

October 1999

# p53 and PCNA expression in benign, atypical and malignant meningiomas

R Ahmed
*Aga Khan University*, rashida.ahmed@aku.edu

I N. Soomro
*Aga Khan University*

S A. Aziz
*Aga Khan University*

S H. Hasan
*Aga Khan University*, sheema.hasan@aku.edu

# P53 and PCNA Expression in Benign, Atypical and Malignant Meningiomas

Pages with reference to book, From 241 To 243

Rashida Ahmed,Irshad Nabi Soomro,Syed Abdul Aziz,Sheema Habibul Hasan ( Department of Pathology, The Aga Khan University Hospital, Karachi. )

## Abstract

**Objective:** Alterations: p53 genes are turning out to be the most common genetic alterations in human cancers. Due to long half-life of mutated p53, its detection is possible by immunohistochemistry. Proliferating cejl nuclear antigen (PCNA) is expressed by dividing cells, hence has been shown to correlate with prognosis. We have used monoclonal antibodies protein DO-7 (p53) and PC1O (PCNA) to see whether their expression correlates with histological grading in meningethelial tumour.
**Material and Methods:** Twenty nine meningiomas (20 benign, 7 atypical and 2 malignant) were selected from the records of our laboratory. p53 and PCNA expression was sought by immunohistochemistry using Peroxidase Anti Peroxidase (PAP) technique.
**Results:** Four benign and 2 atypical meningiomas showed weak staining for p53. Both malignant meningiomas showed strong positivity for p53. Six benign meningiomas had less than 5% PCNA positivity, one 10% positivity and three showed 20% positivity. PCNA positivity ranged for 10-80% in atypical meningiomas. In two malignant meningiomas PCNA positivity was 70% and 90%.
**Conclusion:** It is worthwhile to include p53 and PCNA expression alongwith histologic assessment in predicting outcome of meningiomas. A larger series with complete follow-up is essential in assessing value of these markers which unfortunately remains a dream in our country (JPMA 49:241, 1999).

## Introduction

Alterations in p53 gene are turning out to be the most common genetic alterations in human cancer[1]. Increasing evidence attributes p53 dys-regulation a crucial role in the development of cancer and its prognosis[2]. The expression of mutated p53 has been documented in several epithelial and non-epithelial malignancies, including nervous system. This loss of tumor suppresser activity has also been seen in atypical and meningiomas[3-6].
PCNA is expressed in dividing cells and appears in the nucleas in late Gl phase. It peaks in S phase and subsides in G2 and M phases. Thus its level of expression correlates with cellular proliferative activity and DNA synthesis[5].
Assessment of prognosis in meningiomas is not easy by routine histo-pathological examination. We aimed at identifying criteria based on tumor markers, in the meningothelial tumors, which would help to predict the aggressive behaviour of some of these neoplasms. We carried out a prospective study at The Aga Khan University Medical Centre, Karachi to correlate the histological criteria for predicting aggressive behaviour of the tumor with expression of p53 and PCNA in benign, atypical and frankly malignant tumors.

## Materials and Methods

A total of 29 meningiornas were reported in the period ranging from June 1993 to June 1996, by the Histopathology Department of The Aga Khan University Medical Centre, Karachi. Out of these 20 had been reported as benign, 7 as atypical and 2 as malignant tumors. Out of the 20 benign tumors we

randomly selected every second case for the study whereas all the atypical and malignant meningiomas were included in the study.

The material received for examination was routinely processed, cut at 5m thickness and stained with the standard Haematoxylin and Eosin stain. Tumors were categorised in three groups, i.e. benign, atypical and malignant according to the scores which they secured by using the criteria of hypercellularity, nuclear pleomorphism, mitosis, necrosis, loss of architecture and evidence of brain invasion6. The tumors with scores of 0-4 were called benign, whereas tumors scoring more than 11 as malignant ones (Table).

**Table. Relationship of P53 and PCNA with histologic score of meningiomas.**

| Histologic score | P53 | PCNA % |
|---|---|---|
| a) Benign meningiomas (n=10) | | |
| 1 | - | <5 |
| 1 | - | <5 |
| 1 | + | <5 |
| 3 | - | <5 |
| 1 | + | <5 |
| 2 | - | <5 |
| 3 | + | <5 |
| 2 | ++ | 10 |
| 2 | - | 10 |
| 4 | - | 10 |
| b) Atypical meningiomas (n=7) | | |
| 6 | - | 5 |
| 5 | + | 5 |
| 10 | - | 20 |
| 8 | + | 25 |
| 9 | + | 25 |
| 6 | - | 30 |
| 8 | - | 40 |
| c) Malignant meningiomas (n=2) | | |
| 12 | ++ | 45 |
| 11 | ++ | 35 |

