J3T-1

Two cell lines, J3T-1 and J3T-2, were derived from the same parental canine glioma cell line, J3T. These cells were inoculated to establish brain tumors in athymic mice and rats. Pathologic samples of these animal gliomas were examined to analyze invasive patterns in relation to angiogenesis and were compared with human glioblastoma samples. The molecular profiles of these cell lines were also shown.

Histologically, J3T-1 and J3T-2 tumors exhibited different invasive patterns. J3T-1 cells clustered around newly developed vessels at tumor borders, whereas J3T-2 cells showed diffuse single-cell infiltration into surrounding healthy parenchyma. In human malignant glioma samples, both types of invasion were observed concomitantly. Molecular profiles of these cell lines were analyzed by immunocytochemistry and with quantitative reverse transcription-polymerase chain reaction. Vascular endothelial growth factor, matrix metalloproteinase-9, hypoxia-inducible factor-1, and platelet-derived growth factor were overexpressed in J3T-1 cells rather than in J3T-2 cells, whereas integrin αvβ3, matrix metalloproteinase-2, nestin, and secreted protein acidic and rich in cysteine were overexpressed in J3T-2 cells rather than in J3T-1 cells.

These animal models histologically recapitulated two invasive and angiogenic phenotypes, namely angiogenesis-dependent and angiogenesis-independent invasion, also observed in human glioblastoma. These cell lines provided a reproducible in vitro and in vivo system to analyze the mechanisms of invasion and angiogenesis in glioma progression [1)].

Onishi et al. established a pair of animal models (J3T-1 and J3T-2) with different invasive and angiogenic phenotypes and demonstrated that annexin A2 is expressed at higher levels in J3T-1 than J3T-2 cells. The function of annexin A2 in relation to angiogenesis and invasion was investigated using these models. Stable silencing or overexpression of annexin A2 in J3T-1 and J3T-2 cells (J3T-1shA and J3T-2A cells) was established and used. Thirty human glioblastoma samples were evaluated for expression of annexin A2, vascular endothelial growth factor (VEGF) and platelet-derived growth factor (PDGF). Immunohistochemical and quantitative reverse-transcription polymerase chain reaction analyses revealed higher expression of annexin A2, VEGF and PDGF in J3T-1 and J3T-2A cells. Cultured J3T-1 and J3T-2A cells exhibited higher adhesive ability to endothelial cells. Histopathological analysis of animal brain tumors revealed that J3T-1 and J3T-2A tumors displayed marked angiogenesis and invasion along the neovasculature, whereas J3T-2 and J3T-1shA tumors exhibited diffuse, infiltrative invasion without angiogenesis. Positive expression of annexin A2 was observed in tumor cells surrounding dilated vessels in 25/30 human glioblastoma specimens. Our results reveal that the phenotype of glioma invasion is closely related to angiogenesis. We identify annexin A2 as a factor regulating angiogenesis and invasion of malignant gliomas [2)].

The canine glioblastoma cell line J3T-1 was subcutaneously injected into a 6-week-old female BALB/c nude mice to obtain tumour fragments. Tumor fragments were implanted into adult male mongrel dog brains through surgery. Multiparametric MRI was performed with conventional MRI, diffusion-weighted imaging, and dynamic susceptibility contrast-enhanced perfusion-weighted imaging at one week and two weeks after surgery in a total of 15 surgical success cases. The presence of tumor cells, the necrotic area fraction, and the microvessel density (MVD) of the tumour on the histologic specimen

were assessed. Tumor volume, diffusion, and perfusion parameters were compared at each time point using Wilcoxon signed-rank tests, and the differences between tumor and normal parenchyma were compared using unpaired t-tests. Spearman correlation analysis was performed between the imaging and histologic parameters.

All animals showed a peripheral enhancing lesion on MRI and confirmed the presence of a tumor through histologic analysis (92.3%). The normalized perfusion values did not show significant decreases through at least 2 weeks after the surgery ($P > 0.05$). There was greater cerebral blood volume and flow in the Glioblastoma than in the normal-appearing white matter (1.46 ± 0.25 vs. 1.13 ± 0.16 and 1.30 ± 0.22 vs. 1.02 ± 0.14; $P < 0.001$ and $P < 0.001$, respectively). The MVD in the histologic specimens was correlated with the cerebral blood volume in the Glioblastoma tissue ($r = 0.850$, $P = 0.004$).

The results suggest that the canine Glioblastoma model showed perfusion imaging characteristics similar to those of humans, and it might have the potential as a model to assess novel technical developments for glioblastoma treatment [3].

1)
Inoue S, Ichikawa T, Kurozumi K, Maruo T, Onishi M, Yoshida K, Fujii K, Kambara H, Chiocca EA, Date I. Novel animal glioma models that separately exhibit two different invasive and angiogenic phenotypes of human glioblastomas. World Neurosurg. 2012 Dec;78(6):670-82. doi: 10.1016/j.wneu.2011.09.005. Epub 2011 Nov 7. PMID: 22120277.
2)
Onishi M, Ichikawa T, Kurozumi K, Inoue S, Maruo T, Otani Y, Fujii K, Ishida J, Shimazu Y, Yoshida K, Michiue H, Antonio Chiocca E, Date I. Annexin A2 regulates angiogenesis and invasion phenotypes of malignant glioma. Brain Tumor Pathol. 2015 Jul;32(3):184-94. doi: 10.1007/s10014-015-0216-6. Epub 2015 Feb 20. PMID: 25697644.
3)
Lee S, Choi SH, Cho HR, Koh J, Park CK, Ichikawa T. Multiparametric magnetic resonance imaging features of a canine glioblastoma model. PLoS One. 2021 Jul 9;16(7):e0254448. doi: 10.1371/journal.pone.0254448. PMID: 34242365.