Clinical Neurology and Neurosurgery 178 (2019) 46–50

Contents lists available at ScienceDirect

Clinical Neurology and Neurosurgery

journal homepage: www.elsevier.com/locate/clineuro

# The correlation between phosphorylated Histone H3 (PHH3) and p-STAT3 in Meningiomas

Erdinc Ozek[a,*], Halil Akdag[b], Zeynep Tosuner[c], Anas Abdallah[a], Mustafa Aziz Hatiboglu[a]

[a] Neurosurgery Department, Bezmialem Vakif University, Fatih, Istanbul, Turkey
[b] Bezmialem Vakif University, Fatih, Istanbul, Turkey
[c] Pathology Department, Acibadem University, Atakent, Istanbul, Turkey

ARTICLE INFO

*Keywords:*
Intracranial meningioma
PHH-3
Recurrent meningiomas
STAT-3

ABSTRACT

*Objective:* To assess the correlation between PHH-3 and STAT-3 in grade I and grade II meningiomas.
*Patients and methods:* Medical records were retrospectively reviewed for all cranial cases which diagnosed and underwent surgery at Bezmialem Vakif University Hospital between 2012 and 2017. All cranial grade I and grade II meningioma patients constituted the core sample for this study.
*Results:* This series included 104 (69 female, 35 male) patients, with a median age of 57.3 years. The mean preoperative course was 23.0 ± 40.5 months. The most common symptom was headache (76%) and followed by seizure (24%), weakness (18%) and visual disturbances (14%). Seventy one (68.2%) patients were diagnosed as WHO grade I meningioma and 33 (31.8%) were WHO grade II, grade III meningiomas were excluded from study due to small number of patients. Subtypes of meningioma includes 5 angiomatous (4,8%), 6 fibroblastic (5.7%), 1 meningothelial (0,9%), 11 psammomatous (10,5%), 3 secretory (2,8%), 43 transitional (41,3%) and 33 atypical (31,7%) meningiomas. There is a strong correlation with PHH-3 and Ki-67 (**p**:0,001 >) and mitosis index (**p**:0,001 > ) although there is no correlation with STAT-3 (p:0,260). There is a strong correlation with STAT-3 and Ki-67 (**p**:0,013), although there is no correlation with mitosis index (**p**:0,085) and PHH-3 (**p**:0,260).
*Conclusions:* In our study we also obtain same results with Ki-67 and mitotic index, although correlation with PHH-3 and STAT-3 is firstly determined and there was no statistically significant relation were observed. Depends on the STAT-3 cell proliferation feature, inactivation of these pathways may predict new chemotherapies for grade II meningiomas.

## 1. Introduction

Meningiomas are most common primary central nervous tumors (CNS) that derive from meningothelial cells and typically attached inner surface of dura [1]. Many various cell proliferation markers have been used for grading CNS tumors to determine the patient prognosis and different treatment options. Mitotic activity, which is defined as "the number of mitotic figures per 10 consecutive high-power fields in the highest mitotically active area" is the most reliable prognostic factor used for meningiomas and that compose the World Health Organization (WHO) grading system [2–4]. The WHO classification of meningiomas divided into 3 different subtypes that increasing the risk of recurrence: benign (grade I), atypical (grade II), and anaplastic (grade III) [2]. Meningiomas did not undergo revision in the 2016 WHO classification for brain tumors except brain invasion criteria that added for atypical meningiomas. Various methods and biomarkers have been extensively studied in the literature [5], to determine the assessment of degree of proliferation as a predictor of recurrence. Therefore Ki-67 labeling is used to predict prognostic factor for meningiomas and that is expressed on all cell cycle phases except G0 and does not project sensitively proliferating cells [6–12]. Although Ki-67 widely used, due to inter-laboratory variability in staining and subjective counting methods are the restrictions that have been approved in the literature [13]. For these reasons, new biomarker quest has emerged and mitosis specific marker PHH-3 is started to use in new studies [14–16] and promising results has been obtained. A protein family member signal transducer and activator of transcription 3 (STAT-3) has phosphotyrosine and DNA binding domain that acts as a transcription factor [17]. STAT-3 pathways has been studied previously in atypical and anaplastic meningiomas [18] although its correlation with biomarkers PHH-3, Ki-67, mitotic index and tumor recurrence have not been studied. The purpose of this study is to compare and correlate the prognostic value of PHH-3

* Corresponding author at: Neurosurgery Clinic, Bezmialem University School of Medicine, Fatih, Istanbul, Turkey.
*E-mail address:* erdincozek@gmail.com (E. Ozek).

https://doi.org/10.1016/j.clineuro.2019.01.016
Received 7 November 2018; Received in revised form 19 December 2018; Accepted 24 January 2019
Available online 25 January 2019

and p-STAT-3 with mitotic index, ki-67 labeling index, tumor recurrence presence and WHO classification grades in patients treated for meningiomas.

