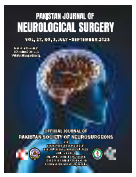

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Original Article

EGFR-1 Expression in Meningioma: Insights into Gender Distribution and Grade

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ABSTRACT

Objective: Meningioma is the most common primary brain. Histopathological grading (from I to III) is essential for prognostication and therapeutic decisions. EGFR-1 overexpression has been implicated in tumor progression and poor outcomes in various cancers. However, its role in meningioma pathophysiology and gender disparities remains unclear. This study aimed to determine the expression of EGFR-1 in grade I and grade II meningioma and explore associations with clinicopathological parameters.

Materials & Methods: This cross-sectional study was conducted at Ayub Teaching Hospital in collaboration with the Neurosurgery Department, Histopathology Laboratory of Ayub Medical College, and Health Diagnostic Laboratory. Non-probability consecutive sampling technique was used to include meningioma patients aged 23-54 years after obtaining ethical approval. Data was collected from medical records, and histopathological examination and immunohistochemical analysis were performed on meningioma tissue samples. Statistical analysis was conducted using SPSS Version 25, and associations between clinicopathological parameters and EGFR-1 expression were evaluated using appropriate tests.

Results: We included 39 meningioma patients, with a female predominance of 22 (56.4%). The majority had grade I tumors (89.7%). EGFR-1 expression was observed as severe 30 (76.9%), moderate 7 (17.9%), and mild 2 (5.1%). No significant association was found between gender and meningioma grade or EGFR-1 expression. However, a significant association was observed between meningioma grade and EGFR-1 expression in grade I tumors ($p = 0.049$), but not in grade II tumors ($p = 0.248$).

Conclusions: A significant correlation between meningioma grade and EGFR-1 expression in Grade I meningioma suggests that EGFR expression could serve as a prognostic biomarker.

Keywords: Meningioma, Histopathology grade, EGFR-1 expression, Immunohistochemistry, Intracranial tumors.

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INTRODUCTION

Meningioma represents the predominant type of primary neoplasms affecting the meninges covering the brain and spinal cord, comprising roughly a quarter of all intracranial tumors. Meningioma is a non-malignant neoplasm originating from the meninges- the protective membranes enveloping the brain and spinal cord.[1,2]Meningioma is categorized into three distinct grades (from I to III) predicated on the extent of the neoplastic changes in the affected cells.[3] The tumors exhibit a diverse range of clinical behaviors, with some displaying a benign and slow-growing nature (grade I), while others exhibit atypical features (grade II) and an elevated risk of recurrence (grade III).[4] The assessment of histopathological grading is of utmost importance for prognostication and therapeutic planning for the patients.[5] The annual occurrence rate of meningioma is estimated to be between 5 and 8 cases per 100,000 individuals.[1] The incidence of meningioma is higher in females than males, and the demographic group most susceptible to this condition is individuals aged 50 years and above.[1,6,7] Meningioma has the potential to result in fatality in certain instances.[1,6,7]

Typically, the identification of meningioma entails a confluence of diagnostic procedures, including imaging modalities like magnetic resonance imaging (MRI) and computed tomography (CT) scans, as well as biopsy after surgical excision of the neoplasm.[1,3] The selection of treatment modalities is dependent upon the grade and site of the neoplasm and may encompass surveillance, surgical intervention, radiotherapy, or a hybrid approach.[8] Meningiomas of minute size and low-grade nature may be subject to surveillance without intervention. Surgical intervention, radiation therapy, or chemotherapy may be necessary for tumors of greater size or higher grade.[6,7]

The transmembrane protein, epidermal growth factor receptor (EGFR), is involved in cellular processes such as growth, proliferation, and survival.[9] Upon binding with EGF, EGFR-1 undergoes activation and initiates a series of intracellular processes that culminate in cellular proliferation and growth.[10, 11] The upregulation of EGFR has been associated with augmented cellular proliferation and growth, thereby potentially facilitating the advancement of neoplastic growth.[13] Studies have demonstrated that EGFR-1 is upregulated in several types of malignancies, such as non-small cell and small cell lung cancers, breast cancer, and glioblastoma multiforme.[11,12] The findings of Mawrin et al, study suggest that there is a correlation between the expression of EGFR-1 and higher histopathological grades as well as unfavorable patient outcomes.[12] This underscores the potential of EGFR-1 as a prognostic marker. Thirteen inhibitors targeting the EGFR have demonstrated efficacy in managing cancer that harbors EGFR mutations.[13, 14] The mechanism of action of EGFR inhibitors involves the inhibition of cancer cell proliferation that is attributed to the over-expression of EGFR receptors.[14] This intervention has the potential to reduce tumor size and enhance overall survival rates.

