Prospective study of Meningioma & diagnostic validity of MIB-1 index in grading of meningioma

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Abstract

Background: Meningioma is one of indolent neoplasm. Meningiomas may make their appearance in childhood or adolescence, but most are encountered in middle or later adult life. Females are affected more commonly than males.

Aims: (1) Categorization of meningioma cases on basis of WHO (2007) classification. (2) Evaluation of clinico-radiological data & correlation with histopathology grade of meningioma. (3) Role of ki67 (MIB-1) index in grading of meningioma. (4) Evaluate the validity of MIB-1 index in grading of meningioma.

Methods and Material: This is prospective study conducted at Department of Pathology at Gujarat Cancer and research Institute, Ahmedabad. Total number of cases enrolled in study was 90 and it was carried out over period of September 2013 to December 2015 after being approved by the ethical committee of the Institute.

Patients selection were done from details of all cases, histomorphology, radiology and immunohistochemistry (IHC) - MIB-1 confirmed by Meningiomas. Surgically resected frozen tissue and specimen section included in study.

Results: MIB-1 LI and the 4 parameters used in the WHO grading system, namely, mitotic index, increased cellularity, sheet-like architecture and small cell formation showed good correlation. There was a poor correlation with certain other histological parameters, such as necrosis, cellular pleomorphism and brain invasion.

MIB-1 labeling index less than 7% mostly are benign in nature and greater than 7% favors atypical or anaplastic.

Conclusions: The MIB-1 LI has highest validity at a level of 7% in the diagnosis of histological atypia in meningiomas. Most of meningiomas, which are missed in appropriate grading on biopsy can be due to focal involvement of atypia.

This study is helpful for pathologist for appropriate diagnostic grading and for the surgeon to take appropriate surgical decision and to follow up the same cases.

In combination, histomorphological features and MIB-1 LI definitely help in identifying potentially aggressive nature of meningiomas.

Key-words: Meningioma, MIB-1 index, brain invasion

Introduction

Meningioma is one of indolent neoplasm. Meningiomas may make their appearance in childhood or adolescence, but most are encountered in middle or later adult life. Females are affected more commonly than males.(1)

Notified some studies suggest a particularly increased prevalence in women with mammary carcinomas,(2) and rare meningiomas do actually harbor metastatic deposits from breast primaries.(3) In addition to that, their frequent expression of progesterone (and, less commonly, estrogen as well as androgen) receptors.(4)

Multifocal meningiomas with type 2 ('central') neurofibromatosis (NF-2),(4) the genetic locus for which resides on chromosome 22q12. The presentation of a case of meningioma in childhood or adolescence should trigger investigation for underlying NF-2.(5) Familial examples occurring outside the setting of classic NF-2 have also 5been described.(5)

From number of epidemiologic studies, Ionizing cranial irradiation emerges as conferring significant risk for subsequent meningioma development,(6) radiation-related lesions being more often multiple, histologically atypical, and clinically aggressive than those arising in sporadic fashion.(7,4).

Most meningiomas arise within the cranial cavity, are dura-based and found in the vicinity of the superior sagittal sinus, over the cerebral convexities or in contact with the falx cerebri. Intracranial meningiomas may also originate within the tela choroidea or stroma of the choroid plexus and rest entirely within the ventricular system.

Meningiomas are classified by WHO(8) mainly in three grades out of which WHO Grade-I meningiomas are most common. WHO Grade-I is devided into subtypes, meningothelial, fibroblastic, psammomas, angiomatous, and transitional type etc. WHO Grade-II meningioma is devided into subtypes, Clear cell and chordoid subtypes, or atypical by criteria. WHO Grade-III Meningiomas is devided into subtypes, Rhabdoid and papillary subtypes, or anaplastic by criteria.

Most of the studies carried out to find the recurrence and aggressive character of meningioma by using various markers like MIB-I LI, p53, BRDU (Bromodeoxyuridine) proliferation indices DNA flow cytometry,(9,10,11,12) MIB-1 LI index has been showed to have strong correlation with recurrence free survival in meningiomas.(1,8,13,14,15,16) strong associations with

recurrence-free survival (RFS) in meningiomas.[9,11,13,14,15,16]

We evaluated the role of ki67 (MIB-1) index in grading of meningioma and its validity in grading of meningioma.

Subjects and Methods

This is prospective study conducted at Department of Pathology at Gujarat Cancer and research Institute, Ahmedabad. Total number of cases enrolled in study was 90 and it was carried out over period of September 2013 to December 2015 after being approved by the ethical committee of the Institute.

