Neck ultrasound as a diagnostic method of cervical lymph node metastasis in patients with Head and Neck squamous cell carcinoma

Author: Alexandra Timmerman
Bachelor of Medicine
Supervisor: Mathias von Beckerath, MD, PhD, University Hospital Örebro
Department of Otolaryngology
Örebro, Sweden
Abstract

**Introduction** Head and Neck squamous cell carcinoma (HNSCC) has a tendency to spread to cervical lymph nodes (LN) but to where and to what extent varies from case to case. Since cervical LN metastasis is such an important prognostic factor, regardless of known LN metastasis, a neck dissection (ND) is often performed where cervical LN are removed to an extent that also varies from case to case and are based on clinical and imaging findings. The preoperative knowledge of the nodal status is therefore of great importance. Computed tomography (CT) constitutes the primary investigation method for these patients in Sweden today. Ultrasound (US) is not a part of the routine investigation but is used sometimes to get a better view of the tumour or a suspicious LN.

**Objective** The aim of the study is to carry out a pilot study investigating the efficiency of neck US compared to CT in finding LN metastasis in patients with HNSCC when performed by an inexperienced clinician.

**Method** This is a prospective pilot study enrolling five patients with HNSCC who were scheduled to undergo surgical treatment including a ND at the ENT clinic at the University Hospital Örebro. An US was performed the day before surgery by an inexperienced clinician. The results were compared to both the results from the routine CT and the histopathological examination, the latter used as gold standard.

**Results** From the five patients enrolled in the study 111 LN were retrieved and histopathological examination confirmed metastasis in two LN from two of the five patients. The two metastasis were also detected using US but not by the CT, giving a sensitivity of 100% and 0% respectively and a specificity of 96.3% and 95.4% respectively.

**Conclusion** The results show that an US in these patients add information about the nodal status even when performed by an inexperienced performer and after a CT has been made.

Key words: Squamous cell carcinoma, head and neck, ultrasound, lymph node metastasis
Abbreviations

CT – Computed tomography
FNAC – Fine-needle aspiration cytology
HNSCC – Head and Neck squamous cell carcinoma
LN – Lymph node
MDT – Multidisciplinary tumour board
MRI – Magnetic resonance imaging
ND – Neck dissection
OPSCC – Oropharyngeal squamous cell carcinoma
OSCC – Oral squamous cell carcinoma
PET – Positron emission tomography
SCC – Squamous cell carcinoma
SNB – Sentinel node biopsy
US – Ultrasound
Table of contents

Abstract ................................................................................................................................. 2
Abbreviations ....................................................................................................................... 3
1. Introduction .................................................................................................................... 5
   1.1 Epidemiology ...................................................................................................... 5
   1.2 Lymphatic drainage ......................................................................................... 5
   1.3 Routine investigation today ............................................................................. 6
   1.4 Ultrasound/Previous research ...................................................................... 6
2. Objective ...................................................................................................................... 7
   2.1 Aim .................................................................................................................... 7
   2.2 Hypothesis ........................................................................................................ 7
3. Method and materials ............................................................................................... 7
   3.1 Study type and Subjects ................................................................................ 7
   3.2 Ultrasound ........................................................................................................ 8
   3.3 Surgical procedure and neck dissection ..................................................... 8
   3.4 Histopathological examination ................................................................ 8
   3.5 Statistics ........................................................................................................... 9
   3.6 Ethics .................................................................................................................. 9
4. Results ......................................................................................................................... 9
5. Discussion .................................................................................................................... 11
6. Conclusion .................................................................................................................. 13
7. Acknowledgements ................................................................................................... 15
8. References .................................................................................................................. 15
Appendix A ....................................................................................................................... 17
1. Introduction

