eosin staining, H&E) of U87 human glioblastoma xenograft in immunosuppressed Wistar rats 4 (d4), 7 (d7) and 14 days (d14) post-inoculation (n=6 animals per time point). Neutrophil infiltration (black arrowhead) is found in d4 group. Necrosis (#) with pseudopalisading of tumor cells (*) is indicated in d7 group. Mitotic figures of the tumor cells (white arrowhead) are indicated in all three groups.

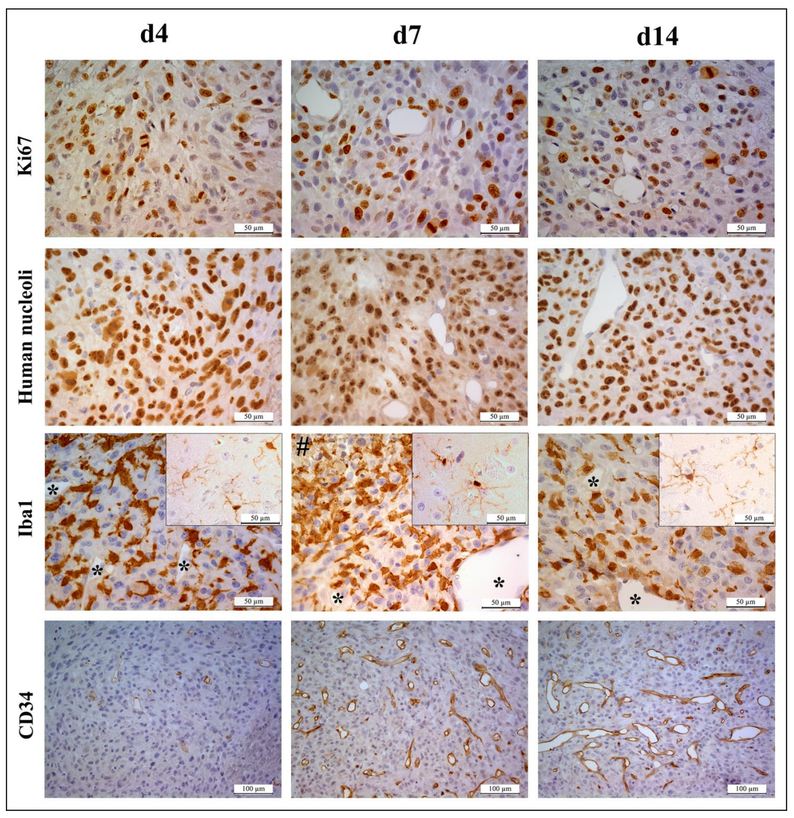

Figure 2. Immunohistochemical features of U87 human glioblastoma xenograft in immunosuppressed Wistar rats 4, 7 and 14 days post-inoculation (n=6 animals per time point): Ki67, Human nucleoli, Iba1 immunostaining of ipsilateral and contralateral side of the tumor - necrotic core is indicated by number sign (#), and blood vessels by asterisk (*), and CD34 immunostaining .

Tumor growth	Ki67 proliferation index (%)	% of the tumor cells within the tumor mass	N (TAM) / mm^2	Areal fraction of blood vessels (%)
d4	52.32±5.98	73.00±3.27	385.91±47.94	0.61±0.16*
d7	56.26±1.79	75.10±2.57	772.14±65.09*	8.13±3.08
d14	53.05±2.02	73.24±3.24	443.00±52.95	8.89±0.80

Table 1. Morphometric analyses on immunohistochemical staining. Results are presented as mean ± SD. *p<0.05

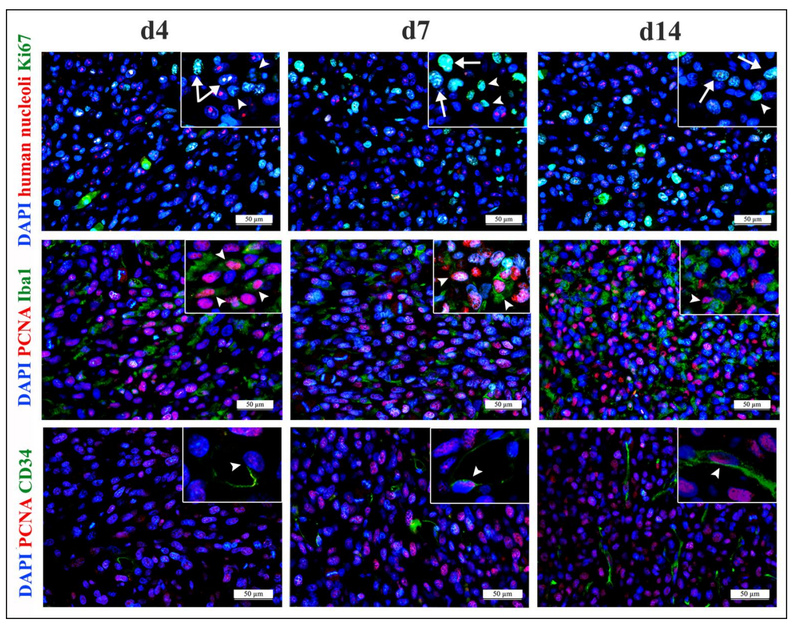

Figure 3. Immunofluorescence images showing colocalization of human nucleoli and Ki67– labelled cells (arrows indicate human nucleoli$^+$ Ki67$^+$ cells, while arrowheads indicate human nucleoli$^-$ Ki67$^+$ cells), PCNA and Iba1 – labelled cells (arrowheads indicate PCNA$^+$ Iba1$^+$ cells), and PCNA and CD34 – labelled cells (arrowheads indicate PCNA$^+$ CD34$^+$ cells).

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