## 2. Patients and methods

### 2.1. Patient data

All patient records were retrospectively reviewed for all cranial tumors that were surgically treated in Bezmialem Vakif University Hospital from January 2012 to June 2017. Patient characteristics, such as patient age at the time of surgery, gender, recurrence rate, and histopathological findings such as WHO grade, progesterone receptor presence, mitotic index, Ki-67 labeling, PHH-3 proliferating rates and activation of p-STAT-3 have been evaluated. This retrospective study has been approved by the local medical ethics committee of Bezmialem Vakif University.

### 2.2. Meningioma tissue collection

All hematoxylin and eosin stained slides of a total number of 104 meningioma cases were reviewed by the pathologist according to their subtype, mitotic activity and grades. Patient series in our study includes consecutive patients who underwent surgery for meningioma. pSTAT-3 and PHH-3 immunohistocemical stainings were performed for the study, not at time of diagnosis. Tissue blocks were then used for immunohistochemistry.

### 2.3. Immunohistochemistry

Four-micrometer-thick sections of formalin-fixed paraffin embedded tissues were placed on 3-aminopropyletxylene-covered slides. Subsequently, they were stained with rabbit polyclonal Santa Cruz Biotechnology p-STAT3 antibody (1/100 titer; clone ser727 rabbit polyclonal antibody) and rabbit polyclonal ZETA Corp PHH3 antibody (1/50 titer; rabbit polyclonal antibody) following the manufacturer`s protocol. Briefly, staining was performed on the Ventana BenchMark Ultra (Ventana Medical Systems Inc.). The staining protocol included Cell Conditioning 1 for 64 min for p-STAT 3 and 30 min for PHH-3, preperoxidase inhibition and primary antibody incubation for 1 h and 20 min at 37° C. Ultra-View Universal DAB Detection Kit (Ventana Medical Systems) was used to detect p-STAT 3 and PHH3 protein expressions. Tissues were counterstained with Hematoxylin for 16 min and Bluing Reagent for 4 min. The slides were examined by the pathologist and nuclear staining ratio for p-STAT3 in tumoral cells and the total number of immunostained mitotic figures per 10 HPFs were evaluated in each case (Figs. 1 and 2).

### 2.4. Statistical analysis

All data are expressed as the median or mean ± standard deviation with the range shown in parentheses. Multivariate analyses were performed to examine the correlations between PHH-3, Ki67 labeling index, mitotic index, p-STAT-3 activation with tumor recurrence presence and WHO meningioma grading system. Differences between groups were assessed by a one-way analysis of variance (ANOVA) using the SPSS 21.0 statistical program (SPSS, Chicago, IL). Significance in the multivariate model was determined using a p value of $< 0.05$, and trend-level effects were defined as $p = 0.05–0.10$. All p values were presented with an odds ratio (OR). When OR could not be calculated, relative/risk ratio (RR) was calculated. The corresponding 95% confidence intervals (CIs) were obtained. If both values are categorical square test were used, if one value is categorical and the other is average it was compared with Mann-Whitney *U* test, if both values are quantity correlation coefficient was calculated.

