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P53 and Ki-67 expression in nasopharyngeal carcinomas

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ABSTRACT

This study was performed to investigate any relation between the histological subtypes of nasopharyngeal carcinomas (NPCs) and the expression of p53 and Ki-67 in NPCs. Fifteen cases which were diagnosed between 2000 and 2003 in Akdeniz University Faculty of Medicine, Department of Pathology and treated with radiotherapy were included in this study. The specimens were stained immunohistochemically with p53 and Ki-67. Twenty-six percent of the cases were undifferentiated carcinoma, 60% were non-keratinizing type and 14% were keratinizing type squamous cell carcinoma (SCC). Ki-67 labeling index was as follows; 11% in undifferentiated carcinomas, 27% in non-keratinizing type and 32% in keratinizing SCC. We found +3 p53 intensity in 25% of undifferentiated carcinomas, 37% of the non-keratinizing SCC and in both of the keratinizing SCC. Although limited number of cases were included in this study, we found correlation between histopathologic subtypes, p53 and Ki-67 expression in NPCs. [Turk J Cancer 2004;34(3):106-109]

KEY WORDS:

p53, Ki-67, nasopharyngeal carcinoma

INTRODUCTION

Nasopharyngeal carcinoma (NPC) is the most common epithelial malignancy of the nasopharynx. NPCs are histologically typed as non-keratinizing, keratinizing and undifferentiated carcinomas. There is strong relation between NPC and EBV infection (1).

There are different clinical outcomes in patients with similar stages of NPC. NPCs are highly radiosensitive tumors. Conventional clinicopathologic parameters are insufficient to predict the prognosis of patients with NPCs treated with radiotherapy and chemotherapy. Thus, new prognostic factors that could reflect the clinical outcome of these patients are needed to be established.

Mutation of the p53 tumor suppressor gene is considered the most common aberration in many cancers. The product of this gene is a nuclear protein thought to be involved in the control of the cell cycle, apoptosis and the maintenance of genomic stability (2). Ki-67, a proliferation antigen, evaluates the proliferative activity of a lesion. The objective of this study was to evaluate the expression of the tumor suppressor gene p53 and Ki-67 labeling index (LI) as prognostic parameters in patients with NPC.

MATERIALS AND METHODS

Histopathologically typical cases of NPC were retrieved from the files of Akdeniz University Faculty of Medicine,

Department of Pathology. Formalin-fixed and paraffin embedded biopsies from 20 NPC cases obtained between 2000 and 2003 who received radiotherapy were analysed. Histological typing followed the WHO classification. 5 cases were excluded because of scanty material in original paraffin blocks.

Immunohistochemical staining was done by the streptavidin-biotin complex method. Formalin-fixed, paraffin-embedded 4μm-thick sections were immunohistochemically stained for the p53 oncoprotein (p53 protein, clone DO-7, Dako, Denmark, code no: M7001) and monoclonal antibody to the Ki-67 nuclear antigen (Ki-67, polyclonal, Dako, Denmark, code no: A0047).

The p53 and Ki-67 immunoreactivities were assessed using a light microscope. In areas having the highest nuclear labeling density, the percentage of tumor nuclei expressing Ki-67 was determined by counting 400 cells per slide, and the percentage was expressed on the available cells. In the case of Ki-67, the quantitative labeling index was used. We scored p53 expression as negative (-), meaning that staining was observed in <5% of the tumor cells, or positive (+), meaning that staining was observed in 5% or more of the tumor cells. The intensity of nuclear staining (taking as a standard the intensity of staining in positive tumor-infiltrating lymphocytes) was assessed and scored as -, +, +2, +3 (none, weak, marked, and strong staining, respectively). Cytoplasmic staining was not considered.

RESULTS

The profile of the patients and the immunohistochemical findings are given in table 1. Two of 15 patients were female. Follow-ups ranged from 4 to 30 months (average 17,6 months). All patients are still alive. 26% of cases were undifferentiated NPC (Figure 1), 60% were non-keratinizing (Figure 2) and 14% were keratinizing squamous cell carcinomas (SCC) (Figure 3).

