PART 1: PREDICTION PRIOR TO TREATMENT
CHAPTER 2

INTERACTION OF QUANTITATIVE $^{18}$F-FDG-PET-CT IMAGING PARAMETERS AND HUMAN PAPILLOMAVIRUS STATUS IN OROPHARYNGEAL SQUAMOUS CELL CARCINOMA

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ABSTRACT

Background. Patients with human papillomavirus (HPV)-positive oropharyngeal squamous cell carcinoma (OPSCC) have a better survival than HPV-negative OPSCC. $^{18}$F-fluorodeoxyglucose positron emission tomography-computed tomography ($^{18}$F-FDG-PET-CT) may also provide prognostic information. We evaluated glycolytic characteristics in HPV-negative and HPV-positive OPSCC.

Methods. Forty-four patients underwent pretreatment $^{18}$F-FDG-PET-CT. Standardized Uptake Values (SUV) and metabolic active tumor volume (MATV) were determined for primary tumors. HPV-status was determined with p16 immunostaining, followed by high-risk HPV DNA detection on the positive cases.

Results. Twenty-seven patients were HPV-positive (61.4%). Median MATV was 2.8 millilitres (ml) [1.6-5.3] for HPV-positive and 6.1 ml [4.5-21.2] for HPV-negative tumors ($p<0.001$). SUV-values are volume dependent (partial volume effect), therefore, MATV was included as covariate in multivariate analysis. The maximum SUV in HPV-positive was 3.9 units lower than in HPV-negative tumors ($p=0.01$).

Conclusion. $^{18}$F-FDG-PET-CT parameters are lower in HPV-positive than in HPV-negative patients. Low pretreatment SUV-values in HPV-positive OPSCC may be explained by HPV-induced tumor changes.
INTRODUCTION

Head and neck squamous cell carcinoma (HNSCC) is the sixth most common cancer worldwide. Tobacco smoking and alcohol consumption are known risk factors for the development of HNSCC, but over the past several years, human papillomavirus (HPV) has been recognized as a major etiological factor in the development of particularly squamous cell carcinomas in the oropharynx (OPSCC). HPV-positive and HPV-negative OPSCC are distinct disease entities. Patients with HPV-positive OPSCC show more favourable treatment response rates and prognosis as compared with HPV-negative OPSCC, despite presenting with regionally more advanced disease. In addition, HPV-related tumors have distinct histological features and have a different genetic route to cancer. In this context, traditional prognostic factors such as tumor size and lymph node invasion provide insufficient risk classification. Identification of other prognostic tumor characteristics may lead to individually customized treatment and thereby higher responses to treatment and less treatment-induced side effects.

Positron emission tomography-computed tomography (PET-CT) combines functional imaging with anatomical localisation. Fluoro-2-deoxyglucose (\(^{18}\text{F}-\text{FDG}\)), a radiolabelled glucose analogue, is the most widely used tracer in oncologic PET studies and enables imaging of metabolically active tissues. Due to increased glycolytic activity, higher concentrations of \(^{18}\text{F}-\text{FDG}\) accumulate in malignant tumors. The standardised uptake value (SUV) is a semi-quantitative measurement of tracer uptake. It has been suggested that patients with high pretreatment SUV-values generally have a less favourable outcome, so it seems that a high rate of glucose metabolism is a characteristic of aggressive tumor behaviour. New functional imaging parameters, such as metabolic active tumor volume (MATV) and total lesion glycolysis (TLG), also have been shown to be of prognostic value in OPSCC and may be additional biomarkers. MATV is the \(^{18}\text{F}-\text{FDG}\)-avid tumor volume and TLG combines SUV\(_{\text{mean}}\) and MATV.

