Association Between Immunohistochemical Expression of Vascular Endothelial Growth Factor (VEGF) and Fibroblast Growth Factor-2 (FGF-2) with Histopathology Grade of Meningioma

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***Abstract*-** Meningioma is one of the intracranial primary tumors that often occurs with the incidence in the United States covering 36% of all tumors in the central nervous system. The growth of solid tumors such as meningiomas is highly dependent on angiogenesis. Vascular Endothelial Growth Factor (VEGF) and Fibroblast growth Factor-2 (FGF-2) are angiogenic factors that mediate positive regulation for angiogenesis. Both VEGF and FGF-2 examinations in previous meningioma have been carried out, but using blood serum samples. This research was conducted using a different method that is immunohistochemically from tumor tissue to assess the association between VEGF and FGF-2 expression with histopathology grade among meningioma patients. Formalin-fixed paraffin-embedded tissue blocks of 32 meningioma patients were immunohistochemically studied for VEGF and FGF-2 expression. The association between VEGF and FGF-2 expression with grade were anylized using SPSS 22 version. Both VEGF and FGF-2 expressions were significantly associated with grade of meningioma ($p < 0.05$). Most negative FGF-2 cases were in grade 1 meningioma (91.6%) and positive FGF-2 were found in 50% in grade 2 meningiomas. While for VEGF expression, although most cases for each VEGF value were in grade 1 meningiomas, but all cases of grade 3 meningioma have high VEGF expression. The positive expression of FGF-2 and the high of VEGF indicate that these two growth factors play a very important role in angiogenesis which triggers cell proliferation, especially for meningioma with higher grade. This can be a prognostic factor and a therapeutic opportunity when surgical intervention and chemotherapy become inadequate management.

***Index Terms*-**VEGF, FGF-2, meningioma, grade, immunohistochemical

I. Introduction

Meningiomas grow from arachnoid cap cells, which are the outer layers of arachnoid material with slow growth.[1,2] The average age of meningioma patients is 65 years and the risk will increase with age and the ratio between women and men 3: 2.[1,3] Although most meningiomas are encapsulated and histologically benign, some of these tumor sites cause very serious and potentially fatal.[4] The fourth revised edition of the World Health Organization (WHO) in 2016 has classified and divided the grade of meningiomas into three, grade 1 as benign (75% of cases), grade 2 as atypical (10-15%) and grade 3 as malignant (1-3%).[1] Higher grade meningiomas show a tendency to be progressive and recurrent.[5] Reported recurrence rates for each grade 1, 2, and 3 meningioma are 7-25% , 29-52%, and 50-94% respectively. Depending on location and grade, treatment options for meningiomas include surgery and postoperative radiation therapy.[6,7]

Vascular Endothelial Growth Factor (VEGF) is a growth factor that has a major role in neovascularization, increased vascular permeability, and peritumoral brain edema.[8,9] Besides VEGF, Fibroblast growth Factor-2 (FGF-2) or also called basic Fibroblast Growth Factor (bFGF) is also one of the angiogenic factors that mediate positive regulation for the angiogenesis.[10] Angiogenic factors such as FGF and VEGF stimulate endothelial cells to secrete several proteases and plasminogen activators, resulting in degradation of the basal membrane of blood vessels, which in turn may allow cell to attack the surrounding matrix. Cells migrate, proliferate, and eventually differentiate to form new lumen containing blood vessels.[11]

At present, several studies have shown promising targeting of angiogenesis regulated by VEGF in meningiomas, especially in patients with recurrent or higher grade meningiomas. Bevacizumab (VEGF inhibitors) as monoclonal antibodies for VEGF receptors can disrupt the binding transduction and signals needed for tumor vascularization which in turn causes regression of tumor blood supply.[12] However, there are several clinical studies that show resistance for VEGF inhibitor. Therefore, there are still many further studies regarding other promising target therapies besides using VEGF inhibitors.[13]

Both VEGF and FGF-2 have been widely studied in several tumors. For meningiomas themselves, examinations of these two angiogenic factors have been carried out using the ELISA method that uses a patient's blood sample. In this study, we interested examining FGF-2 and VEGF at the same time by using a different method that is immunohistochemistry to see its

expression in each histopathological grade of meningioma from the tumor tissue.

