Diagnostic Value of Narrow Band Imaging in Visualization of Pathological Lesions in Larynx and Hypopharynx

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ABSTRACT

Introduction: Narrow Band Imaging (NBI) is an endoscopic optical imaging enhancement technology that improves the contrast of mucosal surface texture and enhances visualization of mucosal and submucosal vasculature. Due to its properties, it can visualize suspected malignant or precancerous lesions earlier than conventional white light endoscopy. The aim of this study was to analyze the benefit of NBI in visualization of precancerous and malignant lesions in preoperative and intraoperative diagnostics and correlation with histopathologic results.

Methods: A total of 589 patients with suspicious laryngeal or hypopharyngeal lesion were investigated using conventional white light endoscopy (WLE) and NBI endoscopy with high-definition TV (HDTV NBI) from 10/2013 to 12/2019. Patients were divided into two groups based on pre-operative NBI examination (group A, 345 patients) and intraoperative NBI examination (group B, 244 patients). All suspicious lesions were graded to 5 types of Ni classification and correlated with histopathologic results. The SPSS version 8.0.4 statistical software package was used for statistical analysis. In diagnosing premalignant and malignant lesions sensitivity, specificity, positive predictive value, and negative predictive value were calculated.

Results: The agreement between NBI endoscopy and histopathological analysis in group A was statistically significant (κ = 0.76, p < 0.001), with a sensitivity of 86.2% (95% IS: 65.4–95.2) and specificity of 90.9% (95% IS: 70.6–94.1). Moreover, in group B was proven almost perfect agreement between NBI and histopathological analysis (κ = 0.8461, p < 0.001), with a sensitivity of 84.0% (95% IS: 60.2–92.4) and specificity of 96.0% (95% IS: 87.0–99.2).

Conclusions: Based on our results, NBI using the Ni classification has great potential in improving diagnosis of precancerous and malignant lesions and correlates strongly with histopathologic results. It serves as a useful adjunct to white light endoscopy in the diagnosis of laryngeal and hypopharyngeal lesions, especially using HDTV NBI.

KEYWORDS
narrow band imaging; white-light endoscopy; Ni classification; precancerous lesion; squamous cell carcinoma; larynx; hypopharynx

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INTRODUCTION

Squamous cell carcinoma (SCC) is one of the most common types of head and neck cancer. Early detection, histopathological diagnosis and treatment significantly improve prognosis and reduce patient morbidity and mortality (1). The most important limiting factor of the early diagnosis is the fact, that precancerous and early malignant changes in the larynx and hypopharynx do not have specific clinical and macroscopical features different from symptoms and mucosal changes triggered by common inflammatory disorders (2). Standard examination methods include rigid or flexible laryngoscopy with white light that lacks sensitivity to precancerous (high-grade dysplasia include carcinoma in situ) or early superficial lesions (T1 or T2 carcinoma). Radiologic examinations such as CT and MRI play a complementary role, as they are the only diagnostic tools that are capable of correctly evaluating deep structures, regional and distant lymph node metastases. Growth and progression of SCC relies on neoangiogenesis, a process where new blood vessels are formed from the surrounding pre-existing blood vessels (1). These new vessels lack the histological architecture and structural anatomy of pre-existing vessels that can be detected by conventional white light endoscopy in later stages of cancer. Earlier detection of these changes is very important for better prognosis of the patients (3). Development of new optical endoscopic methods allowed to detect discrete mucosal changes with pathological vascularization and started rapid progress in the diagnostic process of head and neck tumors. One of endoscopic methods that can visualize these changes and improve the diagnosis is NBI, especially HDTV NBI (2).

NBI as a special endoscopic optical imaging enhancement technology improves the contrast of mucosal surface texture and enhances visualization of mucosal and submucosal vasculature by using specially filtered light (blue and green). The blue light (wavelength 415 nm) corresponds to hemoglobin absorption spectrum and visualizes IPCL (intraepithelial papillary capillary loops). The green light (wavelength 540 nm) penetrates deeper and highlights submucosal vessels. In the definitive image, the mucosal microvascularization appears brown coloured and submucosal vessels are blue coloured (4). Finally, there is a substantial increase of colour contrast between blood vessels and surrounding tissue and a few millimeter changes can be identified. The optimal image is achieved by using HDTV NBI – NBI with high resolution (more than one million pixels) or with ultra high resolution 4K (more than 8 million pixels) (2).