HPV-positive tumors are genetically different from HPV-negative OPSCC but it is largely unknown how they behave with regard to glucose metabolism and whether this plays a role in prognosis. Some conflicting results have been reported on the relation between \(^{18}\text{F}-\text{FDG}-\text{PET-CT}\) parameters and HPV-status. We studied pretreatment \(^{18}\text{F}-\text{FDG}-\text{PET-CT}\) with a uniform dedicated head and neck protocol in a group of well-defined HPV-positive tumors and compared this with a group of HPV-negative OPSCC. HPV-positivity was defined according a validated algorithm, p16 staining followed by high-risk HPV-DNA detection. The objective of this study was to provide supplemental information about differences in glycolytic
characteristics as measured with pretreatment $^{18}$F-FDG-PET-CT between HPV-negative and HPV-positive OPSCC.

**MATERIALS AND METHODS**

*Patients and study design*

Between January 2010 and December 2013, all consecutive patients with a histopathologically proven OPSCC were retrospectively screened for the following inclusion criteria: patients who had undergone a dedicated head and neck $^{18}$F-FDG-PET-CT imaging for tumor staging and patients with a $>$T1 oropharyngeal tumor because it was expected that a reliable volume of interest (VOI) could not be drawn in patients with a T1 tumor. This retrospective study was approved by the institutional review board of the VU University Medical Center, with waiver of informed consent (data were analysed anonymously). Forty-seven patients with a pretreatment $^{18}$F-FDG-PET-CT could be evaluated. Medical records were reviewed for clinical characteristics, including smoking and alcohol intake, TNM-stage and oropharyngeal subsite. Three patients were excluded because the glucose level could not be retrieved. Thus, 44 patients were enrolled in the analysis.

*HPV analysis*

HPV testing was performed with our previously defined and validated test algorithm for HPV detection $^{20,21}$. In short, formalin-fixed and paraffin-embedded tumor tissue was stained by immunohistochemistry for p16 (product of the CDKN2A) and on the p16-immunopositive cases high-risk HPV-DNA was detected with GP5+/6+ polymerase chain reaction (PCR). Only the cases that were positive in the latter assay as well, were classified as HPV-positive.

$^{18}$F-FDG-PET-CT

All patients fasted for at least 6 hours. Mean serum glucose levels were 5.9 mmol/l (range from 4.1 to 10.2 mmol/l). PET-CT was started 63.8 ± 5.6 minutes after intravenous injection of 149-386 mega Becquerel of $^{18}$F-FDG, depending on the body mass index $^{22}$. PET-CT was performed using a dedicated head and neck protocol (scan trajectory jugular notch-orbit; arms down), using an integrated PET-CT system (Gemini TF-64CT; Philips Healthcare, Cleveland, OH, USA; 3D-mode; 4 min emission scans/bed position). Low-dose CT
scanning was performed with 120 kV and 50 mAs prior to emission scanning for attenuation correction and anatomical localisation of $^{18}$F-FDG avid lesions. PET-CT data were reconstructed using a time of flight row-action maximum likelihood algorithm, as implemented by the vendor. Final image matrix size equals 288x288 with a voxel size of 2x2x2 mm. Post reconstruction image resolution equalled 5 mm FWHM.

The primary tumor VOIs for quantitative parameters measurements were drawn using a 3-dimensional (3D) region-growing algorithm, implemented with software developed in-house at the VU University Medical Center, by C. Schouten supervised by a nuclear medicine physician (O. Hoekstra). For each VOI, maximum SUV (SUV$_{\text{max}}$), peak SUV (SUV$_{\text{peak}}$), average SUV using an adaptive threshold of 50% (SUV$_{\text{mean}}$), MATV and TLG (calculated as product of SUV$_{\text{mean}}$ and MATV) were obtained. SUVs were normalized for lean body mass and serum glucose. Cumulative SUV-volume histograms (CSH) were investigated, representing % of total volume with an SUV above a threshold from 0 to 100% of SUV$_{\text{max}}$. The area under the curve (AUC) is a quantitative index of uptake heterogeneity, with lower values corresponding to higher degrees of tumor heterogeneity.