1.1 Epidemiology
Every year there are over 500,000 new cases of Head and Neck squamous cell carcinoma (HNSCC) worldwide. It has been reported to be the sixth most common type of malignancy worldwide [1]. In Sweden, Head and Neck cancer is a less common form of cancer, representing 2.3% of all cancer. Head and Neck cancer is by 95% composed of squamous cell carcinoma, whereas the remaining cases mainly are tumours in the salivary glands. According to the 2014 annual report from the Swedish Head and Neck cancer register (SweHNCR) there were approximately 1,400 new cases of Head and Neck cancer in Sweden. Head and Neck cancer is a heterogeneous group with different origins such as lip, oral cavity, oropharynx, hypopharynx, nasopharynx or larynx. Incidence, aetiology, and prognosis vary among these categories. The two most common groups are oral (OSCC) and oropharyngeal squamous cell carcinoma (OPSCC) with approximately 380 and 350 new cases per year in Sweden respectively [2].

1.2 Lymphatic drainage
HNSCC, like many other types of tumours, tend to spread to different locations than its original site. It primarily does so through the lymphatic system to cervical LNs. In addition to distant lymph node (LN) metastases, the status of cervical LNs is a very important prognostic factor in patients with HNSCC. Clinically palpable cervical LN metastases have the ability to reduce the five-year survival rate with approximately 50% [3]. The incidence of occult LN metastases has been reported to be 20% or greater in HNSCC and, even higher, 34% in OSCC [4,5]. In a study from 1997, Woolgar et al investigated neck-specimens from 154 patients with OPSCC with the aim to map regional LN metastasis. The study showed that metastasis was present in 47 patients where the metastatic findings were bilateral in 8% of the 47 cases [6].

To what extent or localisation the spreading occurs varies from case to case, even in a group with the same type of primary tumour. This is shown in a study from earlier this year carried out by Christensen et al (see table of different spreading locations in Appendix A) [7]. Different sites of the primary tumour imply different risks of
regional spreading. Tumours in oral tongue and floor of mouth tend to spread to regional LNs more often than other primary tumours in the head and neck [8].

1.3 Routine investigation today

The routine investigation in Sweden for detecting cervical LN metastasis is by CT. All patients with HNSCC are discussed at a multidisciplinary tumour board (MDT) where physicians with different disciplines and expertise come together to discuss the patient and imaging findings to determine future surgical and oncological treatment. In some cases, the patient is further investigated by performing a Positron Emission Tomography-Computed Tomography (PET/CT) or Magnetic Resonance Imaging (MRI). US is not a part of the routine investigation today. However, the operating surgeons sometimes use it to get a better understanding of both the primary tumour and suspected pathological LNs prior to the surgery and as an aid for fine-needle aspiration cytology (FNAC). Although there have been many advances in the different techniques of investigating cervical LN metastasis it has been documented that none of them reach the same level of success as the histopathological examination. In a study Rodrigo et al note that none of CT, PET-CT, MRI, US or US-guided fine-needle aspiration (FNA) reached a sensitivity of more than 80-85% [9]. The reason for the low sensitivity could be that these cancer forms often involve micrometastasis, which the histopathological examination is more accurate in detecting [10].

1.4 Ultrasound/Previous research

US is sometimes used preoperatively in pursuit of better understanding of the characteristics and extension of the primary tumour as well as potential LN involvement. However, it is not a part of the routine investigation of patients with HNSCC in Sweden. Several studies have been carried out to compare the efficiency of US with other radiographic methods of investigation in detecting potential LN metastasis in patients with HNSCC. In one study from 2014 Hong et al aimed to investigate the usefulness of US in detecting cervical LN metastasis in 52 patients where the results from the US was compared to those from MRI and PET/CT. The study showed that the US had the highest agreement with the histology results with a sensitivity of 78.9% and specificity of 99% [11]. In another study from 2014, Norling et al carried out a prospective study that included 51 patients with OSCC which had
been classified as N0 by MRI/CT where a preoperative US was performed and compared to the histopathological findings. The study showed that US had a sensitivity of 43.8% and a specificity of 91.4% [12]. In a study from 2000, Stuckensen et al. aimed to compare the use of US, CT, MRI and PET to identify cervical LN metastasis in 52 patients with OSCC. The study showed that US had the highest sensitivity (84%) and PET had the highest specificity (82%) [13].