Patient selection:

Details of all 90 cases, patient's age, primary tumor site, clinical symptoms, operation title, histomormphology, radiology and IHC (MIB-1) confirmed by Meningiomas, were retrieved from patient case file.

Pathological methods:

All craniotomy frozen tissue sent in Normal saline are examined by imprint or crush or squash and paraffin section. Remaining tissue is fixed in 10% formalin overnight as per standard protocol. Then all sections were studied for tumor type, grading and extent or invasion in brain ECT. Same formalin fixed paraffin block was used for IHC i.e. MIB-1 index evaluation.

IHC staining for MIB-1 index.

An IHC stain for MIB-1 index was performed on paraffin section Blocks, using ABC technique with antigen epitope enhancement by heat.

The Diamineobenzidine reaction was used as the final detection step. The slides were counterstained with Mayer's Heamatoxyllin.

The Method for epitope retrieval includes overnight incubation at 60˚C after antigen epitope enhancement and staining by fully automated machine VETANA BENCHMARK XT.

Appropriate Positive and Negative controls were included in all stains to ensure the quality and consistency of staining results.

MIB-1 LI stained slide was scanned under low power (10x objectives) for area of selection of MIB-1 count (i.e. hot spot).

At least 1000 nuclei were counted at high magnification (40 x objectives) without recounting same areas and the average was expressed as percentage. Foci of necrosis were excluded.

In this study, only Cranial meningiomas included and all spinal cord meningiomas are excluded.

Statistical analysis

K Correlation is studied by using Pearson correlation coefficient (R value) and p-value Fisher exact test. R value near to 1 shows good correlation and $P < 0.05$ was taken as the cut off for significance.

Ethical Approval

Approval was sought from the institute's ethics committee.

Results

There were a total of 90 meningiomas diagnosed between periods of September 2013 to December 2015.

On reviewing the slides using the criteria recommended by the WHO 2007 classification, 71 (78.88%) of these were classified as benign: WHO Grade I tumors, 19 (21.12%) were classified as atypical WHO Grade II tumors and 0 (0.0%) were classified as anaplastic WHO Grade III meningiomas.

The overall Age range was 25-76 years, Mean Age at Diagnosis being 48.93 years. In 90 cases 37 are male and 53 are female patient.

Table 1: General profiles of the cases studied

Total number of cases		90
Gender	Male	37
	Female	53
Age (Years)	Mean	48.93
	Range	25-76
Histological Diagnosis	Benign	71
	Atypical	19
	Anaplastic	0

Table 2: Distribution of lesion

Site of lesion	**Number of cases**
Frontal Region	39
Temporal Region	09
Parietal Region	08
Occipital Region	13
CP- Angle	07
Sphenoid Wing	06
Other	08
Total	90

Out of total 90 cases of meningiomas; 39 were located in frontal region, 09 in temporal region, 08 in parietal region, 13 in occipital region, 07 in CP-Angle, 06 in sphenoid wing and 08 are located in other site like parasellar and suprasellar region, parasagital region, olfactory groove, base of skull and retro-orbital region etc.

Table 3: Distribution of tumors according to grade at arbitrary cut-offs of MIB-1 LI

MIB-1 LI	Benign	Atypical	Anaplastic
0-4 %	59	5	0
4.1-7 %	6	3	0
7.1 -11 %	1	2	0
≥ 11 %	5	9	0
TOTAL	71	19	0

The three grades of tumor were distributed using arbitrary cut-offs of the MIB-1 LI.

Table 4: Correlation of MIB-1 LI with individual histological features of atypia

Good correlation	Mitotic Index	R - value	P – value
		0.7380	0.00001
	Cellularity	0.6213	0.00001
	Small cell formation	0.5963	0.00001
	Sheet like architecture	0.7154	0.00001
Nearly statistical significance	Necrosis	0.2325	0.02778
Weak Correlation	Cellular Pleomorphism	0.2608	0.01303
	Brain Invasion	0.2639	0.01196

Tumors with a MIB-1 LI of >7% were more likely to be atypical /Anaplastic than benign. The distribution of meningiomas at the estimated MIB-1 LI cut-off level of 7% is represented pictorially.