EGFR exhibits potential as a target for targeted anti-EGFR therapy in the treatment of meningioma.[13,14] Hence understanding the molecular alterations associated with meningioma development, particularly the role of EGFR-1, can provide valuable insights for improved patient management. Moreover, comprehending gender discrepancies could aid in customizing therapeutic approaches and enhancing the quality of care provided to patients. Hence, we aimed to enhance the existing comprehension of meningioma histopathology and to explore the role of EGFR-1 as a potential biomarker for improved risk stratification and treatment selection by concentrating on Grades I and II meningioma cases. Our study emphasized the gender distribution of meningioma, the histopathological grading, and the pattern of

EGFR-1 expression on immunohistochemistry.

MATERIALS AND METHODS

This cross-sectional study was carried out at Ayub Teaching Hospital, Abbottabad, in collaboration with the Neurosurgery Department, the Histopathology Laboratory of Ayub Medical College, Abbottabad, and the Health Diagnostic Laboratory, Islamabad. The Institutional Ethics Committee of Ayub Teaching Hospital, Abbottabad granted ethical approval for this study. This study utilized a non-probability consecutive sampling technique. Data was collected from the patients, informed consent, who were diagnosed from July 2021 to December 2022.

Inclusion Criteria

All patients, regardless of gender, were diagnosed with meningioma and fell within the age range of 23 to 54 years.

Exclusion Criteria

Patients whose samples were deemed inadequate for IHC staining or who declined to participate.

Data Collection Procedure

The medical records of the patients who were included in the study were utilized to gather clinical and pathological data. We documented several parameters, including age, sex, presenting symptoms, tumor histological subtype and grade, and the positivity status for EGFR-1 expression as determined by immunohistochemistry on a self-developed proforma. The specimens of meningioma tissue were subjected to histopathology examination at the Histopathology Laboratory located at Ayub Medical College in Abbottabad. The biological samples underwent fixation in 10% formalin, tissue processing via an automated tissue processor, paraffin embedding, and sectioning using Shandon™ Finesse™ 325 Manual Microtome, a China-manufactured microtome. The tissue sections underwent Hematoxylin and eosin staining and were subsequently evaluated by two consultant histopathologists with extensive experience. The ultimate determination regarding the grading of the lesion was made through a collaborative agreement. The expression of EGFR-1 in meningioma tissue samples was evaluated through immunohistochemical analysis. Standard protocols were followed for conducting the immunohistochemistry (IHC) analysis at the Health Diagnostic Laboratory in Islamabad. The study employed targeted primary antibodies directed against EGFR-1 and incorporated suitable positive and negative controls.

EGFR 1 protein expression was detected by a semi-quantitative method. The IHC staining for EGFR was scored based on staining intensity as described: 0= no staining; 1+ = faint cytoplasmic staining in >10% of cells on HPF; 2+ = moderate membranous staining in > 10% to 20% of cells on HPF; 3+ = strong membranous staining or staining in > 20% of cells. To validate the reaction, a positive control consisting of lung carcinoma samples and a negative control involving the omission of primary antibodies were utilized for visual comparison. The relevant data, such as the histopathology grade, IHC status, age, and gender of the patients, were recorded on a standardized form.

Data Analysis

The statistical analysis was conducted using the SPSS software version 25. The clinicopathological data was summarized using descriptive statistics and the expression of EGFR-1 was presented in percentage form. The study assessed potential associations between clinicopathological parameters and EGFR-1 expression through the use of suitable statistical tests, such as the chi-square test. The selected level of significance was 5% ($p \leq 0.05$), indicating that results with a p-value

of 0.05 or lower were considered statistically significant.

RESULTS

A cohort of 39 meningioma patients who received surgical intervention for lesion excision were studied. Subsequent histopathological analysis was conducted to grade the lesion, and an immune-histochemical assessment was performed to detect the EGFR-1 gene expression.

Age Distribution of Patients

The study sample had a mean age of 39 ± 8.838 years. This indicates a moderate level of diversity in the age distribution of the participants. The sample encompasses individuals aged between 23 and 54 years.

Gender Distribution of Patients

The sample had a slight predominance of females 22 (56.4%) over males 17 (43.6%).