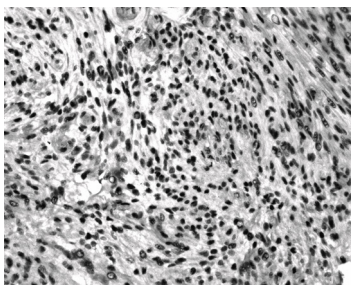

**Fig. 1.** ×200 magnification, diffuse nuclear immunostaining by pSTAT3.

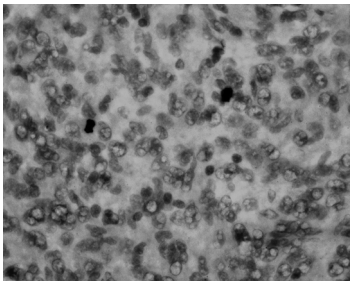

**Fig. 2.** ×400 magnification, two mitotic figures highlighted by PHH-3 immunostain in 1 HPF (high power field area).

## 3. Results

### 3.1. Patients characteristics

One hundred and four patients who were operated due to intracranial meningioma between January 2012-June 2017 years were included to our study. This study included consecutive104 (69 female (61.5%), 35 male (38.5%)] patients with a mean age of 57.53 (range 2–84 years) years. The most common symptom was headache (76%) and followed by seizure (24%), weakness (18%) and visual disturbances (14%). Seventy one (68.2%) patients were diagnosed as WHO grade I meningioma and 33 (31.8%) were WHO grade II, grade III meningiomas were excluded from study due to small number of patients. Subtypes of meningioma includes 5 angiomatous (4,8%), 6 fibroblastic (5.7%), 1 meningothelial (0,9%), 11 psammomatous (10,5%), 3 secretory (2,8%), 43 transitional (41,3%) and 33 atypical (31,7%) meningiomas. The most common anatomic locations were frontal, temporal and parietal which were seen in 24 (23%), 21 (20%) and 19 (18%) patients. The median size of the largest diameter of the tumor was 45,59 mm (range: 12–100 mm).

### 3.2. Surgery and follow-up

The main goal of the surgery for meningiomas is total resection of the tumor, although it could not be possible for every time due to anatomical and surgical difficulties. The surgical resection criteria simpson grading system is important predictor for recurrence regarding pathological grading. In this study median simpson grade was 2 for grade I and grade II meningiomas meningiomas. The mean follow-up period was 33.1 ± 21.3 months (range from 50 days to 68 months). Twenty-six (25%) of the patients had recurrence tumor growing and re-operated or Gamma-Knife treatment was performed. Recurrence rate was 19,7% for grade I meningiomas and 33,3% for grade II meningiomas (Pearson test, p: 0,209)

### 3.3. Histopathological findings

Histopathological markers Ki-67, mitosis index, PHH-3 proliferation and protein p-STAT-3 activation correlation and comparison with

**Table 1**
Histological and characteristic features of Grade I and Grade II meningiomas.

| | Grade I (n:71) | Grade II (n:33) | P value |
|---|---|---|---|
| Mean Age | 56,929 | 58,151 | – |
| Simpson Grade | 2,014 | 1,93 | 0,616 |
| Recurrence Rate | 33,3% | 19,7% | 0,209 |
| Mitotic Index | 0,77 | 4,42 | < **0,001** |
| Ki-67 Value | 5,23 | 14,24 | < **0,001** |
| PHH-3 Proliferation Index | 0,80 | 5,42 | < **0,001** |
| p-STAT-3 Activation Index | 30 | 17,03 | 0,021 |

histopathologic grades, recurrence presence, simpson scoring and progesterone receptor presence were studied.

For grade I (n:71) meningiomas mitosis index mean value was 0,77 (sd:0,78, range 0–3, med:1) and 4,42 (sd:2,53, range 0–10, med:5) for grade 2 (n:33) meningiomas and it is statistically significant (Mann-Whitney p:0,001 >). For grade I meningiomas Ki-67 mean value was 5,23 (sd:3,14, range 1–15, med:4) and 14,24 (sd:8,11, range 4–40, med:12) for grade 2 meningiomas and it is statistically significant (Mann-Whitney p:0,001 >). For grade I meningiomas PHH-3 proliferation index mean value was 0,80 (sd:0,94, range 0–4, med:1) and 5,42 (sd:3,69, range 0–14, med:5) for grade 2 meningiomas and it is statistically significant (Mann-Whitney p:0,001 >). For grade I meningiomas p-STAT-3 activation index value was 30 (sd:30,22, range 0–100, med:20) and 17,03 (sd:25,66, range 0–90, med:5) for grade 2 meningiomas and it is statistically significant (Mann-Whitney p:0,021) (Table 1).