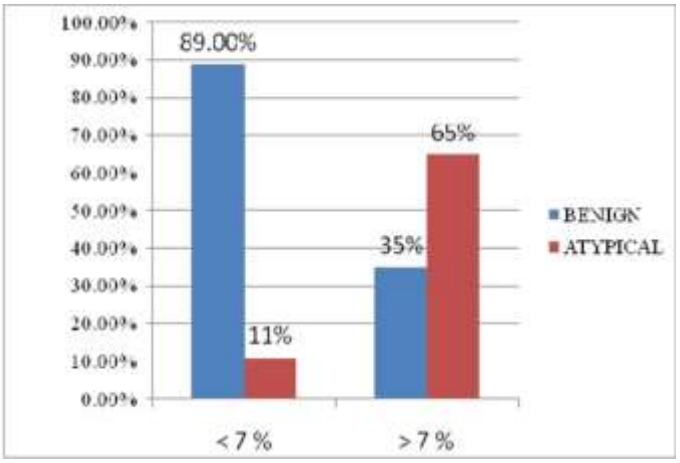

Fig. 1: Distribution of the tumors around the MIB-1 LI at diagnostic level

The correlation of MIB-1 with individual histological characteristics using by the Pearson correlation coefficient (R value) and P-value Fisher exact test (Table 4) shows a good correlation between MIB-1 LI and the 4 parameters used in the WHO grading system, namely, mitotic index, increased cellularity, sheet-like architecture and small cell formation. There was a poor correlation with certain other histological parameters, such as necrosis and cellular pleomorphism. There was also a poor correlation noted with brain invasion.

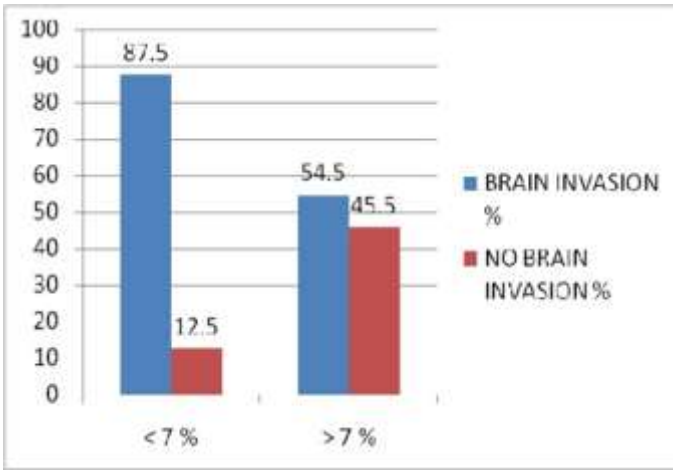

Fig. 2: Distribution of brain invasion with MIB-1 LI at diagnostic level

A histologically stratified distribution of brain invasion with MIB-1 LI at the cut-off level of 7% demonstrates that the MIB-1 levels of brain invasive tumors vary according to their histological grade; 6 of 13 brain invasive meningiomas having a MIB-1 LI > 7% were WHO grade-II while 7 of 13 brain invasive meningiomas with MIB-1 LI < 7% were grade I tumors in Fig. 2.

Table 5: Distribution of Meningiomas histologically stratified at the diagnostic level of the labeling index

MIB-1 LI	Benign	Atypical	Anaplastic
≤ 7%	65	8	0
> 7%	6	11	0

The above table shows the histological stratification at diagnostic level of MIB-1 labeling index. It indicates the cut of range of benign meningiomas to atypical/anaplastic is 7% MIB-1 index. In which MIB-1 labeling index less than 7% mostly are benign in nature and greater than 7% favors atypical or anaplastic.

Table 6: Meningioma grade-II classified according to WHO Classification criteria

		Brain Invasion	No Brain Invasion
Number of cases		13	06
Mitosis ≥ 4 /10HPF		03	06
Variant	Clear cell	0	02
	Atypical/Chordoid	0	04

The above table classified the Meningioma Grade-II according to WHO Classification of meningioma based on Brain Invasion, Mitosis and their variants Clear cell and Atypical or Chordoid by criteria. Total 19 cases of meningioma grade-II; out of them 13 cases graded meningioma grade-II according brain invasion, in which only 3 cases shows ≥4 mitosis/10hpf. Another 6 cases of meningioma graded into Grade-II according to atypia, mitosis index ≥4%, increased cellularity, small cell with high N: C ratio, nuclear pleomorphisms.

Discussion

Meningioma occurs most often in elderly ages. Present study has mean age of 48.93 years with peak incidence in 25 to 76 years age group. Slight female predominance is recorded. Meningiomas were most frequently located in frontal region followed by occipital region and so on. MIB-1LI proved to be helpful in clinching the diagnostic grading and aggressiveness of meningioma.

