**ORIGINAL ARTICLE**

**CYSTIC LYMPH NODE METASTASIS IN PATIENTS WITH HEAD AND NECK CANCER: AN HPV-ASSOCIATED PHENOMENON**

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Abstract: Background. Cystic lymph node metastases have been associated with tonsil cancer. A subset of oropharyngeal cancers contain human papillomavirus (HPV) DNA. The clinical and virologic associations of cystic nodal metastasis in head and neck cancer (HNSCC) were investigated.

Methods. A retrospective review of patients undergoing neck dissection between 2002 and 2004 identified patients with cystic lymph node metastases. Clinical and pathologic information was recorded. The presence of HPV DNA was analyzed using in situ hybridization in a subset of patients.

Results. One hundred neck dissections were reviewed and 20 had cystic nodes. Seventeen of these patients had primary tumors arising in the palatine or lingual tonsil. Three were “unknown primary.” HPV DNA was present in 11 of 13 patients with cystic metastases with tissue available for analysis (87%). No HPV DNA was identified in tumor from 21 patients with solid nodal metastasis ($p < .0001$).

Conclusion. Cystic cervical lymph node metastasis is strongly associated with HPV-related tonsillar HNSCC.

Keywords: head and neck cancer; human papillomavirus; lymph nodal metastasis; cystic neck mass; oropharyngeal cancer

**Historically,** lateral neck cystic masses were commonly judged to be branchial cleft cysts. The proportion of metastatic squamous cell carcinoma in cysts initially presumed to be of branchial cleft origin has been reported to range from 11% to 21%.1–3 In 1881, Von Volkman4 first theorized that malignant lesions of the lateral aspect of the neck arise from remnants of the branchial clefts. At that time, these lesions were labeled “branchiogenic carcinomas” and no attempt was made to search for the primary tumor site. In 1944, Hayes Martin5,6 urged that a biopsy be the initial and not the final step of the clinical investigation and that a thorough search for a primary tumor be made. He contended that these tumors were nearly always metastases from a primary in the head and neck, which could and should be treated.
He proposed that the diagnosis of branchiogenic carcinoma, as an entity distinct from a cystic nodal metastasis, be made only if strict criteria were satisfied. Gourin and Johnson found no examples that met these criteria in their series of 121 patients with an initial diagnosis of lateral cervical cyst, nor did Neel and Pemberton in 319 patients with lateral neck cysts. Thus, it is now thought that branchial cleft carcinoma either does not occur or is exceedingly rare, and instead these are cases of cystic metastasis from a tonsillar primary squamous cell carcinoma (SCC). The role of human papillomavirus (HPV) in HNSC arising in palatine and lingual tonsils is now well documented. HPV DNA can be identified in up to 70% of head and neck squamous cell carcinoma arising in these sites.

The present work is a retrospective study intended to better elucidate the entity of cystic lymph node metastasis as a feature of a subset of head and neck squamous cell cancer.

PATIENTS AND METHODS
A retrospective chart review was undertaken of all neck dissections performed for newly diagnosed head and neck squamous cell carcinoma (HNSCC) in the Department of Otolaryngology–Head and Neck Surgery at Johns Hopkins Medical Institutions between the years 2002 and 2004. Patients who had undergone neck dissections were identified and classified according to primary tumor site. Sixteen patients who had a neck dissection performed after treatment failure with chemoradiation were excluded from the study.

By review of CT/MRI reports, all patients with radiographic evidence of cystic nodal metastases were identified. Cystic nodes were defined as round or ovoid masses with a thin (<2 mm) enhancing capsule, homogeneous fluid content, and no internal complex, irregular, or solid area (Figure 1). Nodes with thicker solid walls and irregular, complex central low attenuation were classified as necrotic (Figure 2).

Demographic information, risk factors, primary tumor site, and T and N classification for each subject were documented (Table 1). Cytology and pathology information such as diagnostic fine-needle aspiration (FNA) results, number of involved nodes, and extranodal extension was recorded.

In Situ Hybridization. HPV-16 detection in formalin-fixed and paraffin-embedded tissues (primary tumor and/or nodal metastasis) was performed using the in situ hybridization catalyzed signal amplification method for biotinylated probes (DAKO GenPoint, Carpinteria, CA). This catalyzed signal amplification system permits visualization of single copies of HPV-16 in infected...
cells [PMID: 9796725]. Briefly, 5-μm tissue sections underwent deparaffinization, heat-induced target retrieval in citrate buffer, and digestion using Proteinase K (Roche Diagnostics, Indianapolis, IN). Slides were subsequently hybridized with a biotinylated HPV-16 type specific probe (DAKO). Signal amplification was performed by consecutive application of streptavidin-HRP complex, biotinyl tyramide, and streptavidin-HRP complex. Visualization of positive hybridization signals was performed by incubation with the chromogenic substrate diaminobenzidine. The presence of punctate hybridization signals within the nuclei of tumor cells was regarded as positive for the presence of HPV-16. Interpretation of staining was performed without knowledge of tumor origin.