For recurrent meningiomas (n:26) mitosis index mean value was 2,85 (sd:2,98, range 0–8, med:1,5) and 1,66 (sd:1,98, range 0–10, med:1) for non-recurrent (n:76) meningiomas (Mann-Whitney p:0,139). For recurrent meningiomas Ki-67 mean value was 9,54 (sd:7,50, range 2–30, med:8) and 7,74 (sd:6,42, range 1–40, med:6) for non-recurrent meningiomas (Mann-Whitney p:0,198). For recurrent meningiomas PHH-3 proliferation index mean value was 3,08 (sd:4,52, range 0–14, med:0,5) and 2,04 (sd:2,42, range 0–10, med:1) for non-recurrent meningiomas (Mann-Whitney p:0,660). For recurrent meningiomas p-STAT-3 activation index mean value was 25,19 (sd:28,37, range 0–80, med:7,5) and 26,16 (sd:30,24, range 0–100, med:10) for non-recurrent meningiomas (Mann-Whitney p:0,697) (Table 2).

For progesterone receptor positive (n:74) meningiomas mitosis index mean value was 1,95 (sd:2,27, range 0–8, med:1) and 1,90 (sd:2,41, range 0–10, med:1) for receptor absent (n:30) meningiomas (Mann-Whitney p:0,941). For progesterone receptor positive meningiomas Ki-67 mean value was 8,18 (sd:7,12, range 1–40, med:6) and 7,87 (sd:5,64, range 1–30, med:6,5) for receptor absent meningiomas (Mann-Whitney p:0,583). For progesterone receptor positive meningiomas PHH-3 proliferation index mean value was 2,34 (sd:3,19, range 0–14, med:1) and 2,10 (sd:2,84, range 0–11, med:1) for receptor absent meningiomas (Mann-Whitney p:0,903). For progesterone receptor positive meningiomas p-STAT-3 activation index mean value was 25,99 (sd:29,41, range 0–100, med:10) and 25,67 (sd:29,73, range 0–90, med:10) for receptor absent meningiomas (Mann-Whitney p:0,370). There were no correlations between progesterone receptor with Ki-67 labeling index, mitosis index, PHH-3 and p-STAT3 (Table 3).

**Table 2**
Correlation between histopathological findings and tumor recurrence.

| | Tumor recurrence (n:26) | No tumor recurrence (n:76) | P Value |
|---|---|---|---|
| Mitosis Index | 2,85 | 1,66 | 0,139 |
| Ki-67 Value | 9,54 | 7,74 | 0,198 |
| PHH-3 Proliferation Index | 3,08 | 2,04 | 0,660 |
| p-STAT-3 Activation Index | 25,19 | 26,16 | 0,697 |

Two deceased patients were excluded from recurrence evaluation.

There is a strong correlation with PHH-3 and Ki-67 (Spearman's Rho, rs:0,563, **p:**0,001 >) and mitosis index (Spearman's Rho, rs:0,532, p:0,001 >) although there is no correlation with p-STAT-3 (Spearman's Rho, rs:-0,111, p:0,260) (Table 4).

There is a strong correlation with p-STAT-3 and Ki-67 (Spearman's Rho, rs:-0,243, p:0,013), although there is no correlation with mitosis index (Spearman's Rho, rs:-0,170, p:0,085) and PHH-3 (Spearman's Rho, rs:-0,111, p:0,260). There is a strong correlation with Ki-67 and mitosis index (Spearman's Rho, rs:0,579, p:0,001 >) (Table 4).

## 4. Discussion

Meningiomas are usually slow growing extra axial intracranial tumors that originate from meninges. Tumor location, extent of surgical resection (Simpson grading), brain invasion, and grading system including mitotic activity are responsible for the risk of recurrence and aggressive behavior of meningiomas [16]. According to recent WHO classification, meningiomas are divided to 3 groups histopathologically according to mitotic figures, brain invasion and cytoarchitectural characteristics [5,19]. However, this classification has limitations such as; independent recurrence rate from grading system, difficulties in identification of mitotic figures, distinction from other chromatin changes, and subjective selection of the highest mitotic area [13]. With the reasons of these disadvantages various biomarkers had emerged to predict prognosis regarding WHO classification of meningiomas.

In our study, we firstly investigate the relations of p-STAT-3 between Ki-67 labeling index and mitosis index with grading system and recurrence presence.

