VIA MEDICA

SHORT COMMUNICATION

Vol. 65, No. 1, pp. 95–98

ISSN 0015–5659
www.fm.viamedica.pl

An evaluation of Ki-67 and PCNA expression in conjunctival and eyelid tumours

Joanna Reszeć, Luiza Kańczuga-Koda, Mariola Sulkowska, Mariusz Koda, Jacek Cylwik, Małgorzata Barwijuk-Machała, Stanisław Sulkowski

Department of Pathology, Medical University, Białystok, Poland

[Received 17 September 2003; Accepted 20 December 2003]

The aim of our study was an evaluation of the expression of cell proliferation markers (PCNA and Ki-67) in conjunctival and eyelid papillomas and squamous and basal cell cancers. A series of 9 cases of squamous cell cancer (SCC), 15 cases of basal cell cancer (BCC) and 43 cases of squamous cell papilloma (SCP) were assessed using the immunohistochemical method with monoclonal antibodies. PCNA overexpression was observed in 100% of SCP, in 88.8% of SCC and in 100% of BCC cases. Ki-67 overexpression was seen in 32.5% of cases of SCP, in 22.2% of SCC and in 66.6% of BCC. The results showed that an evaluation of Ki-67 expression is the most valuable cell proliferation marker.

Key words: Ki-67, PCNA, papilloma, squamous cell cancer, basal cell cancer of conjunctiva and eyelid

INTRODUCTION

Conjunctival and eyelid papilloma, as well as squamous and basal cell cancers, are common tumours of the epithelium and are known to occur in both children and adults. PCNA (proliferating cell nuclear antigen) is a co-factor for DNA polymerase delta in S phase and also during DNA synthesis associated with DNA damage repair mechanism. PCNA expression is a useful marker of cell proliferation in both normal and neoplastic tissues [4]. Ki-67 expression occurs during the phase of the cell cycle designated as late G1, S, M, and G2, while during the G0 phase, the antigen cannot be detected [1]. The aim of our study was to evaluate Ki-67 and PCNA expression in squamous cell papilloma as well as in squamous and basal cell cancers. There are no data related to Ki-67 and PCNA expression in conjunctival and eyelid papilloma.

MATERIAL AND METHODS

A series of 67 benign and malignant conjunctival and eyelid lesions were examined, comprising 9 cases of squamous cell cancer (SCC), 15 cases of basal cell cancer (BCC) and 43 cases of squamous cell papilloma (SCP). Material for histopathological examination was fixed in formalin, routinely processed and paraffin embedded. Immunohistochemical analyses were carried out using monoclonal antibodies DAKO/ /PCNA, M0879; DAKO/Ki-67, M7240. The reaction performed in LSAB was followed by the use of chromogen AEC to visualise the antigen/antibody complex. Scores in SCC and BCC groups were based on the following scale: (–) below 10% of the cells showing positive reaction for PCNA and Ki-67 protein, and (+) above 10% of the cells with positive immunostaining. In the papilloma group we defined as (–) negative immunostaining for the proteins examined or when Ki-67 and PCNA expression was restricted only

Address for correspondence: Joanna Reszeć, Department of Pathology, Medical University, ul. Waszyngtona 13, 15–269 Białystok, Poland, e-mail: joasia@zpk.amb.edu.pl

to the basal layer. PCNA and Ki-67 protein expression was defined as (+) in both the basal and parabasal layers and in the whole thickness of the epithelium. The values obtained were subjected to statistical analysis with the use of the Fisher or chi-square test. Values at $p < 0.05$ were considered significant.