II. MATERIAL AND METHODS

Sample selection

This cross sectional study was conducted in Department of Anatomical Pathology, Universitas Sumatera Utara/ H. Adam Malik General Hospital, Medan and includes 32 cases of meningioma. All samples were obtained through surgical procedure. Inclusion criteria were meningioma cases with adequate clinical data, available and formalin-fixed paraffin embedded tissue block with sufficient tumor tissue. Detailed clinical data were obtained from medical records or pathology archives consisting of age, sex, and location of the tumor. Histological type and grade were determined independently by researchers through hematoxylin and eosin stained slides examination.

Immunohistochemistry protocol and interpretation

The tissue sections were deparaffinized and rehydrated before pretreatment. Endogenous peroxidase was blocked with hydrogen peroxide followed by antigen retrieval. VEGF rabbit monoclonal and FGF-2 (GTX84502, GeneTex, California, America) mouse monoclonal antibodies were used as primary antibody. Diagnostic BioSystems (Diagnostic BioSystems, Pleasanton, CA, USA) polymer kit was used for detection. The reaction was visualized with diaminobenzidine and counterstained with Mayer's hematoxylin followed by dehydration, clearing, and mounting. Positive control were hepar for VEGF and colon for FGF-2. VEGF and FGF-2 expressions were determined independently by researchers. The expression in cytoplasm was analyzed. Immunostaining of VEGF and FGF-2 was evaluated in terms of the proportion and staining intensity of tumor cells. VEGF expression was determined using Histo-score (H-score) with a range of possible scores from 0 to 300. H-score 0-19 was considered low, 20-99 was moderate, and 100-300 was considered high expression. While for FGF-2, the proportion was assessed on the basis of the percentage of immunopositive cells as follows: 0, less than 10%; +1, 10-25%; +2, 26-50%; +3, 51-75%; +4, greater than 75%. Staining intensity was evaluated as negative (0), weak (+1), moderate (+2), and strong (+3). The proportion score (0-4) was multiplied by the intensity score (0-3) and a final score was assigned, 0-4 as negative staining and 5-12 as positive staining.

Statistical analysis

Statistical analysis was performed using SPSS software package version 22.0 (SPSS Inc., Chicago) and Microsoft Excel 2010. Categorical variables were presented in frequency and percentage. Mann-Whitney, Kruskal-Wallis, and Dunn test were applied to find out the association between VEGF and FGF-2 expressions with histopathology grade of meningioma. The p-values < 0.05 were considered significant.

III. RESULT

Patients' characteristics

The mean age for meningioma patients was 44.5 (±11.4) years. Twenty-three patients (71.9%) were females, only 9 patients (28.1%) were males. All the tumors were located in intracranial where convexity was the predominance. The number of the patients with WHO Grade 1 meningioma was 25 (78.1%), with Grade 2 meningioma was 5 (15.6%), and with Grade 3 meningioma was 2 (6.3%). The histological subtypes of meningioma varied and meningothelial meningioma was the majority of this case. Clinical basic characteristic of meningioma patients were summarized in table 1. Representative H&E sections are shown in figure 1.

Table 1. Characteristic of meningioma patients

Characteristics	Number of cases	Percentage (%)
Age, mean ± SD, years	44.5 ± 11.4	
Sex		
Female	23	71.9
Male	9	28.1
Location		
Supratentorial	28	87.5
Convexity	23	82.1
Parietal	8	34.8
Frontal	4	17.4
Temporal	1	4.3
Temporoparietal	6	26.1
Frontoparietal	3	13
Parietoccipital	1	4.4
Sphenoid	2	7.1
Parasagital	1	3.6
Suprasella, parasella	2	7.1
Infratentorial	4	12.5
Cerebellum	1	25
Foramen Magnum	1	25
CPA	2	50
Subtype		
Meningothelial	11	33.4
Fibroblastic	4	12.5
Transtitional	5	15.6
Psammomatous	2	6.3
Angiomatous	1	3.1
Microcyst	2	6.3
Chordoid	1	3.1
Clear cell	1	3.1
Atypical	3	9.3
Anaplastic	2	6.3
Histological grade		
Grade 1	25	78.1
Grade 2	5	15.6
Grade 3	2	6.3