In the present study, 90 cases of cranial meningiomas were studied (Spinal meningiomas are excluded). Our institute is a tertiary care hospital and most of the patients of meningiomas are referred to us for further evaluation and treatment. Patients in present study belonged to the age group in the range of 25-76 years, with mean age being 48.93 years and a sex ratio of male: female is 1:1.43, which was in accordance with other studies. Present study showed that female gender was slightly favored in Meningiomas, which was in accordance with A. Devaprasath et al[9].

Meningiomas are mostly benign nature tumor, but few of them are aggressive behavior although after surgical excision recurrence may occur. Although certain histological characteristics have stronger associations with a decreased recurrence-free survival.[13,16,17] The histological diagnosis of atypia

remains largely subjective, with significant inter-observer variability that creates difficulties in assessing the prognosis and postoperative management of patients.[13,17] The MIB-1 LI is significant in grading and correlation of recurrence of meningiomas. In this study Mitotic index, Cellularity, Small cell formation, Sheet like architecture, Necrosis, Cellular Pleomorphism and Brain Invasion are individually correlated with MIB-1LI. They shows significant correlation between Mitotic index, Cellularity, Small cell formation, technical nearly significant correlation with necrosis and poor correlation with Cellular Pleomorphism and Sheet like architecture. The correlation of necrosis with MIB-1 LI only neared statistical significance (P=0.02778).

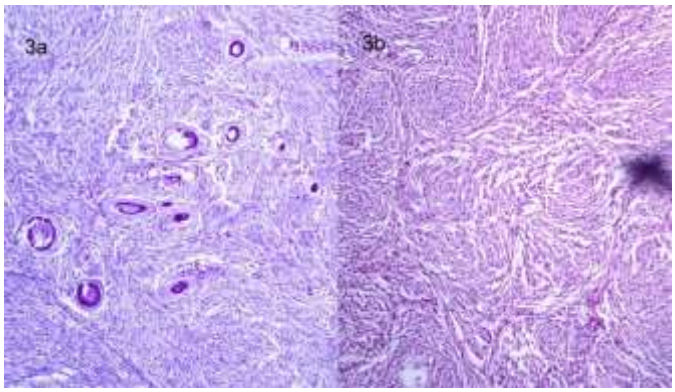

Fig. 3a: Psammomas Meningioma who Grade-I 10x; 3b: Transitional Meningioma who Grade-I 10x

An objective method of counting positively stained nuclei provides a more reproducible method for assessing aggressive behaviour as an adjunct to histology. Our results showed lower MIB-I LI in benign and higher labelling indices in atypical meningiomas in accordance with earlier quoted studies.[9,11,12,14,15,16,18] In A. Devaprasath et al study[9] the MIB-1 LI has highest validity at a level of 7% in the diagnosis of histological atypia in meningiomas and has a good correlation with individual WHO histological features of atypia. The MIB-1 LI used in conjunction with histological features can help in making a recommendation regarding potentially aggressive behavior in meningiomas. And another study of Perry et al[12] demonstrated an association between MIB LI of 4.2% or greater and decreased recurrence-free survival in 425 gross totally resected meningiomas.

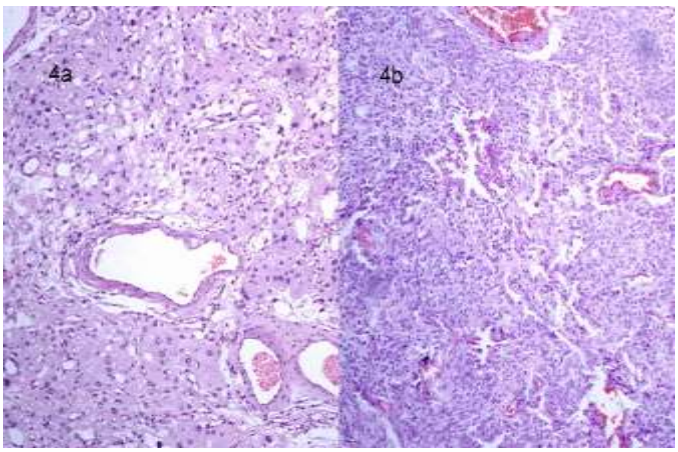

Fig. 4a: Angiomatous meningioma who Grade-I 10x; 4b: Atypical Meningioma who Grade-II 10x