RESULTS AND DISCUSSION

Both PCNA and Ki-67 protein expression was restricted to the nucleus. In the papilloma group (SCP), with ages ranging from 18 to 94 years (mean age 55.8), most cases were localised on the eyelid. Cellular dysplasia was observed in 7 cases of SCP. PCNA protein expression was observed in 43 cases (100%). In the whole thickness of the epithelium PCNA expression was noticed in 11 out of 43 cases of SCP (25.5%) (Fig. 1, 2). In papilloma with dysplasia PCNA expression was restricted to the basal and parabasal layer in 6 out of 7 cases and only in 1 case was positive immunostaining for PCNA seen in the whole thickness of the epithelium. Ki-67 protein expression was assessed as positive in 14 out of 43 cases (32.5%) (Fig. 3). Only in 2 out of 7 cases of papilloma with cellular dysplasia was Ki-67 expression seen and only in the basal and parabasal layers. In the SCC group ages ranged from 42 to 82 years and in the BCC group from 44 to 87 years (the mean age for both groups was 64.9 years). SCCs were evaluated using histological tumour grading (G stage) as G1, G2 and G3, with 3 cases at each stage. In G2 and G3 tumours mainly tricholemmal differentiation was observed and pT ranged from pT2 to pT3. In the SCC group a PCNA positive reaction was observed in 8 out of 9 cases (88.8%), in 2 out of 3 cases at the G1 stage and in 3 out of 3 cases at the G2 stage and G3 stage. Ki-67 protein positive immunostaining was seen only in 22.2% (2 out of 3 cases at the G3 stage) (Fig. 4). The BCC group was divided into 3 subgroups: nodular (4 cases), infiltrative (1 case) and mixed (10 cases). In the BCC group PCNA expression was noticed in 100% of cases: 4 cases of the nodular type, 1 case of infiltrative and 10 cases of the mixed type of basal cell carcinoma (Fig. 5). Ki-67 protein positive reaction was shown in 66.6%: 4 cases of the nodular type of BCC, 1 case of infiltrative and 5 cases of the mixed type of basal cell cancer (Fig. 6).

There is no data connected with Ki-67 and PCNA expression in conjunctival and eyelid papilloma. However Lu et al. [4] showed PCNA overexpression in the basal layer in 20 cases of skin warts, in 32 cases in the basal and parabasal layers and in 36 out of 90 cases of skin warts in the whole thickness of epithelium. In our prior study strong PCNA positive immunostaining was noticed mainly in the basal and parabasal layers of papilloma, as well as in cells of superficial layers in the papilloma with dysplasia [6]. Kręcicki et al. [3] observed PCNA overexpression in 69.9% of dysplastic lesions of the larynx. Rabah et al. [5] observed in laryngeal papillomatosis Ki-67 indices ranging from 9% to 40%. Most of the stained cells were in the basal layer. Koide et al. [2] observed locally PCNA-positive cells in well differentiated and moderately differentiated SCCs of the oesophagus. The PCNA labelling index was significantly higher in the basal cell cancer than in SCC. Stainbeck et al. [7] observed increased PCNA immunoreactivity in highly differentiated squamous cell cancer while, in contrast, more poorly differentiated carcinomas showed strongly elevated proliferative activity throughout the entire tumour mass. Barret et al. [1] showed PCNA expression in more cases than Ki-67 in all types of BCC (nodular, infiltrative, sclerosing and metatypical). A comparison of the immunoreactivity of PCNA and Ki-67 protein in conjunctival and eyelid squamous cell papillomas and squamous and basal cell cancers might suggest that the evaluation of Ki-67 expression is a more particular marker of cell proliferation than PCNA expression. It would seem expedient, therefore, to adapt differentiated standards of proliferate activity and also the markers examined by us in the evaluation of cell proliferation in conjunctival and eyelid carcinogenesis.

ACKNOWLEDGEMENTS

This work is supported by Grant No 3 PO5B 02123 from the State Committee of Scientific Research, Poland.

REFERENCES

1. Barret TL, Smith KJ, Hodge JJ, Butler R, Hall FW, Skelton HG (1997) Immunohistochemical nuclear staining for p53, PCNA, and Ki-67 in different histologic variants of basal cell carcinoma. J Am Acad Dermatol, 37: 430–437.
2. Koide N, Koike S, Watanabe H, Yazawa K, Adachi W, Amano J (1999) Basaloid squamous carcinoma of the esophageus with analysis by in situ nick end labeling and PCNA immunostaining. Hepatogastroenterology, 46: 265–271.
3. Kręcicki T, Jeleń T, Zalesska-Kręcicka M, Szkudlarek T, Szajowski K (1999) Immunohistochemically stained markers (p53,PCNA,bcl-2) in dysplastic lesions of the larynx. Cancer Lett, 143: 23–28.

