**ABSTRACT**

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Title of diploma thesis: DNA methylation changes in oropharyngeal cancer

Oropharyngeal carcinoma (OPC) is a type of head and neck cancer (HNC) that represents the seventh most common malignancy worldwide. The vast majority (more than 90%) of cases are squamous cell carcinomas (SCC). OPC develops in the tissue of the tongue, tonsils, soft palate, and pharynx. In addition to traditional risk factors, human papillomavirus (HPV) has been identified as an additional independent risk factor for the development of these tumors.

Epigenetic alterations refer to heritable changes in gene expression that occur without changes in the underlying DNA sequence and can contribute to carcinogenesis. They include DNA methylation, histone modification and non-coding RNAs effecting gene expression.

This study aimed to investigate methylation levels of selected tumor-suppressor genes in oropharyngeal squamous cell carcinoma (OPSCC) in comparison to normal oropharyngeal tissue. DNA methylation levels of selected tumor-suppressor genes were analyzed using Methylation-Specific Multiplex Ligation-dependent Probe Amplification (MS-MLPA) in metastatic tumor samples, corresponding metastases samples, non-metastatic tumor samples and control tissue samples (non-cancerous palatine tonsils). Using a 15% cut-off for methylation we observed statistically significant higher methylation in the *PAX5*, *CADM1*, *WT1* ($P<0.01$) and *RARβ*, *PAX6* ($P<0.05$) genes of patients with OPC compared with the control group.

Based on results from MS-MLPA and literary review focused on hypermethylation of some promoter regions associated with HPV in cancer of the cervix, we chose analysis of *CADM1* gene using methylation-specific high-resolution melting analysis (MS-HRM). *CADM1* methylation in OPSCC is increased by the presence of HPV infection, which is corresponding to methylation of the *CADM1* gene in the cervix carcinoma. Based on our results it seems that *CADM1* gene methylation levels are probably not influenced by the metastatic process, because methylation levels of corresponding metastases have the same methylation status as their primary tumors. All control samples were unmethylated which also applies to all HPV negative cancer samples.

The findings of this study show promising candidates for prognostic OPSCC biomarkers and may have implications for future individualized therapies based on epigenetic changes.