Due to its properties, NBI can visualize suspicious malignant or precancerous lesions earlier than conventional WLE. NBI shows great potential by improving detection of tumours in upper aerodigestive tract (UADT) and currently is the most progressive technological tool in diagnosis of head and neck cancer (5).

Ni et al. proposed a classification system for description of changes in morphology and architecture of IPCL to distinguish between benign and malignant laryngeal lesions (6). This system describes five patterns of morphological changes in IPCL architecture (Table 1). Type I–III are usually associated with benign changes (Figure 1), type IV represents preneoplastic changes and type V neoplastic changes (Figure 2). Type V is further subdivided into Va, Vb and Vc (Table 1) (5). A new classification system as the result of consensus of European laryngological society was published by Arens et al. in 2016 (7). This system is a simplified version of the Ni classification using a descriptive evaluation of microvascular changes. This classification distinguishes only two types of vascular changes – longitudinal and perpendicular (7).

The aim of this study was 1) to evaluate the prehistological diagnostic value of NBI focusing on laryngeal and hypopharyngeal suspicious lesions and 2) to compare NBI with conventional WLE in this setting.

| Classification | Description |
|----------------|-------------|
| Type I         | Thin, oblique and arborescent vessels are interconnected |
|                | IPCLs are almost invisible |
| Type II        | Diameter of oblique and arborescent vessels is enlarged |
|                | IPCLs are almost invisible |
| Type III       | IPCLs are obscured by white mucosa |
| Type IV        | IPCLs can be recognized as small dots |
| Type Va        | IPCLs appear as solid or hollow, with a brownish, speckled pattern and various shapes |
| Type Vb        | IPCLs appear as irregular, tortuous, line – like shapes |
| Type Vc        | IPCLs appear as brownish speckles or tortuous, line – like shapes with irregular distribution, scattered on the tumour surface |

Fig. 1 Reinke oedema of both vocal cords – benign type of vascularization in NBI (the red arrow is showing oblique and arborescent vessels IPCL type II according to Ni classification).
MATERIAL AND METHODS

The presented study was conceived as a prospective study. Patients with clinically suspicious laryngeal or hypopharyngeal lesion that presented to the Department of Otorhinolaryngology and Head and Neck Surgery in Hradec Králové (Czech Republic) from October 2013 to December 2019 were recruited. A total of 589 patients were investigated using conventional WLE and NBI endoscopy with high-definition TV (HDTV NBI) from 10/2013 to 12/2019. Patients were divided into two groups based on preoperative NBI examination (group A = 345 patients) and intraoperative NBI examination (group B = 244 patients). All suspected lesions were graded to 5 types of Ni classification and correlated with histopathologic analysis.

The main reasons for referral were patients with chronic laryngitis, hoarseness or voice changes of unknown etiology lasting more than 3 weeks, benign-appearing lesions (polyps, cysts, vocal cord granulomas), leukoplakias, erythroplakias, macroscopic tumorous lesions of vocal cords or laryngeal papillomatosis. During the consultation in out-patient department (group A), flexible laryngoscopy was performed. After application of mesocain gel (trimecainhydrochloridum carbethopen-decinibromidum 10 mg/g), flexible videolaryngoscope was inserted through the nasal cavity. For local anesthesia of larynx we used lidocaine spray (10% lidocainum) with specially curved applicator that was applied transorally. Flexible endoscopy was performed with an Olympus ENF-VH or ENF-VQ 3.9 mm flexible endoscope connected to an Elvis Exera III CV 170 light source (Olympus Medical System, Tokyo, Japan) while patients were awake and seated. Each patient was examined by conventional WLE followed by NBI mode where the lesions were assessed in real time and captured. In conventional WLE we evaluated macroscopic characteristics of the lesion (chronic laryngitis, cyst, polyp, granuloma, Reinke oedema, leukplakia, erythroplakia, papilloma, exophytic lesion) and the extent and spreading of the lesion (Figure 2). In NBI mode we described changes in microvascularization graded according to Ni classification. Any lesions considered being precancerous were graded IV, lesions suspected from malignancy were given grade V (Va, Vb or Vc). Endoscopic examination was performed only by three physicians. A clinically indicated biopsy was taken under general anaesthesia in the operating room. The biopsy specimens were fixed in 10% formalin and sent for histopathological examination. The tissue was routinely processed, stained with hematoxylin and eosin and examined by an experienced head and neck pathologist. Histopathological diagnoses were then compared to their respective Ni classification (Table 1). Based on this, sensitivity and specificity of NBI using the Ni classification for detection of laryngeal and hypopharyngeal cancer were calculated. Exclusion criteria were allergy to local anaesthetics, previous laryngeal or hypopharyngeal cancer, or inability to undergo flexible endoscopy due to increased gag reflex.