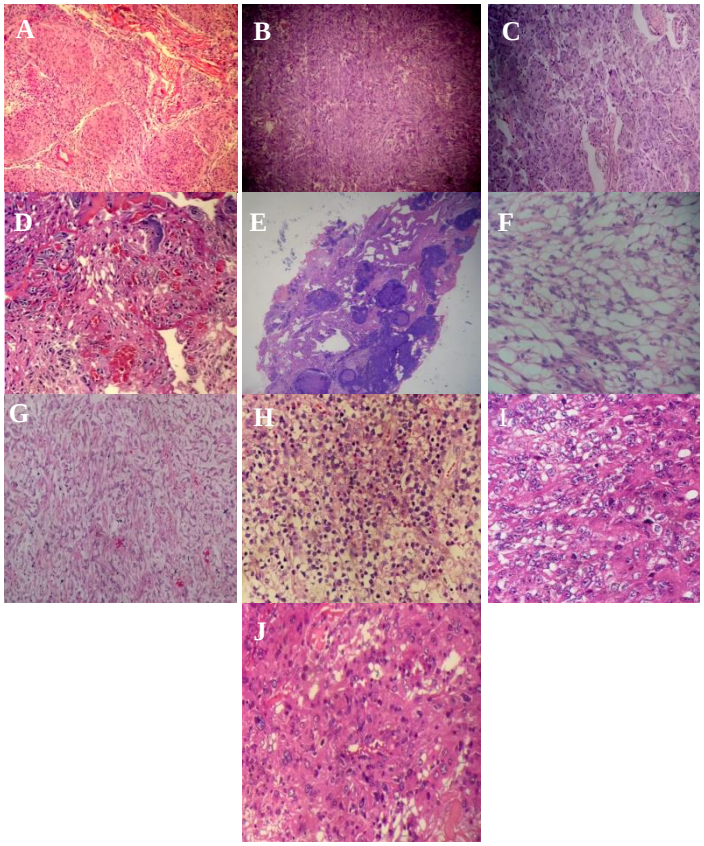

Figure 1. Histological type. A, Meningothelial meningioma. B, Fibroblastic meningioma. C, Transtitional meningioma. D, Angiomatous meningioma. E, Psammomatous meningioma. F, Microcyst meningioma. G, Chordoid meningioma. H, Clear cell meningioma. I, Atypical meningioma. J, Anaplastic meningioma.

VEGF expression

Sixteen out of 32 meningiomas show low score for VEGF, 8 cases for moderate and also high score. In meningiomas grade 1, most of cases were low score but all of meningiomas grade 3 express VEGF in high score (table 2).

Table 2. Association of VEGF expressions with histopathology grade

VEGF	Grade						p-value
	Grade 1		Grade 2		Grade 3		
	n	%	n	%	n	%	
Low	13	81.2	3	18.8	0	0	
Moderate	8	100	0	0	0	0	0.039
High	4	50.0	2	25.0	2	25.0	

***Kruskal-Wallis test**

Generally there was a significant p-value between score VEGF and histopathology grade of meningioma. But based on post hoc test, there is no significant in group of low VEGF versus moderate VEGF (table 3). The intensity of VEGF expression in cytoplasm are shown in figure 3.