**Tumor volume**

Primary tumor volume was obtained from CT scans (Discovery CT590RT, General Electric, Milwaukee, Wisconsin, USA) (with 95 mL intravenous contrast injected at rate of 1 mL/s), used for radiotherapy treatment planning. Time between the staging $^{18}$F-FDG-PET-CT and the radiotherapy planning CT was on average 1-2 weeks. Patients underwent CT scanning in the supine position and were positioned in a 5 point fixation head mask (Posicast® Thermoplastics, Civco Medical Solutions, Reeuwijk, the Netherlands) and scanned in the RT position. A scan was made from the top of the skull to the aortic arch with a slice thickness of 2.5 mm. The gross tumor volume (GTV) using the CT images was delineated, slice by slice, by head and neck radiation oncologists from our hospital. This CT scan was co-registered with a diagnostic MRI scan to ensure optimal delineation. All delineated contours were added and a resulting volume was calculated (Eclipse treatment planning system, Varian Medical Systems, Palo Alto, CA, USA).

In three patients, the time interval between the staging $^{18}$F-FDG-PET-CT and radiotherapy planning CT was delayed due to the administration of induction chemotherapy (n=2) or due to a complication during percutaneous endoscopic gastrostomy (PEG) tube placement (n=1). One patient died before
planning CT was made. In these patients, tumor volume was obtained from axial STIR MRI-images with 4 mm sections. Contours were manually drawn by S. Hakim, under supervision of a head-and-neck radiologist (P. de Graaf), around the border of the primary tumor at each slice position to measure total tumor volume.

**Statistical analysis**

Statistical analyses were performed using SPSS software package (version 20.0; IBM Corp., Armonk, NY, USA). The level of significance was set at $p<0.05$ and hypotheses were tested two-sided. Analysis for differences in patient characteristics between the HPV-positive and HPV-negative group were performed with the Pearson Chi-square ($\chi^2$) test for categorical data. Bonferroni correction was used to compare subgroups. The Student’s $t$ test was used to compare continuous data in case of a normal distribution and the Mann-Whitney $U$ test was used in case of a non-normal distribution. We tested whether there was a significant association between PET-CT parameters and HPV-status in a multivariate analysis to look at the association, after adjusting for other explanatory variables.

**RESULTS**

**Patient characteristics**

The total study group consisted of 39 men and 5 women, with a mean age at the time of diagnosis of 61.0 years (range 46-78). Patient characteristics by HPV-status are shown in Table 1. HPV-positive patients were more likely to present with a T2 primary tumor and N-positive disease. HPV-negative patients were more likely to have a history of heavy smoking and excessive alcohol consumption.
Table 1. General patient characteristics

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>HPV-positive</th>
<th>HPV-negative</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n=27 (61.4%)</td>
<td>n=17 (38.6%)</td>
<td></td>
</tr>
<tr>
<td>Gender</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>24 (88.9)</td>
<td>15 (88.2)</td>
<td>p=0.95</td>
</tr>
<tr>
<td>Female</td>
<td>3 (11.1)</td>
<td>2 (11.8)</td>
<td></td>
</tr>
<tr>
<td>Age at diagnosis, years</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean ± SD</td>
<td>59.7 ± 7.5</td>
<td>63.0 ± 6.4</td>
<td>p=0.14</td>
</tr>
<tr>
<td>Oropharyngeal subsite</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tonsil</td>
<td>12 (44.4)</td>
<td>6 (35.3)</td>
<td>p=0.54</td>
</tr>
<tr>
<td>Base of tongue</td>
<td>12 (44.4)</td>
<td>7 (41.2)</td>
<td></td>
</tr>
<tr>
<td>Oropharynx nos</td>
<td>3 (11.1)</td>
<td>4 (23.5)</td>
<td></td>
</tr>
<tr>
<td>Smoking</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Never (0-5 pack years)</td>
<td>12 (44.4)</td>
<td>0 (0)</td>
<td>p=0.002</td>
</tr>
<tr>
<td>Moderate (6-24 pack years)</td>
<td>6 (22.2)</td>
<td>3 (17.6)</td>
<td></td>
</tr>
<tr>
<td>Heavy (&gt;24 pack years)</td>
<td>9 (33.3)</td>
<td>14 (82.4)</td>
<td></td>
</tr>
<tr>
<td>Alcohol consumption</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Never (0)</td>
<td>4 (14.8)</td>
<td>0 (0)</td>
<td>p=0.005</td>
</tr>
<tr>
<td>Moderate (1-149 unit years)</td>
<td>20 (74.1)</td>
<td>8 (47.1)</td>
<td></td>
</tr>
<tr>
<td>Heavy (&gt;149 unit years)</td>
<td>3 (11.1)</td>
<td>9 (52.9)</td>
<td></td>
</tr>
<tr>
<td>T-classification</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>T2</td>
<td>18 (66.7)</td>
<td>1 (5.9)</td>
<td>p&lt;0.001</td>
</tr>
<tr>
<td>T3-4</td>
<td>9 (33.3)</td>
<td>16 (94.1)</td>
<td></td>
</tr>
<tr>
<td>N-classification</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>N0</td>
<td>0 (0)</td>
<td>3 (17.6)</td>
<td>p=0.04</td>
</tr>
<tr>
<td>N1-3</td>
<td>27 (100)</td>
<td>14 (82.4)</td>
<td></td>
</tr>
</tbody>
</table>