In group B patients underwent examination in general anaesthesia in the operating theatre (direct laryngoscopy or hypopharyngoscopy). After the endotracheal intubation, all anatomical sites were endoscopically evaluated by WLE and NBI using rigid 0° and 30° angled telescopes (Olympus Visera Pro system CV-170 with Olympus OTV S 7 camera head). Attention was paid to the superficial extension of the primary lesion with special emphasis to the additional suspected areas in the entire larynx and hypopharynx. The principles of classification of NBI findings and correlation with histopathological results were identical to the group A.

The SPSS version 8.0.4 statistical software package was used for statistical analysis. Sensitivity, specificity, positive predictive value, and negative predictive value for detection of laryngeal and hypopharyngeal cancer were calculated.
RESULTS

From October 2013 to December 2019, a total of 345 patients were included in group A. There were 248 (71.9%) males and 97 (28.1%) females (male to female ratio 2.5 : 1) aged 34 to 79 years (median 53; mean 58.4 ± 16.5).

In group B total of 244 patients were included. There were 169 (69.3%) males and 75 (30.7%) females (male to female ratio 2.3 : 1) aged 27 to 81 years (median 59; mean 56.2 ± 14.5). Detailed personal data are summarized in Table 2.

**(A)** benign type of vascularization in NBI, **(B)** malignant type of vascularization in NBI.

In group A, we detected pathological findings in 324/345 (94%) patients when using the conventional WLE, especially chronic laryngitis (16%), leukoplakia (22%), erythroplakia (4%), exophytic tumour (35%), papilloma (15%) and Reinke oedema (8%). Using NBI endoscopy, larger extent of the lesion was visualized in 36% and new lesions not detected in white light in 6% cases (Table 3). Histopathological examination was made in 85% (293 patients). If we assumed benign mucosal vascularization in NBI, normal finding was reported by pathologist in 69.1% lesions, dysplastic changes in 26.4%, carcinoma in situ in 1.2% and invasive spinocellular carcinoma in 3.3% (Graph 1A). If we assumed malignant mucosal vascularization in NBI, an invasive spinocellular carcinoma was proven in 46.4% cases, carcinoma in situ in 8.9%, moderate to high-grade dysplasia in 30.4%, low-grade dysplasia in 5.4%, and chronic inflammatory changes without dysplasia in 8.9% cases (Graph 1B).

Character of mucosal vascularization (IPCL) in NBI was correlated with histopathological examination. Sensitivity of NBI in distinction of benign hyper- and parakeratosis, low-grade dysplasia, moderate to high-grade dysplasia from advanced precancerous stages (carcinoma in situ) and invasive spinocellular carcinoma (SCC) was 86.2% (95% IS: 65.4–95.2), while specificity was 90.9% (95% IS: 70.6–94.1). Overall agreement was 86.84% (95% IS: 76.4–93.4). According to Kappa index $\kappa = 0.76$ (95% IS: 0.54–0.96) $(p < 0.001)$, we proved statistically significant agreement between NBI and histopathological examination (Table 3).

**Graph 1** Comparison of vascular pattern of intraepithelial papillary capillary loops in NBI (according to Ni et al. 2011) and the results of histopathological analysis in group A. **(A)** benign type of vascularization in NBI, **(B)** malignant type of vascularization in NBI.

### Tab. 2 Age, sex and comorbidities in two groups of patients.

| Characteristics                  | Group No. I | Group No. II |
|----------------------------------|-------------|--------------|
| men                              | 72.4%       | 69.3%        |
| women                            | 27.6%       | 30.7%        |
| smokers                          | 65.5%       | 71.7%        |
| non-smokers                      | 24.1%       | 19.7%        |
| stop-smokers                     | 10.4%       | 8.6%         |
| age                              | 64.3 y (43–82) | 56.3 y (27–81) |
| hoarseness                       | 79.3%       | 79.1%        |
| chronic laryngitis               | 23.3%       | 29.1%        |
| 1 comorbidity < 60 y             | 74.1%       | 65.8%        |
| ≥ 2 comorbidities > 60 y         | 77.6%       | 72.2%        |
| Extraesophageal reflux           | 17.2%       | 23.4%        |

