Angiogenesis and Brain Oedema in Intracranial Meningiomas: Influence of Vascular Endothelial Growth Factor

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Summary

The correlation between angiographic neovascularization, peritumoural brain oedema (PTBOe) and the expression of vascular endothelial growth factor (VEGF), was analysed in 30 patients with intracranial meningiomas.

Pre-operative angiograms were examined for the existence of either an exclusively dural tumour blush or an additionally pial tumour supply from cerebral arteries. Furthermore the presence of macroscopic tumour-neovascularization and dysplastic changes of tumour-draining cerebral veins was evaluated. VEGF expression was investigated on histological tissue samples, using immunohistochemical techniques. VEGF immunohistochemistry and neuroradiological evaluations were performed in double blind fashion.

Tumour volume and the amount of oedema were calculated by computerized tomography (CT) or magnetic resonance imaging (MRI). The oedema-tumour volume ratio was defined as oedema index (OeI).

Compared to VEGF-negative meningiomas, tumours with striking VEGF staining revealed a significant higher mean oedema index (OeI = 4.2 vs. OeI = 1.5; p < 0.018), and a higher oedema incidence (91.7% vs. 44.4%; p < 0.046). Equally, meningiomas with additionally tumour supply from cerebral arteries were associated with a significant higher mean OeI (OeI = 4.1 vs. OeI = 1.2; p < 0.01) and oedema incidence (94.7% vs. 20.0%; p < 0.0023) than meningiomas with exclusively tumour supply from dural arteries. All meningiomas with striking VEGF-expression were associated with vascular tumour supply from cerebral arteries, but VEGF-negative tumours only in 50% (p < 0.029). These data suggest a link between VEGF-expression, arterial tumour supply and peritumoural brain oedema.

The development of tumour supply from cerebral arteries may be important for formation of meningioma-related oedema. Therefore, VEGF may represent a potent mediator in the evolution of this type of vascularization in meningiomas.

Keywords: Meningioma; vascular endothelial growth factor; pial supply; angiogenesis; brain oedema.

Introduction

Peritumoural brain oedema in meningiomas can aggravate clinical symptoms as well as adversely affect surgical outcome. The oedema-related space-occupying effect may compromise the adjacent brain by distortion or by reduction of cerebral blood flow, and result in an additional increase of intracranial pressure. Electron-microscopic investigations on meningiomas demonstrate that PTBOe is of vasogenic origin [12], and consequently results from a disturbed blood-brain-barrier [17]. According to the primarily extra-axial location of meningiomas, a dysfunction of the blood-brain-barrier, however is difficult to explain, and the pathophysiological mechanisms of oedema-genesis remain not clearly understood. Some pathogenetic hypothesis have already been discussed, like the size of the tumour [14, 36] with possible ischaemia [38], obstruction of the venous drainage [16] due to tumour compression or secretory activity of the tumour [27]. Some authors revealed a high correlation between pial tumour supply of meningiomas and the presence of brain oedema [3, 20]. Recent studies emphasize the importance of vascular endothelial growth factor (VEGF) for the development of tumour-angiogenesis [30, 33], especially in glioblastomas [42]. VEGF was found to be a specific mitogen of endothelial cells in-vitro [22, 25] and a potent promotor of angiogenesis in-vivo [43]. Furthermore, VEGF has an increasing effect on vascular permeability and is identical with the vascular permeability factor (VPF) [22, 25]. VEGF is a homodimeric glycoprotein of 36-46 kD. Molecular cloning of complement DNA (cDNA) revealed 4 types of VEGF in human cells so far. These forms are the result of alternative splicing of messenger ribonucleic acid (mRNA) [18, 40] and are called VEGF121, VEGF165, VEGF189 and VEGF206. The most common and best characterized form is VEGF165, which is a secretable and heparin-binding isoform [9].
This study investigates the relationship between the immunohistochemical VEGF-expression, the angiographic type and degree of arterial tumour supply and the extent of peritumoural brain oedema.

Patients and Methods

Patients

The pre-operative cerebral angiograms, computerized tomographies (CT) or magnetic resonance imaging (MRI) from 30 patients with intracranial meningiomas were evaluated, and immunohistochemical VEGF analysis of surgical specimens was performed. The study included 7 men and 23 women. The mean age was 55.4 years (range 32–79 years). All patients underwent microsurgical removal of the tumour at our facility. The location of the meningiomas is summarized in Table 1. Using criteria of the new World Health Organization (WHO) classification [24], 29 tumours were graded as typical meningiomas (WHO I) and 1 as atypical meningioma (WHO II).

Evaluation of Angiography

The indication for cerebral angiography was based exclusively on clinical aspects, for example to rule out vascular anomalies or to study peritumoural veins, in particular dural sinuses. DSA-technique was used for angiographies and depending on tumour location selective injections of the common carotid, the internal and external carotid, and the vertebral arteries were performed. MRI or CT scans were performed with or without contrast medium in all patients.

Angiographies had to differentiate between exclusively dural or additional pial blood supply from cerebral arteries. A pial vascular supply was suggested, when the dural blush could be separated from the pial one, or, cerebral arteries were directed towards the tumour. Furthermore, angiographically the existence of an exclusive tumour blush was distinguished from a macroscopic tumour-vascularization. An exclusive tumour blush was assumed, if the meningiomas were associated with a diffuse angiographic contrast-staining and additionally only the mean feeding arterial vessels (for example A. meningea media) and their first junction could be proven. In meningiomas with macroscopic tumour-vascularization additional branches of arterial tumour vessels had to be obvious like vessels distal to the 2nd-4th junction. In venous phase of angiography, vessels at the tumour margin were examined for dysplastic venous changes, such as ectasia and distortion of veins with variance in diameter. The resulting volumes of tumour and oedema were then approximated using the formula for a spheroid:

\[ V = \frac{4}{3}\pi \times abc. \]

The relation between PTBOε and tumour volume (oedema index, OεI) was defined as

\[ OεI = \frac{V_{\text{oedema}} + V_{\text{Tumour}}}{V_{\text{Tumour}}}. \]

An OεI = 1 indicates, that no brain oedema was found.

Immunohistochemical Evaluation of VEGF

The tissue samples were snap-frozen in liquid nitrogen at −80°C immediately after operative removal. Subsequently 5 µm thick cryostat-sections were stained using the peroxidase-anti-peroxidase (PAP) method [35] using polyclonal anti-VEGF antibodies (VEGF Ab-1 and VEGF Ab-2, Calbiochem, Cambridge, Massachusetts). Antibody dilution was 1:10. Negative controls consisted of phosphate buffered saline instead of first antibodies. Evaluation of the immunohistochemical staining was carried out independently by two observers. The staining was semiquantitatively assessed to determine both the percentage of labelled cells and the intensity of the immunohistochemical reaction (Fig. 1). The degree of VEGF-staining in meningioma specimens was graduated as follows: 1. negative, if no cells were labelled, 2. weak, if less than about 20% of cells were labelled and 3. strong, if more than about 20% of cells were labelled.

Statistical Evaluation

Oedema indices and incidences of pial blush, oedema and VEGF-expression were statistically evaluated using the Wilcoxon-test and the Fisher’s Exact-test (χ²-test), respectively. All calculations were performed using a statistical analysis system (SAS-System, SAS Institute Inc, Cary NC).

Results

Tumour Vascularization and Oedema

5 meningiomas had only dural supply by external carotid branches, 19 cases revealed an additional pial supply from cerebral arteries (Fig. 2). Unequivocal differentiation between dural and pial vascular supply was not possible in 6 meningiomas. Tumours with pial supply showed a significant higher mean OεI (OεI =