2. Objective

2.1 Aim

The aim of the study is to carry out a pilot study investigating the efficiency of neck US compared to CT in finding LN metastasis in patients with HNSCC when performed by an inexperienced clinician.

2.2 Hypothesis

The hypothesis for this study is that US has a higher sensitivity and specificity than CT in finding LN metastases in patients with HNSCC.

3. Method and materials

3.1 Study type and Subjects

This study is a prospective pilot study carried out at the University Hospital Örebro between August 2016 and January 2017. The subjects included were patients with HNSCC who were scheduled to undergo surgical treatment including ND at the ENT clinic at the University Hospital in Örebro. All patients underwent CT 2-6 weeks prior to the surgery and US the day before the surgical intervention. The inclusion criteria used were: the patients had HNSCC, they were to undergo surgical treatment including a ND at the ENT clinic at the University Hospital Örebro and they had undergone a preoperative CT. Five patients were enrolled in the study. One patient was excluded from the study because of a large secreting skin metastasis on one side of the neck and a previously performed ND on the other side of the neck. One patient declined participating in the study.
3.2 Ultrasound

The US machine used in the study was BK Medical flex focus 500 with the frequency set to 18 MHz. The examination was performed the day before the patient’s head and neck surgery. It was performed by one medical student with no prior practical experience of neck US other than two hours of demonstration of the US machine. The performer was blinded to all previous investigation and documentation regarding the patient. If a LN was found its characteristics were evaluated. The characteristics that were evaluated are known predictors of nodal metastasis and included: size (short axis >10mm), form (round), echogenicity (e.g. calcifications, central necrosis), hilum (absence of) and vascular patterns (peripheral blood flow) [14,15]. After the evaluation, the LNs were given one of three grades of suspicion for malignancy: benign, suspicious, or malignant. If a LN was found that had at least one of the characteristics mentioned above it was documented in still pictures and marked with what level of the neck it was found in (Figure 1.).

3.3 Surgical procedure and neck dissection

In conjunction with the surgical cancer treatment all patients underwent a ND where cervical LNs were removed. The extent of the ND was determined by the MDT and/or the operating surgeon based on clinical examination and imaging findings. During the ND the levels of the neck were removed and put in a container with formalin that was carefully marked with what level it was containing before transported to the Department of Pathology for histopathological examination.

3.4 Histopathological examination

The samples from the ND were received, divided level by level and fixed in formalin. Large LNs/metastases and other suspicious findings are sliced in 3 mm slices before further preparations. All LNs found and other tissue that was to be further investigated was put into a paraffin block to undergo a dehydration process. Thereafter the tissue
was sliced into 4 \( \mu m \) slices, almost equivalent to one layer of cells. From every paraffin block one slice was chosen to be stained using Hematoxyline-eosin (H&E). The last step was the examination performed by a pathologist.

### 3.5 Statistics

The findings from the US were compared to the findings from both the CT and the histopathological examination. The latter was considered to be the gold standard. The sensitivity and specificity was calculated using true and false positive and negative values.

### 3.6 Ethics

All study subjects were given both oral and written information and an informed consent was obtained. All documentation was made under coding. None of the findings were reported to the operating surgeons and the risk of altering the surgical procedure was diminished. Given that this is a pilot study and a student’s project no application for ethical approval from the Ethical Review Board was made.

### 4. Results

Five patients (three men, two women, age-range 39-79 years) were enrolled in the study. Primary tumours were detected in floor of mouth, larynx, tongue and nasal columella. A total of 111 LNs were retrieved and histopathological examination found metastases in two LNs from two of the five patients. As seen in Table 1 all patients but one were regularly using tobacco.