Smoking was defined in pack years (1 pack year=20 cigarettes a day during 1 year)
Alcohol consumption was defined in unit years (1 unit year=one alcohol-containing consumption a day during 1 year).
Abbreviations: HPV; human papillomavirus, oropharynx nos; oropharynx not otherwise specified.

Association between HPV-status and imaging parameters

Representative $^{18}$F-FDG-PET-CT images from an HPV-negative and an HPV-positive patient are shown in Figures 1 and 2. Table 2 summarizes $^{18}$F-FDG-PET-CT imaging parameters in relation to HPV-status. HPV-positive patients had significantly smaller tumor volumes (median 9.6 cm$^3$ [interquartile range 6.4-22.2]) than HPV-negative patients (median 30.6 cm$^3$ [22.2-85.5]) (p<0.001). The median SUV$_{max}$ measurements of the primary tumor site for the HPV-positive and HPV-negative groups were 8.0 [5.9-11.8] and 13.6 [9.8-17.2], respectively (p=0.002) (Figure 3). Median MATV was 2.8 [1.6-5.3] ml for HPV-positive and 6.1 [4.5-21.2] ml for HPV-negative tumors (p<0.001). The median
total lesion glycolysis for HPV-positive and HPV-negative was 17.8 [7.1-40.1] and 55.8 [42.3-211.7], respectively (p<0.001). Comparing AUC-CSH, a marker for tumor heterogeneity, HPV-positive primary tumors are significantly less heterogeneous than HPV-negative tumors, 0.91 [0.84-0.96] versus 0.82 [0.78-0.86] (p=0.003).

Some \(^{18}\text{F}-\text{FDG-PET-CT}\) parameters are volume dependent because of partial volume effects (PVE). Therefore, a more objective assessment of the glucose metabolism parameters is obtained after including MATV in the statistical analysis. MATV was found to be significantly associated with SUV\(_{\text{max}}\) (spearman’s rho= 0.64; p<0.001), SUV\(_{\text{mean}}\) (spearman’s rho= 0.63; p<0.001) and SUV\(_{\text{peak}}\) (spearman’s rho= 0.68; p<0.001). In univariate analysis, the SUV\(_{\text{max}}\) in HPV-positive was 5.1 units lower than in HPV-negative ones. For SUV\(_{\text{peak}}\), a mean difference of 4.1 was found. In multivariate analysis, including MATV as covariate, the SUV\(_{\text{max}}\) in HPV-positive tumors was 3.9 units (p=0.01) lower than in HPV-negative ones. The mean difference in SUV\(_{\text{peak}}\) in multivariate analysis was 3.3 (p=0.008).