### Tab. 3 Overview of own results (Statistics: software SPSS version 8.0.4).

| NBI examination                      | Group No. I | Group No. II |
|--------------------------------------|-------------|--------------|
| Larger extent of lesion in NBI       | 36.0%       | 22.1%        |
| New lesions in NBI                   | 6.0%        | 17.6%        |
| Sensitivity                          | 86.2%       | 90.8%        |
| Specificity                          | 90.9%       | 94.7%        |
| Positive predictive value            | 75.8%       | 93.6%        |
| Negative predictive value            | 95.2%       | 86.5%        |
| Overall agreement 95% IS             | 86.84%      | 89.64%       |
| $\kappa$ index (p < 0.001) 95% IS    | 0.76 (0.54–0.96) | 0.8461 (0.8274–0.9654) |
In group B, larger extent of the lesion was visualized in 22.1% and new lesions not detected in conventional white light in 17.6% cases (Table 3). Histopathological examination verified benign findings (hyperkeratosis, parakeratosis, chronic inflammatory changes, polyps, cysts and granulomas) in 37.3% of cases. Recurrent respiratory papillomatosis was present in 14.3% and low-grade dysplastic changes in 8.6% of cases (Graph 2A). As regards precancerous and malignant changes, we proved high-grade dysplasia in 13.9%, carcinoma in situ in 3.3% and invasive SCC in 22.5% of patients (Graph 2B). Statistical analysis confirmed the sensitivity of 90.8% (95% IS: 82–96) and specificity of 94.7% (95% IS: 79.4–94.6). Overall agreement was 89.64% (95% IS: 79.4–94.6). According to Kappa index $\kappa = 0.8461$ (95% IS: 0.8274–0.9654) ($p < 0.001$) we found statistically significant agreement between NBI and histopathological examination (Table 3).

**DISCUSSION**

Patients with UADT tumors are routinely examined by conventional white light endoscopy. It is the most commonly used method to evaluate superficial spreading of SCC. In early stages of cancerogenesis, the tumors of UADT are hardly distinguished from normal tissue and usually not identified by conventional WLE (5). The growth of epithelial malignant tumors leads to escalated neoangiogenesis, which may be observed in endoscopic practice as changes in the IPCL arrangement, diameter, shape and as a loss of regularity (1). Technological improvement in endoscopic methods is the key for early detection of these changes. Alteration of microvascular architecture becomes more visible in high-grade dysplastic changes in comparison to low-grade dysplastic changes (1).

NBI is easily applicable and well tolerated in local anesthetics in out-patient department. Great practical significance is identification of new suspicious lesions. We found out larger extent of suspicious lesions in 36% of patients, and in 6% of cases we identified new lesions that were not be visible by conventional white light endoscopy. Piazza et al. (2010) proved larger extent of lesions in 26% and new lesions not visible in WLE in 9% (8).

Piazza et al. (2012) also proved the benefit of pre-operative using of NBI with detection up to 20 lesions not visible by white light endoscopy. The diagnostic benefit of NBI in better definition of neoplastic superficial spreading is substantially increased with using HDTV NBI (from 20.8% to 42.7%) (9).

Clinical application of NBI endoscopy requires a special practice and defined “learning curve”. Correct recognition and diagnostics of lesions is long-lasting process. Villaseca et al. (2017) demonstrated, that accuracy of detection of malignant lesions increased with longer-lasting practical experience of the examiner and he recommends at least 200 examinations for correct identification of vascular changes (10). Piazza et al. (2010) demonstrated that 13.7% from 58 positive findings in NBI were false positive, if 70% of the findings have been diagnosed during the first three months of using NBI. The advantage of NBI examination in out-patient department is possibility of data recording and fast video analysis in a short time (8).