**Table 1. Introductory study results**

<table>
<thead>
<tr>
<th>Patient</th>
<th>Tumour origin</th>
<th>Tobacco use</th>
<th>Number of LNs found</th>
<th>HE: # of malignant LNs found</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Floor of mouth</td>
<td>Cigarettes</td>
<td>35</td>
<td>1</td>
</tr>
<tr>
<td>2</td>
<td>Larynx</td>
<td>Snus</td>
<td>23</td>
<td>0</td>
</tr>
<tr>
<td>3</td>
<td>Tongue</td>
<td>-</td>
<td>27</td>
<td>0</td>
</tr>
<tr>
<td>4</td>
<td>Tongue</td>
<td>Snus</td>
<td>4</td>
<td>1</td>
</tr>
<tr>
<td>5</td>
<td>Columella</td>
<td>Cig., Snus</td>
<td>22</td>
<td>0</td>
</tr>
<tr>
<td>Sum for all patients</td>
<td></td>
<td></td>
<td>111</td>
<td>2</td>
</tr>
</tbody>
</table>

LN=Lymph node, HE=Histopathological examination
Table 2 shows number of suspicious or malignant LNs found during CT, US and the histopathological examination as well as in what regions the LNs were found in. As seen in Table 2 US found two out of two metastatic LNs while CT missed both.

Table 2. Locations of suspicious/malignant LNs identified via CT, US and histopathological examination

<table>
<thead>
<tr>
<th>Patient</th>
<th>CT</th>
<th>US</th>
<th>HE (gold standard)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>–</td>
<td>Right IA (JS)*; Left II (JS)</td>
<td>Left II (CM)**</td>
</tr>
<tr>
<td>2</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>3</td>
<td>Left II (JS); Left II (JS); Left II (JS)</td>
<td>Left II (JS)</td>
<td>–</td>
</tr>
<tr>
<td>4</td>
<td>–</td>
<td>Right III (JM)</td>
<td>Right III (CM)</td>
</tr>
<tr>
<td>5</td>
<td>Right II (JS); Left II (JS)</td>
<td>Right II (JS); Left II (JS); Left III (JS)</td>
<td>–</td>
</tr>
</tbody>
</table>

JM=judged malignant, JS=judged suspicious, CM=confirmed malignant, CT=Computed tomography, US=Ultrasound, HE=Histopathological examination; *Not included in results calculations as neck level was not included in specimen sent for histopathological examination; **After comparing sizes and re-evaluating the CT results it was concluded that this LN was the same as the one found in level II even though it was originally marked as level III.

In Table 3 the sensitivity and specificity for US and CT are presented. The calculations were made using the true and false positive and negative results. Also presented in Table 3 the table shows a 100% sensitivity for US and 0% for CT. The specificity for US and CT is 96.3% and 95.4% respectively.

Table 3. Results from US and CT compared to histopathological examination

<table>
<thead>
<tr>
<th>Method</th>
<th>TP</th>
<th>TN</th>
<th>FP</th>
<th>FN</th>
<th>Sensitivity = TP/(TP+FN)</th>
<th>Specificity = TN/(TN+FP)</th>
</tr>
</thead>
<tbody>
<tr>
<td>US</td>
<td>2</td>
<td>105</td>
<td>4</td>
<td>0</td>
<td>1.000</td>
<td>0.963</td>
</tr>
<tr>
<td>CT</td>
<td>0</td>
<td>104</td>
<td>5</td>
<td>2</td>
<td>0.000</td>
<td>0.954</td>
</tr>
</tbody>
</table>

TP=true positive (malignant LNs correctly identified); TN=true negative (benign LNs correctly identified); FP=false positive (benign LNs incorrectly identified as suspicious/malignant); FN=false negative (malignant LNs incorrectly identified as benign)

In Figure 2 Picture taken during neck US. 1. LN with benign characteristics; 2. M. Sternocleidomastoideus; 3, 4. Internal and external carotid artery; 5. Internal Jugular vein

In Figure 3 Suspicious LN. Round shape, hyperechoic structures in node indicates necrosis.
5. Discussion

The results of this study shows that out of 111 LNs retrieved from 5 patients, 2 LNs were confirmed metastatic by the histopathological examination. US found both of the metastatic LNs while CT missed them both. This resulted in a sensitivity of 100% for ultrasound and 0% for CT and a specificity of 96.3% and 95.4% respectively.