The tumors having a score of 5-11 were classified as atypical meningiomas.
The detection of p53 and PCNA was done on formalin fixed, paraffin embedded tissue sections by immunohistochemistry using PAP technique. The antigen retrieval was done by subjecting the sections to microwave heating. Primary antibodies against monoclonal mouse anti human p53 protein DO-7 and

monoclonal mouse anti-proliferating cell nuclear antigen (PCNA) PC10 (both by DAKO) were used followed by incubation with rabbit anti-mouse immunoglobulin, followed by final incubation with polyclonal mouse PAP tomplex. The colour development was done by Di-aminobenzidine. The brown nuclear staining was taken as positive for both the markers. Positivity for PCNA was calculated as number of nuclei/100 cells taking a brown stain whereas for p53, the blue nuclei were taken as negative, mild, brown staining was taken as +, moderate brown colour was ++ and a dark brown colour was considered as +++ (strongly positive). The scoring was done by 2 pathologists independently followed by a combined review of the material.

P53 and PCNA expression in different category of tumors is shown in table. Out of 10 benign meningiomas, 6 were completely negative for p53 and 4 showed very weak nuclear staining. Regarding PCNA, 7 benign tumors showed staining in less than 5% of the nuclei and 3 showed positivity in 10% of the nuclei. In the group of atypical meningiomas, 5 did not show positivity for p53 and 2 showed weak staining. The PCNA positivity in this group ranged from 5-40%. Only 2 malignant tumors were available and showed strong positivity for p53 with high proliferative activity as detected by PCNA ranging from 35% and 45% nuclei.

Tumor suppressor gene p53 is considered to be the policeman of the genome. When mutated it allows non lethal genetic defects to persist ultimately leading to neoplasia. p53 gene is frequently mutated in wide variety of human tumors. Structural rearrangements, deletions and point mutations, result in loss of normal p53 function. The tumor suppresser gene (p53) encodes 53 LD, nuclear phospho-protein, involved in cell cycle control, particularly, in transit from G0/G I to the S phase[2]. There have been few studies in which p53 mutation has been sought in meningiomas and those studies have been in context of progressively increased accumulation in benign, atypical and malignant meningiomas[3-6]. In this study, we studied mutated p53 protein expression in 19 meningiomas by doing immunohistochemistry. An attempt was made to

correlate p53 expression with number of cells in cycle as detected by the proliferation marker PCNA and correlating both the result with histological grading obtained on H & E staining. It appears that the expression of p53 be an uncommon event in benign and atypical meningiomas and only occurs when the malignant transformation has occurred. PCNA whose efficacy as a proliferation marker is well tested[7,8] showed correspondingly higher score in three categories of Meningiomas. However, there were 4 tumors in benign category with score of 10%. The score of atypical ranged from 5-40% and 2 malignant tumors showed a score higher than 35%. Although only follow up of these patients will confirm the potential of PCNA as a prognostic marker, it appears at this stage, that adequate categorisation can be done by this marker and will complement the scoring system in meningeal tumors and thus minimise the subjectivity and observer bias. The overlap category of atypical meningiomas with a higher score and PCNA ositivity needs follow up for recurrence. We admit that the sample size in this study is relatively small (specially the malignant meningiomas which are very rare tumors) but we think it is worth while to co-relate the three criteria (histological grading, p53 and PCNA) to predict the probable outcome of the histologically aggressi e menigiomas.

**References**

1. Arnold J, Jamil M, Cathy A Finalay. The p53 tumour suppressor gene. Nature, 1991:351:453-56.
2. Anna R, Massimo R, Eliana FA, et at. p53 over expression in the multi-step process of esophageal carcinogenesis. Am. J. Surg. Pathol., 1995;19:1419-22.
3. Wang JL, Zhang ZJ, Hartrnan SA, et at. Mutations in the p53 tumour suppressor gene in human meningiornas. Int. J. Cancer, 1995:64:223-28.
4. Ohgaki H, Eibl RFI, Schwab M, et at. Mutations of the p53 tumour suppressor gene in neoplasms of the human nervous system. Mol. Carcinog., 1993:8:74¬80.

5. Commentary by HK NG on atypical and malignant meningiomas. Advances in anatomic pathology vol 1 Raven Press Ltd., New York, 1994, pp. 44-48.
6. Prayson RA. Address Malignant Meningioma: a clinicopathologic study of 23 patients including MIBI and p53 immunohistocheniistry. Am. J. Clin. Pathol., 1996;105:719-26.
7. Arnold B, Gelb 0, Kamel W, et at. Estimation of tumour growth fractions in archival formalin fixed, paraffin embedded tissue anti PCNA/cycline monoclonal antibodies. Am. J. Pathol., 1992;141:1453-58.
8. Shih YH, Kadota Y, Sato K, et at. Immunohistochemical expression of proliferating cell nuclear antigen (PCNA) in intracranial glioma and mcningioma. Chung Hua I Hsueh Tsa Chih, 1994:53:193-97.