Table 3. Post hoc test in each value of VEGF

Post Hoc	p-value*
Low vs Moderate	0,168
Low vs High	0,029
Moderate vs High	0,007

***Dunn test**

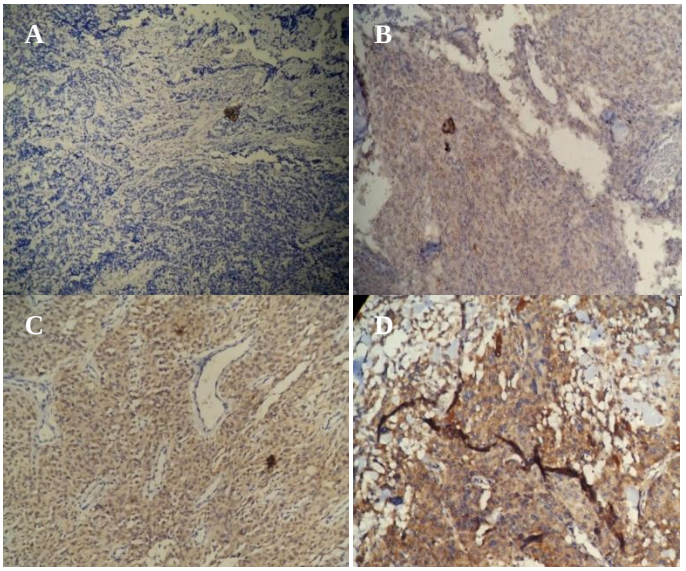

Figure 3. Immunohistochemical VEGF expression. A, Negative intensity. B, Mild intensity. C, Moderate intensity. D, Strong intensity.

FGF-2 expression

Twenty-four out of 32 meningioma cases were negative for FGF-2 and most cases are in grade 1, while positive expression was seen in eight cases and 50% cases are in grade 2 (table 4). The intensity of FGF-2 expression in cytoplasm are shown in figure 3.

Table 4. Association of FGF-2 expressions with histopathology grade

FGF-2	Grade						p-value*
	Grade 1		Grade 2		Grade 3		
	n	%	n	%	n	%	
Negative	22	91.6	1	4.2	1	4.2	0.002
Positive	3	37.5	4	50.0	1	12.5	

***Mann-Whitney test**

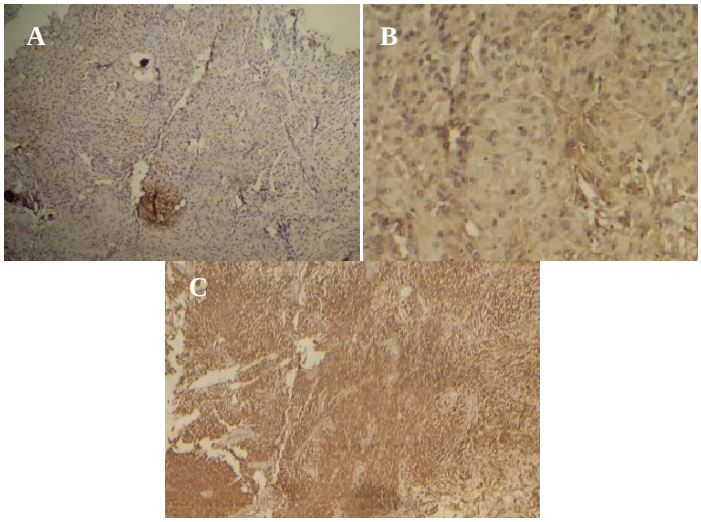

Figure 3. Immunohistochemical FGF-2 expression. A, Negative intensity. B, Mild intensity. C, Moderate intensity.

IV. DISCUSSION

Meningiomas account for 25-30% of all primary tumors in the central nervous system, are slow-growing tumors, most common in middle-ages and elderly with a peak incidence of the sixth to seventh decades.[1] The prevalence rate in the United States confirmed by pathology examination of 97.5 per 100,000 populations.[4]

Several studies have also reported an association between meningioma and VEGF expression, with different results. Although higher vascularization is found in high-grade tumors, the correlation between histological grade and VEGF expression in meningiomas is still unclear. Most brain tumors secrete excessive VEGF, which leads to tumors with abnormal vascular permeability. This hyperpermeability allows fluid to leak from the intravascular space to the brain parenchyma causing an increase in interstitial fluid pressure and vasogenic cerebral edema. A positive correlation between VEGF and edema has been found in a number of previous studies.[8,14-16]

VEGF is a regulator of vascularity, angiogenesis, and vascular permeability. This may play an important role in the formation of peritumoral brain edema (PTBE) associated with meningioma.[15] High VEGF expression and the presence of PTBE have been proposed as predictors of recurrence in benign meningioma (grade 1).[17] There were 4 cases of grade 1 meningioma in this study with a high VEGF score, but the PTBE status was not evaluated by the author and no long-term observation was carried out on these four patients, so it cannot be predicted whether the recurrence in these patients is increasing.