Cervical LN metastasis in patients with HNSCC is a well-established negative prognostic factor. Presence of one or more metastatic LN can reduce the survival rate by about 50% [15,16]. Even though it will in many cases be unnecessary, many institutions treat the neck even when clinically N0 because of the high probability of occult metastasis [17]. Accurate preoperative knowledge of cervical LN status is therefore critical. US is a widely available, rapid, low-cost and none-invasive method of assessing the cervical LN status. It is also possible to perform the US as late as the day before the surgery as results are obtained immediately, an advantage compared to Sentinel Node Biopsy (SNB) and FNAC. In the following, the results for US are compared with those for CT. Then this study’s results for US are compared to those in previous studies. The discussion also highlights the danger of false negative results and presents the relevance of this study for clinical practise. Last, limitations are discussed.

First, the results for US are compared to those for CT. In this study, US found both of the two metastatic LNs resulting in a sensitivity of 100%. CT, on the other hand, was unable to identify any metastatic LNs resulting in a sensitivity of 0%. Previous studies comparing US and CT, such as Stuckensen et al and Gheeta et al, also show a lower sensitivity for CT (66% and 50% respectively) than for US (84% and 100% respectively) [13,18]. Possible reasons for US having a higher sensitivity than CT are that US is able to show more characteristics of the LNs while CT primarily evaluates size. CT is also limited when it comes to detecting and evaluating smaller nodes [19]. A third reason is that the CT was made several weeks before the removal of the LNs and that the cancer had been given time to produce structural changes in the nodes. Even though there are reasons for the sensitivity of US being higher than for CT, they cannot explain the big difference in sensitivity in this study. The conclusion must be that 0% for CT is misleading and probably an effect of the low number of patients. This study shows that the specificity for US and CT is similar to each other and
relatively high at 96.3% and 95.4% respectively. This means that when suspecting metastasis both US and CT is likely to be right.

Second, the US results obtained in this study are compared to those of previous studies. Other studies have obtained US sensitivity results of 78.9% (Hong et al), 84% (Stuckensen et al) and 100% (Gheeta et al) [11,13,18]. The previous study with the most similar results to this study is Gheeta et al. The similarity may be due to the low number of patients enrolled in the study, as larger numbers of patients increase the risk of missing something and therefore getting a lower sensitivity. The previous study with the most different results to this study is Norling et al, which found that the sensitivity for US was only 43.8% [12]. The reason for the difference may be that they only enrolled patients with necks previously classified as N0 and therefore included patients with less obvious tumour spreading.

When comparing this study’s specificity for US with that of other studies the results differ a lot between different studies. Both Hong et al and Norling et al have similar results to this study with a specificity for US at 99% and 91.4% respectively [11,12]. However, Stuckensen et al and Gheeta et al found the specificity to be lower at 68% and 25% respectively [13,18]. They also found, unlike this study’s results, the specificity for CT to be higher than for US at 74% and 100% respectively. The reason for this might be the large number of nodes US was able to find in combination of a lacking ability to differentiate between benign and malignant appearance, which leads to a high false positive result, and consequently low specificity [13,18]. When comparing results with previous studies one has to take in consideration that they are a couple of years old which might affect the results since the US machine has developed during recent years and might give better pictures today.

Third, avoiding false negative values is important for all diagnostic methods. False negative means that the method could not find what was later confirmed to be malignant using the gold standard method. US was in this small population able to produce a false negative value of zero. However, the false negative value of two for CT is a remarkable finding given that it is the primary method of searching for cervical LN metastasis in these patients. One challenge for all radiological staging of
the neck is micrometastasis. The reason for that is that it has not yet occurred enough structural changes in the LN that would make it possible to detect it. In a study where US was added in patients with N0 OSCC Norling et al found that micrometastasis was present in about 10% of patients, none of which were detected by US [12]. One possibility is that CT, having more difficulty in detecting and evaluating smaller LNs, is more vulnerable in this matter.