Prevalence of Grade I Meningioma

Table 1 presents data pertaining to the frequency and percentage distribution of various characteristics exhibited by the study population. A significant proportion of patients were diagnosed with Grade I meningioma 35 (89.7%), and a significant proportion of them had pronounced EGFR-1 expression 30 (76.9%).

Table 1: Characteristics of the study population.

Variables		Frequency	Percent
Gender	Male	17	43.6%
	Female	22	56.4%
Grade of Meningioma	Grade I	35	89.7%
	Grade II	4	10.3%
EGFR-1 Expression	Mild	2	5.1%
	Moderate	7	17.9%
	Severe	30	76.9%
	Total	39	100.0%

Table 2 shows the correlation between gender and meningioma grade, as well as the association between gender and EGFR-1 expression. The majority of the patients were diagnosed with grade I meningioma 35 (89.7%), with 14 being male and 21 being female. Severe EGFR-1 expression was observed in 30 individuals, consisting of 14 males and 16 females. We did not find a statistically significant association between gender and either EGFR-1 expression or meningioma grade.

Cross-tabulation analysis of gender, EGFR-1 expression, and grade of meningioma is presented in Table 3. The statistical analysis using the chi-square test revealed a noteworthy correlation between the intensity of EGFR-1 expression and the grade of meningioma in Grade I cases. The Pearson Chi-Square test resulted in a p-value of 0.049, indicating statistical significance. However, no statistically significant correlation was noted between EGFR-1 expression and gender.

Table 2: Cross-tabulation between Gender and EGFR-1 and Grade of Meningioma.

Parameters		Gender		Total	Chi-Square Test
		Male	Female		
EGFR-1 Expression	Mild	1	1	2	P = 0.673
	Moderate	2	5	7	
	Severe	14	16	30	
Total		17	22	39	
Grade of Meningioma	Grade I	14	21	35	P = 0.181
	Grade II	3	1	4	
Total		17	22	39	

DISCUSSION

Meningioma is a prevalent neoplasm located within the cranium, constituting roughly a quarter of all primary intracranial

Table 3: Cross-tabulation of Gender, EGFR-1 expression, and Grade of Meningioma.

Meningioma Grade			EGFR-1 Expression			Total	Chi-Square Test
			Mild	Moderate	Severe		
Grade I	Gender	Male		0	14	14	0.049 (significant result)
		Female		5	16	21	
	Total			5	30	35	
Grade II	Gender	Male	1	2		3	0.248
		Female	1	0		1	
	Total		2	2		4	
Total	Gender	Male	1	2	14	17	0.063
		Female	1	5	16	22	
	Total		2	7	30	39	

tumors. Comprehending the molecular attributes of meningioma, encompassing genetic modifications and protein manifestation is imperative for precise diagnosis, prognosis, and focused therapeutic approaches. The mean age of our patients was 39 ± 8.838 years. We observed a wide age range of individuals, ranging from 23 to 54 years, which underscores the prevalence of meningioma across various age cohorts. The observed age distribution is consistent with prior research on meningioma, which has documented a comparable age spectrum among those afflicted.[2,3]

A marginally greater incidence of meningioma in females (56.4%) as opposed to males (43.6%) was observed in our cohort. The observed gender disparity in the incidence of these tumors may be ascribed to genetic predisposition and hormonal influences, particularly estrogen, in their pathogenesis.[15] The application of chi-square tests for statistical analysis did not yield a statistically significant correlation between meningioma grade or EGFR-1 expression and gender. Our results align with the current literature wherein no statistically significant gender-based variations in meningioma are observed. [15, 16] However, the prevalence of EGFR-1 overexpression in meningioma is said to be higher in women as compared to men.[1,2,17] It is noteworthy that the lack of a substantial correlation in our study does not inevitably indicate that gender does not exert an impact on meningioma. Instead, it indicates that within our cohort, gender in isolation may not serve as a potent prognosticator of meningioma grade or EGFR-1 expression.