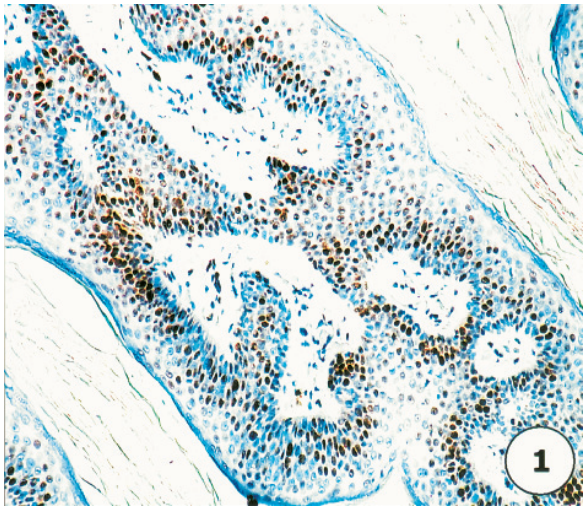

Figure 1. PCNA overexpression observed in the basal and parabasal layers of epithelium in eyelid squamous cell papilloma. Magn. 100 ×.

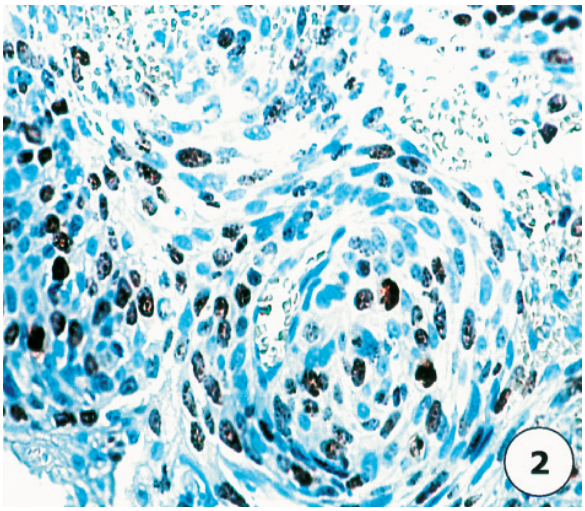

Figure 2. Ki-67 nuclear positive immunostaining seen in squamous cell cancer with tricholemmal differentiation of the eyelid. Magn. 100 ×.

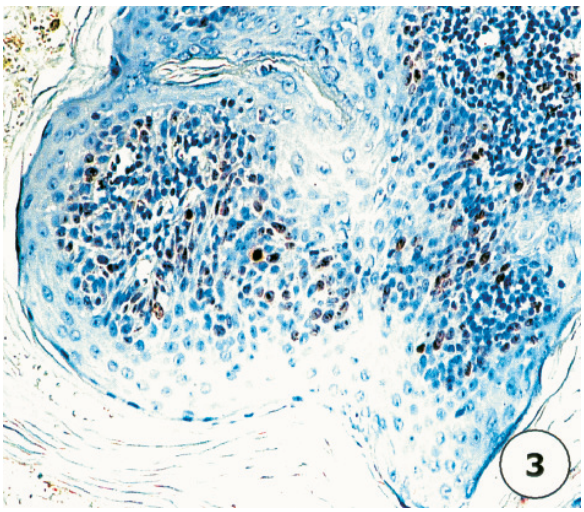

Figure 3. Ki-67 nuclear overexpression in squamous cell papilloma of the eyelid seen in the basal and parabasal layers. Magn. 100 ×.