Piazza et al. (2010) assessed 279 patients with laryngeal cancer and reported a sensitivity of only 61% using flexible NBI alone, but the sensitivity increased substantially to 98% and specificity to 90% when NBI was coupled with HDTV (8). Shoeffel-Havakuk et al. (2017) compared NBI with WLE for the diagnosis of laryngeal cancer and reported a sensitivity of 58.6% when using NBI. Sensitivities reported in this study were based on three expert assessments of NBI and WLE images, not histopathological diagnosis like in other studies (11). Chang et al. (2016) consider NBI as an effective technique with high diagnostic accuracy (98.9%), sensitivity (97.2%) and specificity (100%), and therefore recommend using flexible endoscopy with NBI and taking biopsy in out-patient department. Diagnostic accuracy was not affected by size, localization, “learning” or history of head and neck cancer (12). In recent
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In our study we achieved sensitivity 86.2% and specificity 90.6% and according to Kappa index there was substantial agreement between NBI and histopathological results ($\kappa = 0.76; p < 0.001$).

NBI has also limitations in preoperative diagnosis. To obtain a valid interpretation it is necessary to keep minimal distance between distal part of endoscope to the examined mucosa (3 mm and less) which can be difficult in patients with strong gag reflex even after using local anaesthetics. Another disadvantage is the impossibility to assess vertical extent of lesion. The cause of false negative findings might be due to hyperkeratosis making the assessment of pathological vascularization impossible (3, 18). In our study there were two patients with verrucous SCC where it was not possible to assess IPCL. In one patient with adenoid cystic carcinoma superficial pathological vascularization was not identified due to submucosal spreading of the tumour. Assessment of leukoplakia in NBI is controversial as well, described by a well-known phenomenon – the “umbrella effect”, which reflects the submucosal vascular pattern being hidden under the hyperkeratotic plaque (Klimza et al. 2017) (19). To overcome the “umbrella effect”, the NBI was used to categorise submucosal vascular pattern surrounding the plaque, as illustrated in Figure 3B (Klimza et al. 2017, Stanikova et al. 2017) (18, 19). Laryngeal recurrent respiratory papillomatosis (RRP) is another problematic lesion that can be confused with SCC (Figure 4). In our study we found 27 patients with RRP (8.3%), 3 cases were initially suspected from malignancy and proved to be false positive. However, if RRP is suspected other synchronous papillomatous lesions (Jackowska et al. 2018) can be detected in NBI. In our study NBI revealed other lesions not visible in WLE in 14/27 (51.8%) (20).

In peroperative using of HDTV NBI we demonstrated significant difference in comparison with conventional WLE. We identified larger extent of lesions in 22.1% and new suspicious lesions not visible by WLE in 17.6%. The sensitivity was 90.8%, specificity 94.7% and according to Kappa index ($\kappa = 0.8461; p < 0.001$) almost perfect agreement between NBI and histopathological examination has been proven. We had low amount of false positive results and high number of true negative results, and therefore the specificity is so remarkable.
It is difficult to determine margins between physiological and pathological findings when taking “ideal” biopsy in conventional WLE. There are many studies that evaluate positive intraoperative effect of NBI to reduce the number of positive resection margins (21, 22). They proved benefit of NBI in intraoperative diagnostics for better visualization of margins in superficial spreading lesions what is important in endoscopic surgery. Garofolo et al. (2015) proved significant reduction of positive resection margins using NBI (from 23.7% to 3.6%) in patients with early glottic carcinoma (cordectomy type I and II) (21). Piazza et al. (2010) confirmed the significance of NBI in 26 laryngeal tumors where HDTV NBI used intraoperatively led to enlargement of resection margins (23). In a recent study performed by Klimza et al. (2019) pathological lesions not visible in WLE were identified in 13.6% and histopathologically confirmed in 88.8% (33.3% invasive SCC, 66.6% high-grade dysplasia and carcinoma in situ) (22). Orita et al. (2008) confirmed intraoperative benefit of NBI during the endoscopic hypopharyngeal surgery (24).

CONCLUSION

The results in all above mentioned published studies as well as our study confirmed that NBI is a reliable method in early detection of precancerous and malignant changes, for discriminating between benign and malignant patterns in prehistological diagnosis.

Great practical significance is identification of new suspicious lesions not visible by conventional white light endoscopy that we proved preoperatively in 6% and intra-operatively in 17.6% of cases. We confirmed low amount of false positive results and high number of true negative results, and therefore the specificity was so remarkable. Especially in peroperative using of HDTV NBI we demonstrated almost perfect agreement between NBI and histopathological examination.

NBI has been proved to be a “gold standard” and routine diagnostic tool for assessment of laryngeal and hypopharyngeal pathology.

CONFLICT OF INTEREST

The authors declare that there is no actual or potential conflict of interest in relation to this article.

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