ORIGINAL ARTICLE



## EXPRESSION FEATURES OF HUMAN PAPILLOMA VIRUS TYPE 16 AND ANTI-EPSTEIN-BARR VIRUS IN PLEOMORPHIC ADENOMA, SURROUNDING AND INTACT SALIVARY GLAND

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## **ABSTRACT**

The aim is to reveal the expression features of MCA to human papilloma virus type 16 and anti-Epstein-Barr virus in the pleomorphic adenoma, surrounding and intact salivary gland.

Materials and methods: It was used surgical and biopsy material from 30 patients, represented by pleomorphic adenomas with surrounding to tumor tissue of the salivary gland and intact tissue of the salivary gland (the distance between the tumor and the intact salivary gland – 10 mm). Immunohistochemical study was performed using mouse monoclonal antibody (MCA) to human papilloma virus type 16 (clone CAMVIR-1, «Diagnostic BioSystems», USA) and anti-Epstein-Barr virus (LMP, clone CS. 1-4, «Dako», Denmark). Visualization was performed, using an EnVision<sup>TM</sup> FLEX detection system (Dako, Denmark). Antigen unmasking was carried out in citrate buffer pH 6.0 at 95°C. Primary antibodies were incubated at room temperature for 30 minutes, secondary antibodies — 20 minutes. Sections were counterstained with Gill hematoxylin. We assessed the immunohistochemical reaction by a semi-quantitative method by counting the percentage of positively stained cells in the field of view of a microscope × 400. Microspecimens were studied and photoarchived on an Olympus BX-41 microscope (Japan).

Results: In this study it was detected a positive immunohistochemical reaction with MCA to human papilloma virus type 16 and anti-Epstein-Barr virus, respectively, in 26 (86.7%) and 8 (26.7%) cases. Epithelial, mixed and mesenchymal variants of pleomorphic adenoma of the salivary glands are characterized, respectively, by the severely expressed, moderately expressed and minimally expressed of MCA to human papilloma virus type 16 and anti-Epstein-Barr virus. The parenchymal component of pleomorphic adenoma is characterized by more marked expression of these markers as compared to the stromal component.

The epithelial cells of the salivary glands, surrounding the pleomorphic adenoma, as well as intact salivary glands, express MCA to human papilloma virus type 16 and anti-Epstein-Barr virus. The severity of the expression of these markers in the salivary gland is determined by the histological variant of the tumor (severely expressed in the epithelial variant, moderately expressed in the mixed variant, and minimally expressed in the mesenchymal variant).

Conclusions: The immunohistochemical study has shown that the Epstein-Barr virus and, especially, human papilloma virus type 16 can act as exogenous trigger factors involved in the development of pleomorphic adenoma of the salivary glands. The revealed immunohistochemical features of MCA expression to human papilloma virus type 16 and anti-Epstein-Barr virus in the salivary gland surrounding the pleomorphic adenoma and in the intact tissue of the salivary gland make it possible to recommend the extracapsular dissection of the tumor with resection of the adjacent intact tissue of the salivary gland at a distance of 10 mm in patients with pleomorphic adenoma.

KEY WORDS: anti-Epstein-Barr virus, human papilloma virus type 16, immunohistochemistry, pleomorphic adenoma, surrounding the tumor and intact salivary gland

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## INTRODUCTION

The problem of virus-associated oncogenesis is extremely relevant and is of scientific and practical interest for doctors of various specialties. Prevailing virus-associated tumors varies greatly across the world [1]. The study of viral nature of human tumors has continued for many years. However, today there are many controversial and unresolved issues related to the search for causal relationships and significant correlations between viral infection and the tumors manifestation [2].

Virus-associated tumors include pleomorphic adenoma of the salivary glands. The latter is the most common

benign salivary glands neoplasm, accounting for 33.2% to 68.4% of all cases [3]. This tumor most commonly occurs in the parotid gland but it may involve submandibular, lingual and minor salivary glands, too [4].

The human papilloma virus type 16 and the Epstein-Barr virus play a significant role in the genesis of pleomorphic adenomas of the salivary glands development, showed in our earlier morphological studies [5] and the works of other scientists [6]. Immunohistochemical research methods using monoclonal antibodies (MCA) play an important role in establishing the viral genesis of pleomorphic adenomas development. To date, the question of