Some studies suggest that VEGF expression correlates with meningioma vascularization while others do not. One of the reasons for this difference is the involvement of angiomatous meningiomas and meningiomas with higher grade, such as atypical and anaplastic meningiomas. Angiomatous meningiomas usually show the highest microvascular density (MVD), but VEGF levels are not always the highest. Higher grade meningiomas show an increase in VEGF without an increase in MVD. Lamszus, et al. reported that VEGF is contained in protein extracts from human meningioma tissue induced capillary-like tube formation and in vitro endothelial cell migration. These findings indicate that VEGF is involved in angiogenesis in meningiomas, but increased VEGF expression does not result in an increase in the number of blood vessels with an increase in histological grade.[9]

Meanwhile, in order to grow bigger and attack the surrounding tissues as well as grade 2 and 3 meningiomas, tumors must be able to induce new blood vessel growth. New blood vessel growth is a regular process that depends on coordinated signaling by growth factors and cell adhesion receptors, one of which is VEGF.[17] This is illustrated by the significant correlation between moderate and high VEGF expression and grade in this study.

VEGF expression in meningiomas has been shown to vary with respect to grade and histological subtype. Research by Dharmalingam, et al. note that no significant association can be shown in all grade of meningiomas. In addition, the level of expression as described by VEGF assessment and expression patterns (focal or diffuse) did not differ significantly between grades. The study noted that transitional meningioma had a high VEGF positivity (81.8%), followed by meningothelial meningioma (66.7%), while fibroblastic meningioma showed the least (12.5%). Angiomatous meningiomas express very high VEGF expressions because of the features inherent in this subtype. Denizot, et al. showed, by quantitative analysis of VEGF by ELISA, that fibroblastic meningiomas showed lower VEGF level compared to meningothelial and transitional subtypes.[18] While Mahzouni, et al. explained that there was a significant relationship between VEGF expression and grade but there was no significant relationship between VEGF expression and subtype.[19] In this study, there was a relationship between VEGF expression and grade, while no analytical test was carried out between VEGF expression and subtype because not all meningioma subtypes were found. However, it can be seen that from 4 cases of meningioma grade 1 samples there was a high VEGF score, for 1 case of angiomatous subtype, 1 fibroblastic, and 2 cases of transitional subtypes.

VEGF expression is also correlated with survival progression where it is concluded that VEGF is significantly correlated with tumor development and recurrence.[16] This is seen in the case of grade 3 meningioma in this study. All cases of grade 3 meningioma express high VEGF because indeed the grade increases, the higher the rate of recurrence of this neoplasm. But the limitations of this study, no long-term observation of these meningioma patients will be done whether they will experience a recurrence or even previously this patients had a history of meningioma disease.

Significant FGF-2 levels and receptors, FGFR1 and FGFR2 have been detected in meningiomas. FGF becomes a mitogen, differentiation and angiogenic agent that can influence tumor development as an autocrine growth factor.[20] FGF exerts extensive mitogenic activity by stimulating the growth of fibroblasts, endothelial cells and cancer cells. FGF-2 is an important regulator of cell growth and differentiation in physiological and pathological conditions.[21] FGF-2 is present in any part of the body and has a strong affinity for glycosaminoglycans such as sulfate deposits. FGF-2 is stored by binding of glycosaminoglycans to the cell surface or extracellular matrix. When FGF-2 is actually needed, the cell secretes various

enzymes such as heparinase that release FGF-2 from glycosaminoglycans so that it can reach the target cell. Previous research has suggested the role of FGF-2 as a prognostic marker for various types of malignancies.[22]