RESULTS

One hundred patients who underwent neck dissection as part of their initial treatment of suspected HNSCC metastasis were identified. Primary sites were laryngeal (n = 23), hypopharyngeal (n = 3), oropharyngeal (n = 44), oral cavity tumors (n = 22), unknown (n = 6), and skin (n = 2). Neck dissection pathology reports showed 38 patients with no nodal involvement, 24 with 1 involved lymph node, and 38 with ≥2 involved nodes. Thirty-two patients had extranodal extension.

Twenty patients were found to have cystic nodal metastases, as confirmed by radiologic report and review of films. The age range of these patients was 35 to 80 years (mean, 57.9 years). Ten patients were smokers, 5 used alcohol regularly, and 6 (31%) were nonsmokers and non-drinkers. Ten patients with cystic lymph node metastasis (CLNM) were found to have a tongue base primary, 7 had a palatine tonsil primary, and 3 had a histologically unknown primary. Thus 17 of 20 patients with cystic lymph node metastases had tumors arising from the palatine or lingual tonsil. Overall, 20% of HNSCC with neck metastasis from all primary sites had CLNM, including 41% of patients with tongue base and tonsil primaries (p <.001). Fifteen patients with cystic nodes had had an FNA performed. Results of cytopathologic evaluation were positive for carcinoma in 12 (80%) of these patients.

Tissue samples from the lymph node metastasis and/or primary site was available from 15 of the 20 patients with cystic nodal metastases. In situ hybridization of 13 of 15 patients (87%) confirmed the presence of HPV DNA in tumor cells, including 2 metastases from unknown primary site. Tumor from both nodal metastasis and the primary site was available for 3 patients, and in each patient, both primary and metastatic cells contained HPV. Cells harvested by FNA were available for analysis in 1 patient and were also strongly HPV positive. HPV in situ hybridization results were also determined for 21 patients with solid lymph node metastases. Nine of these patients had some evidence of central necrosis in at least 1 node. The primary sites involved were oral cavity (n = 11), oropharynx (n = 4), hypopharynx (n = 2), and larynx (n = 4). HPV DNA was present in none of these patients (chi-square comparing cystic and solid nodes for HPV: p <.0001).

DISCUSSION

The appearance of cystic cervical nodal metastases is a distinctive, specific, and easily recognizable phenomenon that occurs in a subset of HNSCC. It has been recognized for some time that there is a marked predilection for cystic morphology in cervical metastases to be associated with SCC of Waldeyer’s ring. The incidence of
cystic lymph nodes arising from SCC of Waldeyer’s ring has been reported to be as high as 33% to 50%. The fact that 3 of our cases were examples of “unknown primary” HNSCC is consistent with the association of cystic nodes with oropharyngeal primary disease. One published study reports that 10% of malignant cystic metastasis ultimately found to originate in Waldeyer’s ring were cases in which no primary was initially apparent. In a series of unknown primary HNSCC from our institution, all primary tumors that were eventually detected were in the tonsil or tongue base.

The association between HPV and tonsil/tongue base cancer has been well documented in the past decade. In our institution, the vast majority of HNSCC cases with biologically active HPV are cases from the tonsil or base of tongue, and over 50% of all tonsil/base of tongue HNSCC contained HPV. In these sites, HPV-16 infection specifically targets the specialized reticulated epithelium that lines the tonsillar crypts. Integration of HPV as indicated by in situ hybridization is a rigorous method to determine the biologically functional presence of the virus in cancer cells. Not all HPV-associated oropharyngeal cancers spawn cystic nodes (only 41% of palatine or lingual tonsil cancers in our population had cystic nodal metastases), but the overwhelming majority of cystic nodes were from HPV-associated tonsillar cancers (palatine and lingual).

In a series of 101 patients with SCC arising in Waldeyer’s ring, Regauer et al found 61% to harbor cystic metastasis. Of these, 35 were of primary palatine tonsil SCC, 22 were of primary base of tongue tumors, and 2 patients were of nasopharyngeal tumors. As nasopharyngeal carcinoma is rare in our geographical area and initially not treated surgically, none appeared in our series.

A small number of published studies identify the association between oropharyngeal HNSCC and cystic nodal metastasis. In one such series of 136 patients with cystic metastasis, Thompson and Heffner found the primary site to be palatine tonsil in 64%. They stressed that this group of tumors represents a subtype of tonsil cancer. The unique feature of this subtype of tonsil cancer is a propensity for moderately large cystic metastasis in lymph nodes of the jugulodigastric region coupled with relatively small or occult primary tumors. Histologically, these tumors arise from a transitional type, tonsillar crypt epithelium, as distinct from well-differentiated SCC, which may also arise in the oropharynx.