**Table 2.** Imaging parameters in relation to HPV-status

<table>
<thead>
<tr>
<th>(^{18}\text{F}-\text{FDG-PET-CT})</th>
<th>HPV-positive (n=27) (61.4%)</th>
<th>HPV-negative (n=17) (38.6%)</th>
<th>(p)-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tumor volume* ((\text{cm}^3))</td>
<td>9.6 [6.4-22.2]</td>
<td>30.6 [22.2-85.5]</td>
<td>(&lt;0.001)</td>
</tr>
<tr>
<td>SUV(_{\text{max}})</td>
<td>8.0 [5.9-11.8]</td>
<td>13.6 [9.8-17.2]</td>
<td>(=0.002)</td>
</tr>
<tr>
<td>SUV(_{\text{mean}})</td>
<td>5.9 [4.5-8.5]</td>
<td>9.9 [6.9-11.9]</td>
<td>(=0.003)</td>
</tr>
<tr>
<td>SUV(_{\text{peak}})</td>
<td>7.1 [5.6-9.6]</td>
<td>11.7 [9.1-14.3]</td>
<td>(=0.001)</td>
</tr>
<tr>
<td>MATV (ml)</td>
<td>2.8 [1.6-5.3]</td>
<td>6.1 [4.5-21.2]</td>
<td>(=0.001)</td>
</tr>
<tr>
<td>TLG</td>
<td>17.8 [7.1-40.1]</td>
<td>55.8 [42.3-211.7]</td>
<td>(=0.001)</td>
</tr>
<tr>
<td>AUC-CSH (%)</td>
<td>0.91 [0.84-0.96]</td>
<td>0.82 [0.78-0.86]</td>
<td>(=0.003)</td>
</tr>
</tbody>
</table>

* Measured with contrast-enhanced CT

Values are presented as median [interquartile range].

Abbreviations: AUC-CSH: area under the curve- cumulative SUV-volume histograms, HPV; human papillomavirus, MATV; metabolic active tumor volume SUV; standardized uptake value, TLG; total lesion glycolysis; \(^{18}\text{F}-\text{FDG-PET-CT}\); \(^{18}\text{F}\)-fluorodeoxyglucose positron emission tomography-computed tomography.
Figure 1.
A 54-year old patient was diagnosed with an HPV-negative OPSCC in the right palatine tonsil. Axial and coronal $^{18}$F-FDG-PET-CT images before treatment showing increased $^{18}$F-FDG uptake in the primary tumor. (A) Axial CT, (B) axial PET, (C) axial fused, (D) coronal CT, (E) coronal PET and (F) coronal fused. A VOI was drawn for the primary lesion; $\text{SUV}_{\text{mean}}$ 12.5, $\text{SUV}_{\text{max}}$ 18.6, $\text{SUV}_{\text{peak}}$ 13.6, MATV 42.2 and TLG 528.3.
Figure 2.
A 48-year old patient was diagnosed with an HPV-positive OPSCC in the right palatine tonsil. Axial and coronal $^{18}$F-FDG-PET-CT images before treatment showing increased $^{18}$F-FDG uptake in the primary tumor. (A) Axial CT, (B) axial PET, (C) axial fused, (D) coronal CT, (E) coronal PET and (F) coronal fused. A VOI was drawn for the primary lesion; SUV$_{\text{mean}}$ 7.6, SUV$_{\text{max}}$ 10.3, SUV$_{\text{peak}}$ 9.6, MATV 2.8 and TLG 21.6.
Figure 3.
Association between SUV$_{\text{peak}}$ (A) and SUV$_{\text{max}}$ (B) with HPV-status in patients with OPSCC. SUV$_{\text{peak}}$ in HPV-positive OPSCC is 7.1 [5.6-9.6] and 11.7 [9.1-14.3] in HPV-negative OPSCC (p=0.001). SUV$_{\text{max}}$ in HPV-positive OPSCC is 8.0 [5.9-11.8] and 13.6 [9.8-17.2] in HPV-negative OPSCC (p=0.002).
HPV-related squamous cell carcinoma of the oropharynx is a clinically and histopathologically distinct disease entity. Patients with HPV-positive OPSCC show higher response rates to chemoradiotherapy and have a better overall survival compared to HPV-negative OPSCC. Traditional prognostic factors such as tumor size and lymph node invasion are inadequate to classify patients into risk groups. Therefore, the identification of other independent tumor characteristics that may select patients for tailored treatment regimens becomes of interest. Our results showed significantly lower glycolytic parameters and decreased tumor heterogeneity as measured with \(^{18}\text{F-FDG-PET-CT}\) in HPV-positive OPSCC as compared to HPV-negative ones.