The relationship between FGF-2 and histopathological grade of meningioma has also been investigated by Risfandy, et al. but using a patient's blood serum sample and not from the tumor tissue. From the analytical test it was found that there was no significant relationship between this two growth factors.[23] In contrast to the immunohistochemical studies in this study, a significant association was found between FGF-2 expression and histopathological grade of meningioma. This may be due to the different number and distribution of samples. Risfandy only classified meningioma grades into 1 and 2, while the authors divide the grade according to the WHO classification of 1, 2, and 3. Although considered not significant association, Risfandy et al revealed that higher serum FGF-2 levels were found in meningiomas with higher grade. This can show that FGF-2 as a growth factor plays a role in cell proliferation and angiogenesis more in higher grade meningiomas. Angiogenesis itself is controlled by a balance of promoters and inhibitors. FGF-2 has received attention as another very potential angiogenic growth factor beside VEGF. Although the detailed mechanism of the relationship between FGF signals and the onset of the emergence of malignant tumors is still unclear. However, at this time, mutation of FGF receptor was assumed to activate the FGF signal constantly and ultimately encourage the growth and metastasis of tumor cells.[22]

This study used FGF-2 immunohistochemical techniques in meningioma cases where most meningiomas are benign (grade 1). Denizot, et al. also carried out analysis of FGF-2 levels through tumor tissue samples, but used the quantitative ELISA method. Based on the results obtained, there was no significant association in FGF-2 levels among the subtypes of meningioma grade 1.[24] Study of Baritaki, et al. also evaluated FGF-2 from tissue samples through qRT-PCR examination for cases of brain tumors including glioma, meningioma, and metastasis. All cases of meningioma show higher expression compared to normal brain tissue. But unfortunately in their study, all cases of meningiomas are included in the benign group, so it cannot be seen in comparison with cases of higher grade meningiomas.[25] In this study although most cases of meningiomas gave negative expression results in 24 of 32 cases and did not take normal brain tissue samples for comparison, but there is a significant association between FGF-2 expression and histopathological grade of meningioma.

It is also important to think that in most studies, serum and intratumoral levels of FGF-2 were associated with decreased survival rate. High intratumoral and serum FGF-2 levels are also associated with recurrence of various types of cancer such as lung, bladder, breast, esophagus and Hodgkin Lymphoma.[21,26] Donnem, et al. explained in their study that high FGF-2 expression in the NSCLC case had a 37% rate for 5 years of survival, while for low FGF-2 expression, survival rate had increased to 59% .[26]

Both FGF-2 and VEGF expressions in this study had significant association with histopathological grade of meningioma. Synergistic action of FGF and VEGF has been observed and crosstalk between these two families has been suggested and considered an important arrangement in the formation of blood vessels and this mechanism has long been a basic question in vascular biology that is not understood. Data from the Murakami study, et al. shows how these two angiogenic growth factors regulate the neovascularization process in a coordinated way.[27]

VEGF and FGF-2 show the same MVD in tumor xenografts, but tumor growth rates induced FGF-2 were significantly higher than VEGF in vivo. These findings indicate that abnormal leakage in tumor blood vessels can affect tumor growth, supplying oxygen and nutrients in insufficient quantities. On the other hand, blood vessels were induced by FGF-2 allow tumors to access an adequate supply of oxygen and nutrients and the disposal of waste products; as a result, tumors can continue to grow with a high growth rate. However, abnormal leakage of blood vessels induced by VEGF may be useful for effectively delivering large molecular size anticancer drugs into tumors due to increased endothelial fenestration with low pericyte protection.[28]

Beside interactions with the VEGF, FGF-2 can control the function of growth factors and other chemokines, such as PDGF, HGF, and MCP-1. Although the exact mechanism of this interaction remains unclear, it is interesting to speculate that FGF-2 can modulate many neovascularizing processs.[27] VEGF and FGF-2 play a unique role in synergistic enhancement of endogenous PDGF-B-PDGFRß signaling, to promote the formation of mature blood vessels in addition to the long-known mitogenic effects.[28]