The authors stated that MIB-1 is a useful ancillary study for routine evaluation of meningiomas, particularly those with borderline atypia. They admitted, however, that a MIB LI of 4.2% may not be applicable to other institutions owing to variability in staining and counting methodologies among different laboratories. The difference in cut-off values could be attributed to the use of an electronic image analyzer by Perry et al[12] as opposed to the manually counted MIB-1 LI in our study. Furthermore, we studied the validity of the MIB-1 LI in predicting histological atypia and not its association with a decreased recurrence-free survival, as was done by Perry et al.[12]

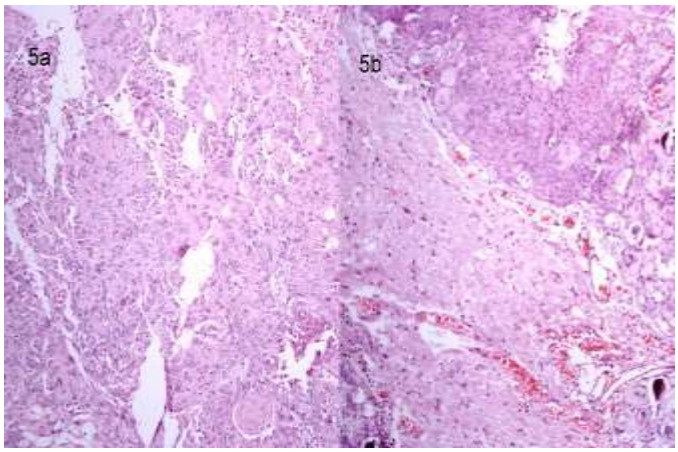

Fig. 5a: Clear cell meningioma who Grade-II 10x Brain; 5b: Brain invasion in meningioma who Grade-II 10x

Present study shows highest correlation between MIB-1 LI and individual WHO histological features of atypia and the MIB-1 LI has been showed to have strong associations with recurrence-free survival (RFS) in meningiomas[11,13-16]. Six meningiomas cases show >7% MIB-1 LI although histomorphologial feature are of meningioma WHO grade-1. >7% MIB-1 LI is in favor of atypical/ anaplastic nature of meningiomas. As meningioma WHO grade –II histomorphological diagnoses due to brain invasion of lesion, these 8 case shows MIB-1 LI is less than 7%. So it shows poor correlation. Few cases had incomplete resection; residual mass is treated by radiotherapy. Problem is that clinical follow-up periods vary between studies, and the length of the postoperative follow-up is important, especially, as was mentioned earlier, in the case of non-recurrent tumors.

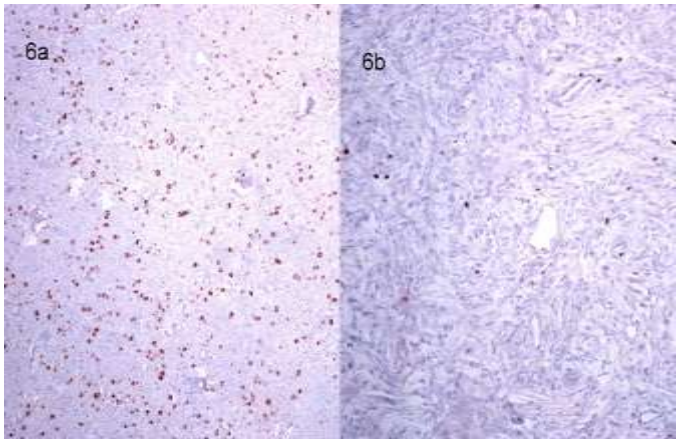

Fig. 6a: High count MIB-1 LI in meningioma who Grade-II; 6b: Low count MIB-1 LI in Meningioma Grade-I

While we do find that MIB-1 LI is a useful objective method, at no point can it be considered a supplement for histological investigations. However, as the criteria for histological grading are subject to considerable inter-observer variation, [19] the MIB-1 LI would be a useful adjunct to histological grading. Tumours that do not have any histological features of malignancy but have a high score should be reviewed carefully.

Conclusion

To conclude, the case study of meningioma (90 cases) has illustrated for diagnostic validity of MIB-1 LI in grading of meningioma (WHO classification of meningiomas) of patients presenting at our institute.

The MIB-1 LI has highest validity at a level of 7% in the diagnosis of histological atypia in meningiomas and has a good correlation with individual WHO histological features of atypia. Most of meningiomas, which are missed in appropriate grading on biopsy, can be due to focal involvement of atypia. These case scenarios are very well caught by MIB-1LI index. Brain invasion have poor correlation with MIB1 LI.