A better overall survival after CRT is seen in patients with an HPV-positive OPSCC compared to HPV-negative patients, mainly due to its increased radiosensitivity. Besides, patients with a complete response to CRT exhibited relatively low pretreatment SUV-values than patients with a partial or non-response to non-surgical treatment. Distinct histological features in HPV-positive and HPV-negative OPSCC may be responsible for the different pretreatment SUV-values, potentially due to a different distribution of hypoxic regions within the tumor as a result of other metabolic demands. \(^{18}\text{F-FDG-PET-CT}\) reflects metabolic activity which is indirectly related to tumor oxygenation as hypoxia increases glucose metabolism. Considering the radiosensitivity of HPV-related OPSCC, hypothetically the amount of hypoxia would be low and HPV-positive patients should have relatively low pretreatment SUV-values compared to HPV-negative patients. Indeed, in this study we have found that \(^{18}\text{F-FDG-PET-CT}\) parameters are significantly lower in patients with HPV-positive OPSCC compared with HPV-negative patients. This hypothesis can be best confirmed by a PET-CT study with a specific hypoxia tracer, such as \(^{18}\text{F-fluoroazomycin arabinoside (^{18}\text{F-FAZA})}\).

Heterogeneous histopathology or morphology may be more accurately reflected by a quantitative index of uptake heterogeneity, such as AUC-CSH, than other metabolic PET-CT parameters. Probably, factors such as necrosis and hypoxia within a tumor contribute to tumor heterogeneity in \(^{18}\text{F-FDG-uptake}\). As HPV-positive and HPV-negative OPSCC are morphologically distinct diseases, it is of interest to quantify tumor heterogeneity in \(^{18}\text{F-FDG uptake}\). We found that primary tumors of HPV-positive patients are more homogeneous than tumors of HPV-negative patients, using AUC-CSH as a heterogeneity index. A previous study by Tahari et al. showed that HPV-negative primaries are more heterogeneous by comparison of the ratio \(\text{SUV}_{\text{max}}\) by \(\text{SUV}_{\text{mean}}\). These results suggest that HPV-negative
OPSCC contain more areas with necrosis and hypoxia, contributing to the reduced radiosensitivity of HPV-negative tumors.

Joo et al. found a significant difference between primary tumor $\text{SUV}_{\text{max}}$ and HPV-status in 78 patients with OPSCC and concluded that $^{18}$F-FDG-PET-CT could be used as an adjuvant diagnostic tool for determination of HPV-status. Tahari et al. also showed that HPV-negative primary tumors have significantly higher metabolic rates, such as $\text{SUV}_{\text{max}}$, $\text{SUV}_{\text{peak}}$ and $\text{SUV}_{\text{mean}}$, as compared with HPV-positive patients. In nodal disease, statistical significance was not found for the same parameters. Our results are in accordance with results of these previous studies. However, two methodological comments can be placed. First, accurate quantification of $^{18}$F-FDG-uptake is size dependent, because of the partial volume effect (PVE). This process is affected by tumor size and shape, spatial resolution of the scanner, background activity in surrounding tissue, image sampling and voxel size. The PVE effect is most prominent in small tumors and results in an underestimation of the accumulation of radioisotope in tumor. HPV-positive tumors typically present with a small primary tumor at diagnosis. Previous mentioned studies did not take the PVE into account. Statistical analysis in our study showed a significant association of $\text{MATV}$ with $\text{SUV}_{\text{values}}$. We therefore tested the association between HPV-status and PET-CT imaging parameters using a multivariate analysis as well, including $\text{MATV}$ as covariate. In multivariate analysis, $\text{SUV}_{\text{max}}$ and $\text{SUV}_{\text{peak}}$ remained significantly associated with HPV-status. Second, in our institute we use a validated algorithm for HPV-detection: p16 immunostaining, followed by a GP 5+/6 PCR on the p16-immunoposititive cases. This algorithm showed an accuracy of 98%. Joo et al. and Tahari et al. assessed HPV status by high risk in situ hybridization. This technique shows a sensitivity and specificity of 88%. HPV analysis by in situ hybridization could have affected their results, with both an underrepresentation of HPV-positive cases and more false positive patients.