The limitation of US is often thought to be experience of the examiners. This study, given that it is a small pilot study, cannot compare the power of US and CT. However, this study shows that a medical student with no prior experience in head and neck surgery or US can find disease not shown on a previous CT. An US examination performed by an experienced surgeon, with basic knowledge of US, would probably be beneficial to the patient. Since many patients have already undergone a CT scan when they arrive at the hospital specialized in this type of tumour surgery an US might initially be viewed as a complement to the CT. This study will also help calculating the power needed in a future and more extensive study.

One possible challenge in this study was the few categories – benign, suspicious, or malignant – used in the evaluation. The effect of this would be that many nodes that meet only one of the criteria gets classified as truly suspicious nodes. This might be avoided by using more grading categories as well as ranking the importance of the different characteristics.

One limitation to the study is the challenge of correctly defining and naming the different levels of the neck. The borders are not defined by clear anatomical structures and one can suspect that there is some variation in how levels are defined. This might be a problem in both the evaluation of the CT scan, performing the US and during the ND. Different positioning of the patient may make the naming even harder.

6. Conclusion

The results indicate that a preoperative US the day before neck surgery can add information about the cervical LN status in patients with HNSCC when performed by
an inexperienced clinician and even though a routine CT has already been made. Further studies must be made to come to any conclusion regarding the efficiency of US compared to CT in this matter. We propose further studies of the use of preoperative US. We also propose however that, given that US is a non-invasive and cost-effective method of assessing the nodal status of the neck, it should take a place in the routine investigation of these patients, especially when the CT was made early in the process.
7. Acknowledgements

First, I would like to thank my supervisor Mathias von Beckerath, MD, PhD, Department of Otolaryngology at the University Hospital Örebro, for his help organizing the study and guidance throughout my writing process. I would also like to thank the personnel at the ENT clinic at the University Hospital Örebro for their help in carrying out this study. Last, I would like to thank my boyfriend Fredrik Bauer for his encouragement and technical support during the entire project.

8. References


Appendix A

Distribution of all harvested sentinel LNs stratified by neck side and tumor subsite

<table>
<thead>
<tr>
<th>Subsite</th>
<th>Ipsilateral neck side level</th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>Floor of mouth</td>
<td>14%</td>
<td>(7/50)</td>
<td>42%</td>
<td>(21/50)</td>
<td>16%</td>
</tr>
<tr>
<td>Tongue*</td>
<td>6%</td>
<td>(2/34)</td>
<td>50%</td>
<td>(17/34)</td>
<td>32%</td>
</tr>
<tr>
<td>Other subsites</td>
<td>50%</td>
<td>(5/10)</td>
<td>30%</td>
<td>(3/10)</td>
<td>10%</td>
</tr>
<tr>
<td>All subsites</td>
<td>15%</td>
<td>(14/94)</td>
<td>43%</td>
<td>(40/94)</td>
<td>21%</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Subsite</th>
<th>Contralateral neck side level</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>Floor of mouth</td>
<td>10%</td>
<td>(5/50)</td>
<td>8%</td>
<td>4%</td>
</tr>
<tr>
<td>Tongue*</td>
<td>3%</td>
<td>(1/34)</td>
<td>–</td>
<td>6%</td>
</tr>
<tr>
<td>Other subsites</td>
<td>–</td>
<td>10%</td>
<td>(1/10)</td>
<td>–</td>
</tr>
<tr>
<td>All subsites</td>
<td>6%</td>
<td>(6/94)</td>
<td>5%</td>
<td>(5/94)</td>
</tr>
</tbody>
</table>

Floor of mouth tumors showed a higher frequency of lymphatic drainage to both the contralateral level 1 and 2 compared with tongue tumors
* Anterior 2/3 of tongue and inferior tongue
** Buccal, hard palate, gingiva and retromolar trigone

Source: [7]