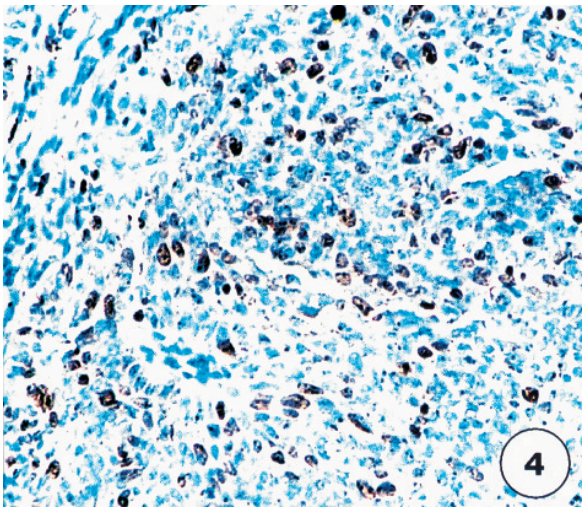

Figure 4. Ki-67 nuclear immunostaining seen in squamous cell cancer. Magn 200 ×.

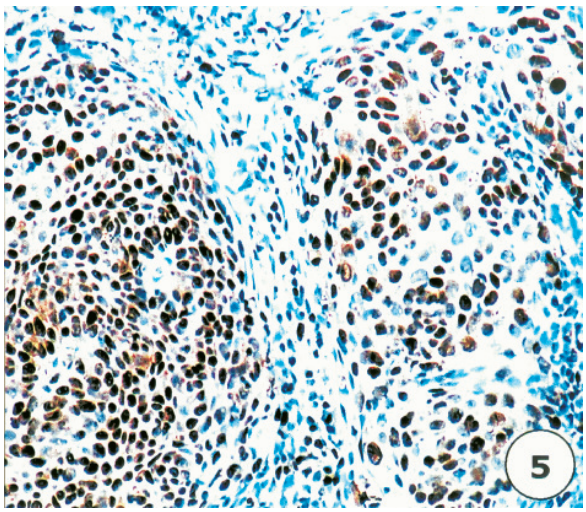

Figure 5. PCNA positive immunostaining observed in basal cell cancer. Magn. 200 ×.

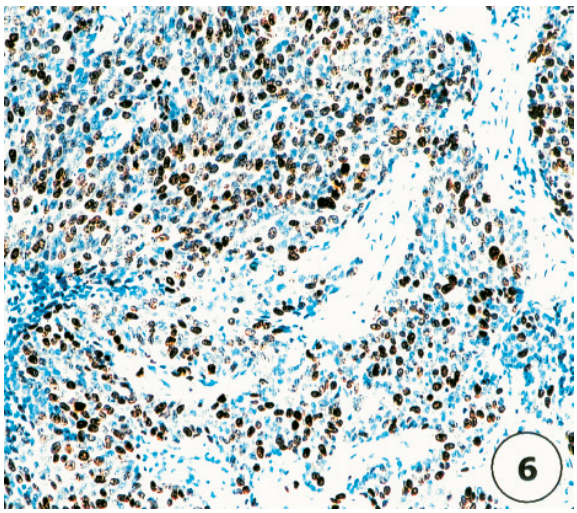

Figure 6. Ki-67 nuclear immunostaining observed in basal cell cancer. Magn. 100 ×.

4. Lu S, Syrjanen K, Havu VK, Syrjanen S (1996) Expression of PCNA is associated with the presence of HPV DNA in skin warts. Arch Dermatol Res, 289: 35–39.
5. Rabah R, Sakr W, Thomas R, Lancaster WD, Gregoire L (2000) Human papillomavirus type, Proliferative activity and p53. Arch Pathol Lab Med, 124: 721–724.
6. Reszec J, Sulkowska M, Famulski W, Guzinska-Ustymowicz K, Sulkowski S (2002) The expression of tumorigenesis markers in oral papilloma. Pol J Pathol, 53: 195–200.
7. Stainbeck RG, Moege J, Heselmeyer KM, Klebe W, Neugebauer W, Borg B, Auer GU (1993) DNA content and PCNA immunoreactivity in oral precancerous and cancerous lesions. Eur J Cancer B Oral Oncol, 29: 279–284.