Histone H3 protein phosphorylation is a specific step during mitosis [20,21]. Histone H3 phosphorylation includes two structurally processes that formed by transcriptional activation and chromosome compaction during cell division [22]. For proper sequence of chromosomes on the metaphase plate histone H3 phosphorylation is crucial and it is responsible from metaphase to anaphase transition [22,23]. Dephosphorylation of Histone H3 starts with late anaphase and ends before chromosome decondensation in early telophase [24]. PHH-3 labeling is a strong method for detecting mitotic figures that offers more objective choice of mitotic index when compared to conventional counting mitosis per unit area [3]. In our study PHH-3 proliferation index has a strong correlation with Ki-67 labeling index and mitosis index although has no correlation with p-STAT-3 activation. PHH-3 is also significantly associated with WHO grading system, although not associated with tumor recurrence conversely to literature. When compared to recent and largest study performed by Winther et al. [13] similar results have been demonstrated excluding association between recurrence and PHH-3. They have been founded that recurrence free survival time is significantly associated with PHH-3 proliferation. We only compare the tumor recurrence presence between PHH-3 and found to be significantly.

STAT-3 is a member of phosphothyrosine protein family that acts like transcription factor on DNA binding domain. STATs play numerous role on cellular functions such as cell proliferation, apoptosis and angiogenesis. Some growth factors such as PDGF and EGF with IL-6 activate the latent STAT-3 in cell cytoplasm [25–27]. After activation of the latent STAT in cell cytoplasm by growth factor and cytokines result nucleus dimerization and translocation. JAK-STAT3 pathway and STAT3 activation by different pathways in meningiomas have been studied firstly by Magrassi et al. in 1999 [25]. Johnson et al. [18] in 2008 were reported the correlation between p-STAT-3 and grade II meningiomas. Magrassi et al. [25] have been founded that p-STAT-3 levels are significantly higher in 17 grade I meningioma than control dural tissue. Johnson et al. [18] were demonstrated the p-STAT-3 levels are higher in Grade II and grade III meningiomas when compared to grade I meningiomas.

**Table 3**
Histopathological findings by presence of progesterone receptor.

| | Progesteron Receptor Positive (n:74) | Progesterone Receptor Negative (n:30) | P Value |
|---|---|---|---|
| Mitosis Index | 1,95 | 1,90 | 0,941 |
| Ki-67 Value | 8,18 | 7,87 | 0,583 |
| PHH-3 Proliferation Index | 2,34 | 2,10 | 0,903 |
| p-STAT-3 Activation Index | 25,99 | 25,67 | 0,370 |

**Table 4**
Correlations between Ki-67, mitosis index, PHH-3 and p-STAT-3.

| | Mitosis Index | | PHH-3 | | STAT-3 | |
|---|---|---|---|---|---|---|
| | *P* value | Rs | *P* value | Rs | *P* value | Rs |
| PHH-3 | **0,001** | 0,532 | | | | |
| p-STAT-3 | 0,085 | −0,171 | 0,260 | −0,111 | | |
| Ki-67 Labeling | **0,001** | 0,579 | **0,001** | 0,563 | **0,013** | −0,243 |

Bold values provide statistically significant values.

Both expressions of p-STAT-3 and PHH-3 have been studied in meningiomas, however their role for the behavior of meningiomas has not been well identified. The rationale of this study evaluating the relationship between p-STAT-3 and PHH-3 was that there might be correlation between these two markers, since the evidence suggest that both p-STAT-3 and PHH-3 would contribute the diagnosis and the prognosis of patients with meningiomas. This would also provide data for future research to identify the role of p-STAT-3 for targeted therapies. However, we did not observe a correlation between p-STAT-3 and PHH-3 in the current research. Johnson et al also reported STAT-3 activation and risk of recurrence in meningioma in 2017. They have been founded that p-STAT3 has no correlation with grade I and grade II meningiomas and also has no correlation with tumor recurrence presence. This actual study contains small group of grade I and grade II meningiomas (6 Grade I and 7 Grade II). Although in our study of 71 of grade I and 33 of grade II meningiomas were examined. Conversely to Johnson et al our study demonstrates p-STAT3 and PHH-3 has a strong correlation with grade II meningiomas when compared to grade I meningiomas although similar to actual study there was no correlation with tumor recurrence presence statistically [28].

In the lighting of the literature this is the first study that demonstrates the correlations of p-STAT-3 with Ki-67 labeling, conventional mitotic index, new biomarker PHH-3 and tumor recurrence. In our study p-STAT-3 levels are founded to be significantly higher in grade II meningiomas than grade I meningiomas, although there was no statistically relation with tumor recurrence, Ki-67 labeling index, mitotic index and PHH-3 proliferation.