The counting of mitosis/10 HPF helped in histomorphological grading the meningiomas according to WHO 2007 classification of Meningioma. In combination; histomorphological features and MIB-1 LI definitely help in identifying potentially aggressive nature of meningiomas. So, it is an important diagnostic tool in challenging and doubtful cases.

This study is helpful for pathologist for appropriate diagnostic grading and for the surgeon to take appropriate surgical decision and on following up the same cases.

References

1. Juan, R. (2011), Rosai and Ackerman's Surgical Pathology- Central Nervous System 2:28:2389.
2. Longstreth Jr WT, Dennis LK, McGuire VM, Drangsholt MT, Koepsell TD: Epidemiology of intracranial meningioma. Cancer 1993;72:639-648.
3. Zon LI, Johns WD, Stomper PC, Kaplan WD, Connolly J L, Morris JH, Harris JR, Henderson IC, Skarin AT: Breas t carcinoma metastatic to a meningioma. Case report and review of the literature. Arch Intern Med 1989;149:959-962.
4. Perry A, Louis DN, Scheithauer BW, Budka H, von Deimling; A: Meningiomas. In: Louis DN, Ohgaki H, Wiestler OD, Cavenee WK, ed. WHO classification of tumours of the nervous system, Lyon: IARC;2007:164-172.
5. Perry A, Giannini C, Raghavan R, Scheithauer BW, Bane rjee R, Margraf L, Bowers DC, Lytle RA, Newsham IF, Gutmann DH: Aggressive phenotypic and genotypic features in pediatric and NF2-associated meningiomas: a clinicopathologic study of 53 cases. J Neuropathol Exp Neurol 2001;60:994-1003.
6. Longstreth Jr WT, Dennis LK, McGuire VM, Drangsholt MT, Koepsell TD: Epidemiology of intracranial meningioma. Cancer 1993;72:639-648.
7. Al-Mefty O, Topsakal C, Pravdenkova S, Sawyer JR, Harrison MJ: Radiation-induced meningiomas: clinical, pathological, cytokinetic, and cytogenetic characteristics. J Neurosurg 2004;100:1002-1013.
8. Amir Rezaee, Frank Gaillard et al: WHO Classification of Tumours of the Central Nervous System, 4th Ed, Vol 1, 2007.
9. Devaprasath A, Chacko G: Diagnostic validity of the Ki-67 labelling index using the MIB-1 monoclonal antibody in the grading of meningiomas 2003;51(3):336-340.
10. Langford LA, Cooksley B, deMonte F. Comparison of MIB-1 and BRDU proliferation indices in meningiomas. Hum Pathol 1996;27:350-4.
11. Prayson RA. Malignant meningiomas- A Clinicopathological study of 23 patients including MIB-1 and p53 immunohistochemistry. Am J Clin Pathol 1996;105:719-26.
12. Perry A, Stafford SL, Scheithauer BW, Suman VJ, Lohse CM. The prognostic significance of MIB-1, p53, and DNA Flow cytometry in completely resected primary Meningiomas. Cancer 1998;82:2262-9.
13. Ohta M, Iwaki T, Kitamoto T, Takeshita I, Tateishi J, Fukui M. MIB-1 Staining index and Scoring of Histological Features in Meningioma. Cancer 1994;74:3176-89.
14. Abramovich CM, Prayson RA. MIB-1 labeling indices in benign, aggressive and malignant meningiomas: a study of 90 tumors. Hum Pathol 1998;29:1420-7.
15. Abramovich CM, Prayson RA. Histopathologic features and MIB-1 Labeling Indices in Recurrent and Nonrecurrent Meningiomas. Arch Pathol Lab Med 1999;123:793-800.
16. Perry A, Stafford SL, Scheithauer BW, Suman VJ, Lohse CM. Meningioma Grading- An Analysis of Histologic Parameters. Am J Surg Pathol 1997;21:1455-65.
17. Karabagli Pinar, Aydinsav: Proliferative Indices (MIB-1) in Meningiomas: Correlation With The Histological Subtypes and Grades: Journal of Neurological Sciences 01/2006.
18. Perry A, Stafford SL, Scheithauer BW, Lohse CM, Wollan PC. "Malignancy" in Meningiomas. Cancer 1999;85:2046-56.
19. Louis DN, Scheithauer BW, Budka H, von Deimling A, Kepes JJ. Meningiomas In: Kleihues P, Cavenee WK, editors. Pathology and Genetics. Tumors of the Nervous System: WHO Classification of Tumors. 1st edn. Lyon: IARC Press; 2000. pp. 176-84.