Upon analyzing the distribution of meningioma grades, our investigation revealed that a significant proportion of the population 35 (89.7%) exhibited Grade I meningioma, whereas a minor fraction 4 (10.3%) displayed Grade II meningioma. The observed frequency of Grade I meningioma in our study cohort is in line with the documented global incidence rates and the overall pattern observed in meningioma cases. [2] The aforementioned results are in line with the existing body of literature on meningioma, which consistently indicates that Grade I meningiomas are the most prevalent subtype.[2,15]

Concerning the expression of EGFR-1, our data revealed that a significant proportion of the participants demonstrated intense expression 30 (76.9%), while a smaller percentage exhibited moderate 7 (17.9%) and mild 4 (5.1%) intensity of EGFR expression. These results are consistent with the recent studies conducted by Lee et al and Arnli et al, which emphasized the elevated occurrence of EGFR-1 manifestation in meningioma.[18,19] The upregulation of EGFR-1 has been linked to neoplastic advancement and inferior prognostic results in diverse malignancies, such as gliomas and breast carcinoma.[20]

The over-expression of EGFR-1 in meningioma indicates its potential as a therapeutic target and a prognostic indicator.[19,20]

The cross-tabulation analysis conducted to investigate the correlation between meningioma grade, EGFR-1 expression, and gender yielded a noteworthy correlation between the expression of EGFR-1 and the grade of meningioma in Grade I cases (p=0.049). Our results second the findings documented by Smith et al. who observed a comparable correlation in a more extensive sample of individuals with meningioma.[15] Our results also align with those of recently published literature, which demonstrated a statistically significant increase in EGFR expression levels in grade I meningioma compared to grade II meningioma.[16,21] Furthermore, Telugu and colleagues discovered that the over-expression of EGFR was correlated with an unfavorable prognosis among individuals diagnosed with grade I meningioma.[16] Ciardiello et al, conducted a recent study that revealed that the expression of EGFR was linked to heightened tumor growth and invasion in cell lines of meningioma.[22,23] Ciardiello and colleagues have reported that the use of EGFR inhibitors can effectively target the over-expression of EGFR, resulting in a reduction in tumor growth and invasion in cell lines.[22] We did not find any significant correlation between the intensity of EGFR-1 expression and the grade of meningioma in Grade II meningioma cases (p=0.248). The insignificance of the findings can be attributed to the limited sample size of Grade II meningioma cases in our study (n=4).

Numerous studies have been conducted to examine the expression of EGFR-1 in meningioma, yielding diverse outcomes. In 2016, a research article was published in the Journal of Neurosciences in Rural Practice that examined the expression of EGFR-1 in a cohort of 100 meningiomas, comprising 80 grade I tumors and 18 grade II tumors, through the utilization of immunohistochemistry. The research revealed a notable increase in the expression of EGFR-1 in grade I and II tumors, indicating the potential involvement of EGFR-1 in the advancement of meningioma.[16] Many studies have revealed that the expression of EGFR-1 was linked to an increased recurrence rate and a reduced time to recurrence, indicating that EGFR-1 could serve as a prognostic indicator for meningioma.[11,12,23] According to a recent study conducted by Li et al., the expression of EGFR was linked to an unfavorable prognosis among patients diagnosed with meningioma.[23] The research revealed that individuals exhibiting elevated levels of EGFR expression exhibited a reduced overall survival duration in comparison to those with lower levels of EGFR expression. A recent study has reported that the expression of EGFR was observed to be more prevalent in meningioma of grade II and III as compared to those of grade I.[23] The research additionally discovered that the expression of EGFR was correlated with an increased likelihood of tumor recurrence and mortality.

CONCLUSION

A statistically significant higher intensity of EGFR expression in Grade I meningioma indicates that the intensity of EGFR expression could serve as a valuable prognostic biomarker.

LIMITATIONS

A small sample size and study at a single institution may limit the generalizability of our findings.

RECOMMENDATIONS

Future studies with larger sample sizes are recommended to validate our results. Multi-center studies involving diverse populations would provide a more comprehensive understanding of meningioma characteristics.

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Additional Information

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Conflicts of Interest:
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AUTHORS CONTRIBUTIONS

Sr.#	Author's Full Name	Intellectual Contribution to Paper in Terms of:
1.	Shagufta Naeem& Shabana Naz	1. Study design and methodology.
2.	Shagufta Naeem, Hamza Javed Sikandar Ahmed Zahid	2. Paper writing.
3.	Arshad Khan, Ehtesham & Ahmed Khan Afridi	3. Data collection and calculations.
4.	Shagufta Naeem, Shabana Naz & Hamza Javed	4. Analysis of data and interpretation of results.
5.	Shagufta Naeem, Shabana Naz & Hamza Javed	5. Literature review and referencing.
6.	Arshad Khan, Ehtesham Ahmed Khan Afridi & Sikandar Ahmed Zahid	6. Editing and quality insurer.