P53 staining intensity was +2 in 50% of undifferentiated carcinomas and +3 in 25% of these cases. Thirty-seven percent of non-keratinizing SCCs and 2 keratinizing SCC

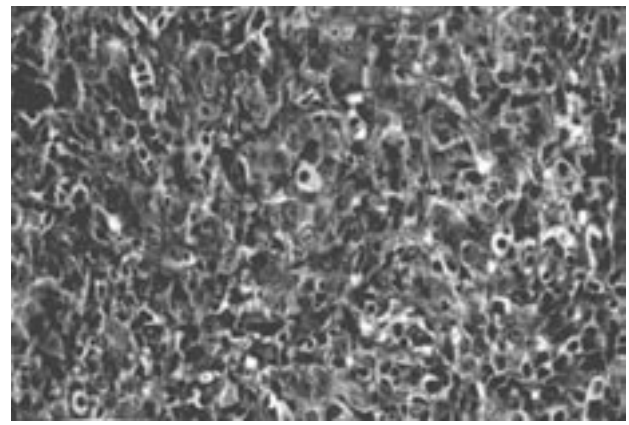

Fig 1. Histopathological appearance of undifferentiated carcinoma, H&E, x400

Table 1
The profile of the patients and the immunohistochemical findings

Sex	Follow-up (mo)	Histological type	p53(%)	Intensity of staining	Ki67 LI (%)
M	30	Undifferentiated carcinoma	10	+2	10
F	28	SCC, keratizing	10	+3	5
M	6	SCC, non-keratinizing	5	+2	10
M	8	SCC, keratinizing	90	+3	60
M	29	Undifferentiated carcinoma	10	+1	5
M	27	SCC, non-keratinizing	40	+2	10
M	24	SCC, non-keratinizing	90	+3	60
F	25	SCC, non-keratinizing	90	+2	40
M	22	SCC, non-keratinizing	70	+3	15
M	16	SCC, non-keratinizing	30	+2	10
M	17	SCC, non-keratinizing	60	+2	35
M	14	SCC, non-keratinizing	80	+3	25
M	7	SCC, non-keratinizing	40	+2	25
M	4	Undifferentiated carcinoma	50	+3	10
M	7	Undifferentiated carcinoma	40	+2	10