In many experimental trials, angiogenesis-driven FGF-2 is blocked by VEGF inhibition, which shows that FGF-2 controls the upstream VEGF angiogenesis by modulating VEGF function. This hierarchical regulation seems to play a similar role in lymphangiogenesis, as lymphatic growth induced by FGF-2 is inhibited by the blockade of VEGFR3 signaling.[28] Although VEGF inhibitor therapy can cause vascular disintegration and regression not only in many experimental studies, but also in human clinical trials, blood vessels rapidly rebound when VEGF inhibition is withdrawn.[27] In normal tissue, while VEGF-induced vessels contain high endothelial fenestration that mediate high permeability, blood vessels induced by FGF-2 do not have vascular fenestration.[28] The combination of inhibition of FGF-2 and VEGF signals may have a much deeper effect on blood vessels, because a rebound in vascular growth may not occur without VEGFR2 expression. FGF-2 signaling is indispensable for the maintenance of VEGFR2 expression, and its inhibition has a profound effect on biological processes that depend on VEGF.[27] Since recent years, the FGF-2 signaling pathway has been stated to contribute in tumor angiogenesis, cell proliferation, and resistance to chemotherapy and VEGF inhibitor therapy for several types of human cancer in preclinical and clinical studies.[28]

V. CONCLUSION

There is significant association between VEGF and FGF-2 expression with histopatholgy grade of meningioma.

COMPETING INTERESTS

The authors have no relevant financial interest in the products or companies described in this article.

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ETHICAL APPROVAL

Health Research Ethical Committee, Universitas Sumatera Utara, Medan, Indonesia approved this study.

REFERENCES

[1] Perry A, Louis DN, Budka H, Deimling AV, Sahm F, Rushing EJ, et al. Meningioma. In: Louis DN, Ohgaki H, Wiestier OD, Cavenee WK, Ellison DW, eds. WHO Classification of Tumours of the Central Nervous System. Lyon: IARC; 2016. pp. 232-45.

[2] Fathi AR, Roelcke U. Meningioma. Curr Neurol Neurosci Rep. 2013; 13: 337.

[3] Hortobagyi T, Benczi J, Varkoly G, Kouhsari MC, Klekner A. Meningioma Recurrence. Open Med. 2016; 11: 168-73.

[4] Wiemels J, Wrensch M, Claus EB. Epidemiology and Etiology of Meningioma. J Neurooncol. 2010; 99: 307-14.

[5] Abbritti RV, Polito F, Cucinotta M, Giudice CL, Caffo M, Tomasello C. Meningiomas and Proteomics: Focus on New Potential Biomarkers and Molecular Pathways. Cancer Genomics and Proteomics. 2016; 13: 369-80.

[6] Sharma P, Katiyar V, Sharma R, Gurjar HK, Krishnan S. Role of Tyrosine Kinase Inhibitors in Recurrent Meningiomas: Controversies and Promises. Neurosurgey. 2018; 82(6): 181-3.

[7] Shibuya M. Pathology and Molecular Genetics of Meningioma: Recent Advances. Neurol Med Chir. 2015; 55(1): 14-27.

[8] Schmid S, Enein FA, Pfisterer W, Birkner T, Stadek C, Knosp E. Vascular Endothelial Growth Factor: The Major Factor for Tumor Neovascularization and Edema Formation in Meningioma Patients. Neurosurgery. 2010; 67(6): 1703-8.

[9] Sakuma T, Nakagawa T, Ido K, Takeuchi H, Sato K, Kubota T. Expression of vascular endothelial growth factor-A and mRNA stability factor HuR in human meningiomas. J Neurooncol. 2008; 88: 143-55.

[10] Vujasinovic T, Buta M, Markicevic M, Vukosavljevic D. Angiogenesis: bFGF and VEGF in Breast Carcinoma. Arch Oncol. 2006; 14(3-4): 126-30.

[11] Cross MJ, Welsh LC. FGF and VEGF function in angiogenesis: signalling pathways,biological responses and therapeutic inhibition. TRENDS in Pharmacological Sciences. 2001; 22(4): 201-7.

[12] Franke AJ, Skelton WP, Woody LE, Bregy A, Shah AH, Vakharia K, et al. Role of bevacizumab for treatment-refractory meningiomas: A systematic analysis and literature review. Surgical Neurology International. 2018; 9(133): 1-10.

[13] Lieu C, Heymach J, Overman M, Tran H, Kopetz S. Beyond VEGF: Inhibition of the Fibroblast Growth Factor Pathway and Antiangiogenesis. Clin Cancer Res. 2011; 17(19): 1-10.