$^{18}$F-FDG-PET-CT is a diagnostic modality used in head and neck oncology for pretreatment staging, treatment planning and assessment of treatment response and has additional prognostic value. $^{18}$F-FDG is often increased in malignant tissues, when compared to healthy tissue, caused by an higher glucose metabolism coinciding with an up-regulated glucose uptake through overexpression of glucose transporters (glut). Increased $\text{SUV}_{\text{values}}$ of the primary tumor, using $\text{SUV}_{\text{values}}$, is a poor prognostic factor in HNSCC treated with definitive radiotherapy or concomitant with chemotherapy. Also, several studies have reported on $\text{MATV}$ and $\text{TLG}$ as prognostic imaging markers. Recent studies show conflicting results whether $^{18}$F-FDG-PET-CT parameters and HPV-status are independently associated with survival outcomes. Cheng et al. studied 60 patients with OPSCC and multivariate
analysis revealed that TLG and HPV-positivity were independently associated with overall survival\(^1\). However, only 12 patients (20%) were HPV-positive. Garsa et al. showed that MATV is a significant predictor of disease free survival and overall survival in subgroups of patients (p16-positive (n=25) and p16-negative (n=18))\(^1\). Although p16 immunohistochemistry is a surrogate marker for an HPV-infection, a considerable number of patients are p16-positive but HPV-negative\(^2\). On the contrary, in a multivariate analysis with 78 patients from Joo et al., only HPV-positivity and not SUV\(_{\text{max}}\) was associated with 5-year disease-specific survival\(^1\). Further long-term follow-up studies with sufficient OPSCC patients who undergo \(^{18}\)F-FDG-PET-CT and HPV analysis can answer the question if \(^{18}\)F-FDG-PET-CT imaging parameters and HPV-status are independent prognostic factors. However, this is unlikely if there is a strong association between these tumor characteristics. This study shows that imaging parameters derived from \(^{18}\)F-FDG-PET-CT are significantly associated with HPV-status in patients with OPSCC.

We acknowledge several limitations to this study. First, due to the retrospective study design that only included patients with a dedicated head and neck \(^{18}\)F-FDG-PET-CT, selection bias may have occurred. Secondly, a limited number of patients with OPSCC was included. Despite these limitations, we found a significant association between HPV-status and \(^{18}\)F-FDG-PET-CT parameters after adjusting for MATV and we believe our study provides additional information to previously reported studies on this subject.

**CONCLUSION**

Glycolytic parameters from \(^{18}\)F-FDG-PET-CT are significantly lower in HPV-positive as compared with HPV-negative patients. Besides, HPV-positive OPSCC show lower degrees of tumor heterogeneity. In multivariate analysis, including MATV as a covariate, SUV\(_{\text{max}}\) and SUV\(_{\text{peak}}\) remained significantly associated with HPV-status. Low pretreatment \(^{18}\)F-FDG-PET-CT parameters, which are associated with a more favourable prognosis, may be explained by HPV-induced tumor changes.

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REFERENCES