Progesterone receptor (PR) presents in to two third of meningiomas [29,30] however many studies have been performed to determine the correlation of PR with Ki-67 and mitotic index [29–33]. Iplikcioglu et al. [34] founded that there was no correlation between PR with Ki-67 and mitotic index. In our study we also obtain same results with Ki-67 and mitotic index, although correlation with PHH-3 and p-STAT-3 is firstly determined and there was no statistically significant relation were observed.

## 5. Conclusion

Findings from this study suggest that PHH-3 proliferation index is a new, easy process and a strong predictor for prognosis and may provide further contribution for treatment. p-STAT-3 activation is significantly higher in grade II meningiomas and has correlation between Ki-67 labeling, although there is no correlation with mitotic index, PHH-3 and tumor recurrence that demonstrates p-STAT-3 may not be a good predictor for prognosis as PHH-3. However, depending on the p-STAT-3 cell proliferation feature, inactivation of these pathways may predict new chemotherapies for grade II meningiomas.

## References


[1] A. Perry, D.N. Louis, B.W. Scheithauer, et al., Meningiomas, in: D.N. Louis, H. Ohgaki, O.D. Wiestler (Eds.), WHO Classification of Tumours of the Central Nervous System, IARC, Lyon, France, 2007, pp. 1641–1677.
[2] D.N. Louis, H. Ohgaki, O.D. Wiestler, et al., The 2007 WHO classification of tumours of the central nervous system, Acta Neuropathol. (114) (2007) 97–109.
[3] A. Olar, K.M. Wani, E.P. Sulman, et al., Mitotic index is an independent predictor of recurrence-free survival in meningioma, Brain Pathol. 25 (2015) 266–275.
[4] T. Backer-Grondahl, B.H. Moen, S.H. Sundstrom, et al., Histopathology and prognosis in human meningiomas, APMIS. 122 (2014) 856–866.
[5] D.N. Louis, A. Perry, G. Reifenberger, et al., The 2016 World Health Organization classification of tumors of the central nervous system: a summary, Acta Neuropathol. (131) (2016) 803–820.
[6] A. Matsuno, T. Fujimaki, T. Sasaki, et al., Clinical and histopathological analysis of proliferative potentials of recurrent and non-recurrent meningiomas, Acta Neuropathol. 91 (1996) 504–510.
[7] A. Perry, S.L. Stafford, B.W. Scheithauer, et al., The prognostic significance of MIB-1, p53, and DNA flow cytometry in completely resected primary meningiomas, Cancer. 82 (1998) 2262–2269.
[8] E. Abry, I.O. Thomassen, O.O. Salvesen, et al., The significance of Ki-67/MIB-1 labeling index in human meningiomas: a literature study, Pathol. Res. Pract. 206 (2010) 810–815.
[9] S.H. Torp, C.F. Lindboe, B.H. Gronberg, et al., Prognostic significance of Ki67/MIB-1 proliferation index in meningiomas, Clin. Neuropathol. 24 (2005) 170–174.
[10] J. Bruna, M. Brell, I. Ferrer, et al., Ki-67 proliferative index predicts clinical outcome in patients with atypical or anaplastic meningioma, Neuropathology 27 (2007) 114–120.
[11] D.M. Ho, C.Y. Hsu, L.T. Ting, et al., Histopathology and MIB-1 labeling index predicted recurrence of meningiomas: a proposal of diagnostic criteria for patients with atypical meningioma, Cancer 94 (2002) 1538–1547.
[12] G. Cattoretti, M.H. Becker, G. Key, et al., Monoclonal antibodies against recombinant parts of the Ki-67 antigen (MIB 1 and MIB 3) detect proliferating cells in microwave-processed formalin-fixed paraffin sections, J. Pathol. 168 (1992) 357–363.
[13] T.L. Winther, M.B. Arnli, Ø Salvesen, et al., Phosphohistone-H3 proliferation index is superior to mitotic index and MIB-1 expression as a predictor of recurrence in human meningiomas, Am. J. Clin. Pathol. 146 (2016) 510–520.
[14] G. Gauchotte, C. Vigouroux, F. Rech, et al., Expression of minichromosome maintenance MCM6 protein in meningiomas is strongly correlated with histologic grade and clinical outcome, Am. J. Surg. Pathol. 36 (2012) 283–291.
[15] Y.J. Kim, R. Ketter, W.I. Steudel, et al., Prognostic significance of the mitotic index using the mitosis marker anti-phosphohistone H3 in meningiomas, Am. J. Clin. Pathol. 128 (2007) 118–125.
[16] T. Ribalta, I.E. McCutcheon, K.D. Aldape, et al., The mitosis- specific antibody anti-phosphohistone-H3 (PHH3) facilitates rapid reliable grading of meningiomas according to WHO 2000 criteria, Am. J. Surg. Pathol. (28) (2004) 1532–1536.
[17] J. Bromberg, STAT proteins and oncogenesis, J. Clin. Invest. 109 (2002) 1139–1142.
[18] M.D. Johnson, M.O. Connell, F. Vito, et al., Increased STAT-3 and synchronous activation of Raf-1-MEK-1- MAPK, and phosphatidylinositol 3-Kinase-Akt-mTOR pathways in atypical and anaplastic meningiomas, J. Neurooncol. (92) (2009) 129–136.
[19] I. Elmaci, A.M. Altinoz, R. Sari, et al., Phosphorylated histone H3 (PHH3) as a novel cell proliferation marker and prognosticator for meningeal tumors: a short review, Appl. Immunohistochem. Mol. Morphol. (2017) 00:000–000.
[20] K. Shibata, K. Ajiro, Cell cycle-dependent suppressive effect of histone H1 on mitosis-specific H3 phosphorylation, J. Biol. Chem. 268 (25) (1993) 18431–18434.
[21] K. Shibata, M. Inagaki, K. Ajiro, Mitosis-specific histone H3 phosphorylation in vitro in nucleosome structures, Eur. J. Biochem./FEBS 192 (1) (1990) 87–93.
[22] F. Hans, S. Dimitrov, Histone H3 phosphorylation and cell division, Oncogene 20 (2001) 3021–3027.
[23] R. Pacaud, M. Cheray, A. Nadaradjane, et al., Histone H3 phosphorylation in GBM: a new rational to guide the use of kinase inhibitors in anti-GBM therapy, Theranostics 5 (2015) 12–22.
[24] A. Olar, Phosphohistone H3: implications for clinical practice and risk assessment in meningioma, Neuro Oncol. 17 (2015) 631–633.
[25] L. Magrassi, C. De-Fraja, L. Conti, et al., Expression of the JAK and STAT superfamilies in human meningiomas, J. Neurosurg. (91) (1999) 440–446.
[26] L. Klampfer, Signal transducers and activators of transcription (STATs): novel targets of chemo-preventive and chemotherapeutic drugs, Curr. Cancer Drug Targets