[14] Retnani DP, Fauziah D. Ekspresi Vascular Endothelial Growth Factor (VEGF) Dikaitkan dengan Pertumbuhan Tumor dan Edema Peritumoral. Majalah Patologi. 2013; 22(3): 24-9.

[15] Ding YS, Wang HD, Tang K, Hu ZG, Jin W, Yan W. Expression of Vascular Endothelial Growth Factor in Human Meningiomas and Peritumoral Brain Areas. Annals of Clinical and Laboratory Science. 2008; 38(4): 344-51.

[16] Reszec J, Hermanowicz A, Rutkowski R, Turek G, Mariak Z, Chyczewski L. Expression of MMP-9 and VEGF in Meningiomas and Their Correlation with Peritumoral Brain Edema. BioMed Research International. 2015; 1-8.

[17] Salokorpi N, Yrjana S, Tuominen H, Karttunen A, Haljasvaara R, Pihlajaniemi T, et al. Expression of VEGF and Collagen XVIII in Meningiomas: Correlations With Histopathological and MRI Characteristics. Acta Neurochir. 2013; 155: 989-96.

[18] Dharmalingam P. Vascular Endothelial Growth Factor Expression and Angiogenesis in Various Grades and Subtypes of Meningioma. Indian J pathol Microbiol. 2013; 56(4): 349-54.

[19] Mahzouni P, Aghili E, Sabaghi B. An Immunohistochemical Study of Vascular Endothelial Growth Factor Expression in Meningioma and Its Correlation with Tumor Grade. Middle East Journal of Cancer. 2018; 9(4): 288-94.

[20] Perry A. Tumours of the Meningens. Chapter 36 Greenfields Neuropathology 9th edition Love S, Perry A, Ironside J, Budka H. USA: CRC Press; 2015. pp. 1803-22.

[21] Akl MR, Nagpal P, Ayoub NM, Tai B, Sathyen A. Prabhu SA, et al. Molecular and Clinical Significance of Fibroblast Growth Factor 2 (FGF2/bFGF) in Malignancies of Solid and Hematological Cancers for Personalized Therapies. Oncotarget. 2016; 7(28): 44735-62.

[22] Jibiki N, Saito N, Kameoka S, Kobayashi M. Clinical Significance of Fibroblast Growth Factor (FGF) Expression in Colorectal Cancer. Int Surg. 2014; 99: 493-9.

[23] Risfandi M, Tandean S. Fibroblast Growth Factor 2 (Fgf-2) Serum Related Relationship With The Degree Of Intracranial Meningiomas Patients In Haji Adam Malik Hospital, North Sumatera. International Journal of ChemTech Research. 2017; 10(13): 369-73.

[24] Denizot Y, Armas RD, Caire F, Moreau JJ, Pommepuy I, Truffinet V, et al. The Quantitative Analysis of bFGF and VEGF by Elisa in Human Meningiomas. Hindawi Publishing Corporation. 2006; :1-3.

[25] Baritaki S, Chatzinikola AM, Vakis AF, Soulitzis N, Karabetsos DA, Neonakis I, et al. YY1 Over-Expression in Human Brain Gliomas and Meningiomas Correlates with TGF-ß1, IGF-1 and FGF-2 mRNA Levels. Cancer Investigation. 2009; 27: 184-92.

[26] Donnem T, Al-Shibli K, Al-Saad S, Busund LT, Bremnes RM. Prognostic Impact of Fibroblast Growth Factor 2 in Non-small Cell Lung Cancer. Journal of Thoracic Oncology. 2009; 4(5): 578-85.

[27] Murakami M, Nguyen LT, Hatanaka K, Schachterle W, Chen PY, Zhuang ZW, et al. FGF-dependent Regulation of VEGF Receptor 2 Expression in Mice. The Journal of Clinical Investigation. 2011; 121 (7): 2668-78.

[28] Hori Y, Ito K, Hamamichi S, Ozawa Y, Matsui J, Umeda IO. Functional Characterization of VEGF- and FGF-induced Tumor Blood Vessel Models in Human Cancer Xenografts. Anticancer Research. 2017; 37: 6629-38.

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