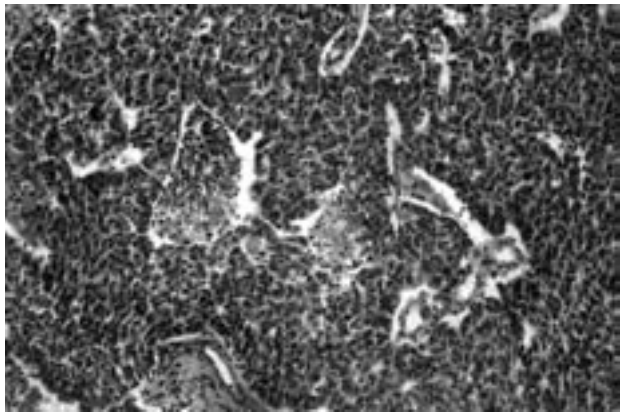

Fig 2. Histopathological appearance of non-keratinizing SCC, H&E, x200

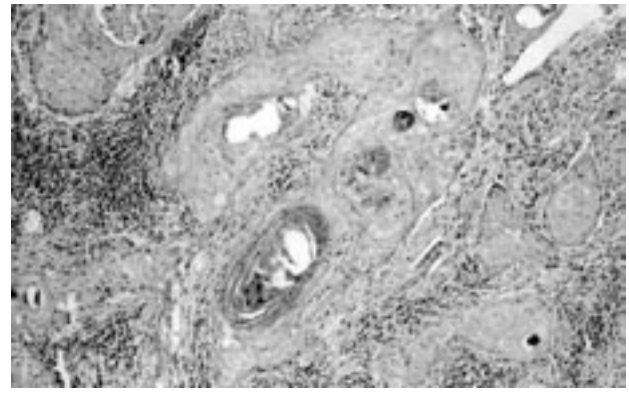

Fig 3. Histopathological appearance of keratinizing SCC, H&E, x100

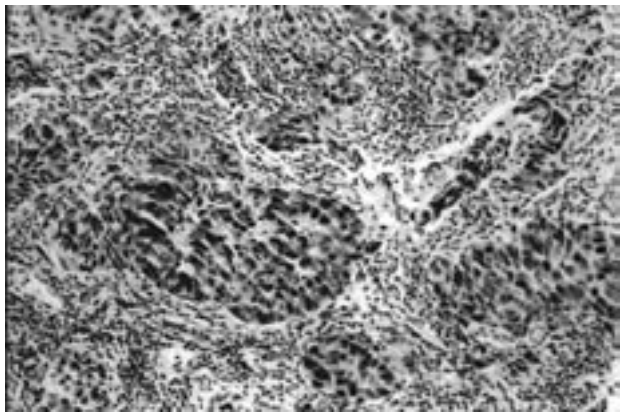

Fig 4. p53 immunoreactivity in non-keratinizing SCC, x200

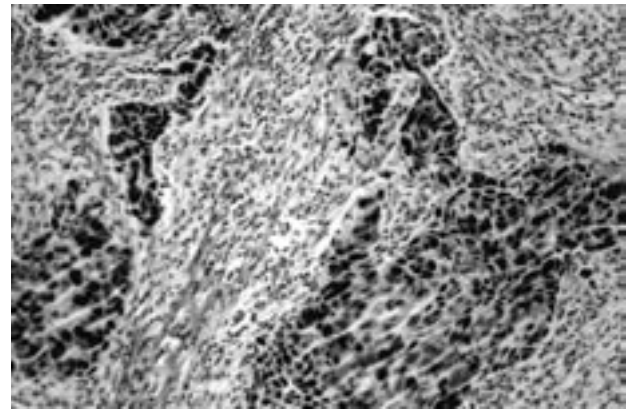

Fig 5. Ki-67 immunoreactivity in keratinizing SCC, x400

cases (100%) had +3 nuclear staining intensity for p53 (Figure 4).

Ki-67 LI was 11% for undifferentiated carcinomas, 27% for non-keratinizing SCCs and 32% for well differentiated SCCs (Figure 5).

DISCUSSION

We studied 15 NPC cases to establish correlation between p53 expression, Ki-67 LI and histopathological subtypes. The tumor suppressor gene, p53, regulates cell proliferation and apoptosis. It is the most common mutation found in many cancers. P53 protein overexpression was observed in the nuclei of tumoral cells. Cases were divided into two groups according to their immunoreactivity intensity and percentage of p53 positive tumor cells.

Li et al. (3) stated that p53 dysfunction in NPCs may be involved in the regulation of NPC cell growth. Gulley et al. (4) evaluated the association between EBV infection and p53 accumulation in NPCs. They found that EBV-infected tumors are more likely to express p53 than are tumors lacking EBV, and they concluded that there is a strong correlation between the presence of EBV and the accumulation p53 in tumor cells.

Masuda et al. (5) studied p53 and Ki-67 expression in NPCs and they found no association between the Ki-67 LI and clinicopathological parameters, radiosensitivity, distant metastases, or survival of patients with NPCs, suggesting that, in these tumors, the Ki-67 level lacks clinical significance. However, they suggested that the overexpression of p53 is a clinical prognostic factor associated with resistance to treatment with agents that damage DNA in patients with NPCs.

We observed strong positivity for p53 in keratinizing SCCs and weaker staining in undifferentiated cases but the number of our cases is not enough to make a conclusion.

Ki-67 is a nuclear antigen that is expressed in proliferating cells during the G1, S, G2 and M phases of the cell cycle. Its expression is used as a marker cell proliferation. Many studies have confirmed in a variety of human malignant tumors between a high Ki-67 LI and a poor prognosis. However, other studies, in prostate cancer and carcinomas of head and neck have shown no correlation between Ki-67 index and prognosis (5).

Genc et al. (6) also studied p53 and Ki-67 in NPCs. According to them, a high percentage of p53 and Ki-67 expression was associated with the stage and tumor behavior. Yazici et al. (7) also found similar results with p53.

In our study the Ki67 LI was higher in keratinizing SCCs which have the worst prognosis and it was least in undifferentiated cases which are known to have the best response to therapy.

Although limited number of cases are included in this study, we found correlation between histopathologic subtypes, p53 and Ki-67 expression in NPCs. The Ki-67 LI was higher in cases with high percentage of p53 positive tumor cells.

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