(6) (2006) 107–121.
[27] M. Kortylewski, Hua Y Stat3 as a potential target for cancer immunotherapy, J Immunotherapy (30) (2007) 131–139.
[28] M. Johnson, M. O'Connell, K. Walter, STAT3 activation and risk of recurrence in meningiomas, Oncol. Lett. 13 (2017) 2432–2436.
[29] P.E. Fewings, R.D. Battersby, W.R. Timperley, Long-term follow up of progesterone receptor status in benign meningioma: a prognostic indicator of recurrence? J. Neurosurg. 92 (2000) 401–405.
[30] A. Brandis, S. Mirzai, M. Tatagiba, et al., Immunohistochemical detection of female sex hormone receptors in meningiomas: correlation with clinical and histological features, Neurosurgery 33 (1993) 212–217.
[31] D.W. Hsu, J.T. Efird, E.T. Hedley-Whyte, Progesterone and estrogen receptors in meningiomas: prognostic considerations, J. Neurosurg. 86 (1997) 113–120.
[32] G. Nagashima, M. Aoyagi, H. Wakimoto, et al., Immunohistochemical detection of progesterone receptors and the correlation with Ki-67 labeling Indices in paraffin-embedded sections of meningiomas, Neurosurgery 37 (1995) 478–482.
[33] S. Pravdenkova, O. Al-Mefty, Sawyer, et al., Progesterone and estrogen receptors: opposing prognostic indicators in meningiomas, J. Neurosurg. 105 (2006) 163–173.
[34] A.C. Iplikcioglu, M.A. Hatiboglu, E. Ozek, et al., Is progesterone receptor status really a prognostic factor for intracranial meningiomas? Clin. Neurol. Neurosurg. (2014) 119–122.