The phenomenon of cystic metastasis is not limited to oropharyngeal primary malignancies and has also been reported to occur in association with primary cancer of the thyroid (papillary type), esophagus, and uterine cervix. It is noteworthy that uterine cervix SCC is also an HPV-related cancer.

In many radiographic studies, cystic lymph nodes are grouped together with central lymph node necrosis because of the common presence of hypodense nodal content. The process of central necrosis is thought to occur when cancer growing in the medullary portion of a lymph node outstrips its blood supply. Our results indicate that cystic nodal metastasis may be distinct from cases of necrotic nodal degeneration in which the area of central lucency is more irregular in shape and content, and the viable rim of tumor is variable in thickness. The content of necrotic nodes with caseous debris is also distinctively different from the serous fluid in a cystic node. Among the cancers producing solid metastases with central necrosis, HPV DNA was identified in 0 of 9 patients available for in situ hybridization, including 3 patients with oropharyngeal primaries.

In 1 ultrasonographic study, Ying explains that intranodal necrosis can be classified into coagulation necrosis and cystic necrosis, which can be differentiated by ultrasound. The author states that coagulation necrosis appears as an intranodal echogenic focus, whereas cystic necrosis appears as an echolucent area within the lymph nodes.

Morphologically, cystic nodal metastases are well circumscribed and usually surrounded by a smooth fibrous capsule. They are filled with fluid that may be thin and straw colored initially and over time or following manipulation (perhaps including FNA) may develop into granular, thick, tenacious, brown, and/or hemorrhagic fluid.

The mechanism of cyst formation in cystic metastases has not been fully elucidated. The phenomenon of cystic nodal metastasis has been postulated to occur secondary to pseudocystic change resulting from spontaneous degradation of keratin and cellular debris within the carcinomatous lymph node deposit. However, some favor the formation of a true cyst lined by neoplastic epithelium in the central portion of the lymph node with lymphoid elements occupying the periphery of the node. A collection of cellular debris has been observed in needle aspirates of cystic metastases. It has been suggested that the carcinomatous surface epithelium does not desquamate in...
the early stages of cyst formation, thus accounting for the hypocellularity of many cystic aspirates. Because microcystic spaces and cyst formation are often demonstrated in primary squamous cell carcinoma of Waldeyer’s ring, cyst formation has been proposed as an intrinsic property of transformed keratinocytes from these sites, which, when metastatic, are still able to simulate the growth behavior and histological patterns of the parent cell.

Patients seen with cystic nodal metastases frequently report a sudden recognition of the presence of a neck mass indicative of enlargement at a rate beyond that expected as due to tumor cell proliferation alone. This “overnight” appearance of a large neck mass could occur because of the sudden blockage of the flow of lymphatic fluid passing through a node that has an established metastatic colony filling a potential space lined with tumor cells.

Investigation of lymph node metastases in HNSCC typically includes an FNA. Schwarz et al19 demonstrated that FNA had an overall sensitivity of 92% and a 100% positive predictive value for the diagnosis of SCCs of the head and neck. This, however, has not been the common experience with cystic metastases. Gourin and Johnson1 reported a false-negative rate of 42% with FNA aspirate of cystic nodes. Another study20 reported sensitivities ranging from 33% to 50%. The difficulty of FNA diagnosis in cases of cystic metastases may be due to hypocellularity, inflammatory cells, epithelial cell changes due to inflammation, and presence of cellular debris.21 Cytologic features suggesting malignancy may be lacking, resulting in a mistaken diagnosis of benign squamous cyst. Confirmation of malignancy may not occur until excisional biopsy. However, in our series, FNA results were positive for malignant cells in 80% of cystic nodal patients, indicating that FNA remains of great value in the setting of cystic metastases. Furthermore, we have shown that analysis of FNA specimens for HPV correlates strongly with metastatic carcinoma from tonsillar HNSCC.22

In our patients with cystic nodal metastases, the mean age was 58 years, and most were either smokers or alcohol users. However, the literature indicates that cystic SCC metastases may not be associated with the usual risk factors of smoking and alcohol abuse,23 and are often seen in a younger patient population than those with solid metastatic SCC.24 These features are consistent with the demographics reported for HPV related HNSCC.

**CONCLUSION**

This series clearly demonstrates the relationship between cystic lymph node metastases, HPV, and tonsil/tongue base HNSCC. A cystic lymph node within the neck of an adult should raise a suspicion of a malignant process arising in the lymphoepithelial tissue of the tonsil or tongue base. Initial investigation should include imaging studies and FNA. If FNA is inconclusive, the neck node should be biopsied, with frozen section evaluation and neck dissection if cancer is confirmed. A panendoscopy and tonsillectomy should be performed as well. Molecular analysis of needle aspirate material or excisional biopsy for HPV DNA can provide powerful evidence of a lingual or palatine tonsillar origin of cystic nodal metastases.

**REFERENCES**