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Evidence-Based Series 5-9 Version 2

A Quality Initiative of the Program in Evidence-Based Care (PEBC), Cancer Care Ontario (CCO)

Routine HPV Testing in Head and Neck Squamous Cell Carcinoma

The Expert Panel on HPV Testing in Head and Neck Squamous Cell Carcinoma

January 13, 2020

An assessment conducted in December 2023 deferred review of Evidence-Based Series 5-9 Version 2. This means that the document remains current until it is assessed again next year. The PEBC has a formal and standardized process to ensure the currency of each document (PEBC Assessment & Review Protocol)

EBS 5-9v2 is comprised of 4 sections. You can access the summary and full report here:
https://www.cancercareontario.ca/en/guidelines-advice/types-of-cancer/581

Section 1:	Guideline Recommendations (ENDORSED)
Section 2:	Evidentiary Base
Section 3:	EBS Development Methods and External Review Process
Section 4:	Document Assessment and Review

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Evidence-Based Series 5-9

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Routine HPV Testing in Head and Neck Squamous Cell Carcinoma

Guideline Report History

GUIDELINE VERSION	SYSTEMATIC REVIEW		PUBLICATIONS	NOTES and KEY CHANGES
	Search Dates	Data		
Original May 13, 2013	1996 to Apr 2013	Full Report	Web publication	N.A.
Version 2 January 13, 2020	2013 to Feb 2019	New data found in Section 4: Document Assessment and Review	Updated web publication	2013 recommendations are ENDORSED

Evidence-Based Series 5-9: Section 1

A Quality Initiative of the Program in Evidence-Based Care (PEBC), Cancer Care Ontario (CCO)

Routine HPV Testing in Head and Neck Squamous Cell Carcinoma: Guideline Recommendations

The 2013 guideline recommendations have been ENDORSED, which means that the recommendations are still current and relevant for decision making. Please see Section 4: Document Assessment and Review for a summary of updated evidence published between 2013 and 2019, and for details on how this guideline was ENDORSED.

GUIDELINE OBJECTIVES

To evaluate the appropriateness of, and make recommendations on, routine testing for human papillomavirus (HPV) status in adult patients with primary, or neck nodal metastatic, squamous cell carcinoma (SCC) of the head and neck.

TARGET POPULATION

Adult patients with squamous cell carcinomas arising in oropharynx, larynx, hypopharynx, nasopharynx, sinonasal tract, or oral cavity subsites or an unknown primary head and neck site.

INTENDED USERS

This guideline is targeted for:

1. Clinicians involved in the delivery of care of adult patients with head and neck squamous cell carcinoma (HNSCC).
2. Pathologists involved in the evaluation of HNSCCs.

RECOMMENDATIONS, KEY EVIDENCE, AND JUSTIFICATION

RECOMMENDATION 1

The tumours of all adult patients presenting with oropharyngeal squamous cell carcinomas should be routinely tested for HPV status.

Summary of Key Evidence for Recommendation 1

- A meta-analysis showed a definite survival benefit for HPV-positive patients compared to those whose tumour was HPV negative in terms of overall survival (OS) (HR: 0.43 (95%CI: 0.32-0.58), progression-free survival (PFS) (HR: 0.40, 95%CI: 0.28-0.56), and disease-specific survival (DSS) (HR: 0.45 (95%CI: 0.27-0.76).
- A published data meta-analysis by Ragin and Taioli (1) demonstrated that patients with HPV-positive oropharyngeal tumours had a 28% reduced risk of death compared to

patients with HPV-negative oropharyngeal tumours (HR: 0.72, 95%CI: 0.5-1.0). Similar results were calculated for disease-specific survival (DSS) (HR: 0.51, 95%CI: 0.4-0.7). However, no benefit in overall survival (OS) or DSS was seen in HPV-positive versus negative patients with non-oropharyngeal tumours.

Justification for Recommendation 1
There is evidence from a meta-analysis of randomized trials that HPV-positivity is a strong predictor of prognosis in patients with oropharyngeal squamous cell carcinoma. In addition, it is likely that HPV status will influence management decisions in the near future and is now regarded as a mandatory stratification factor for clinical trials. Therefore, even though at this time no recommendation can be made to base clinical management decisions on HPV status, the valuable prognostic benefits of HPV testing are sufficient to warrant routine testing.

Qualifying Statements for Recommendation 1
- The above recommendation only applies to patients with squamous cell carcinoma of the oropharynx, which includes tonsil, base of tongue, soft palate and associated pharyngeal walls. The data and recommendation do not apply to patients with non-oropharyngeal cancers.
- Altering management decisions based on results from HPV testing is not recommended beyond the context of a clinical trial at this time.

RECOMMENDATION 2
It is recommended that the neck nodal tissue of patients with metastatic squamous cell carcinoma to neck nodes from an unknown head and neck primary be routinely tested for HPV status.

Summary of Key Evidence for Recommendation 2
- Twelve studies (2-13) found the prevalence of HPV-positive lymph node metastases ranged from 0%-19% in patients with non-oropharyngeal primary sites compared to 66%-87% in those whose primary tumour originated in the oropharynx.

Justification for Recommendation 2
The evidence indicates that there is relationship between HPV positivity and whether the initial cancer arises in the oropharynx or not. As detection of the primary tumour offers a reduction in morbidity due to the benefits of localized treatment, the additional diagnostic information provided by HPV status is sufficient to warrant routine testing of these tissues.

Qualifying Statements for Recommendation 2
Currently, there are no standardized protocols or extensive published experience regarding the performance of p16 immunohistochemical (IHC) or HPV in situ hybridization (ISH) in fine-needle aspiration (FNA) or cytology material from metastatic squamous cell carcinoma to cervical lymph nodes.

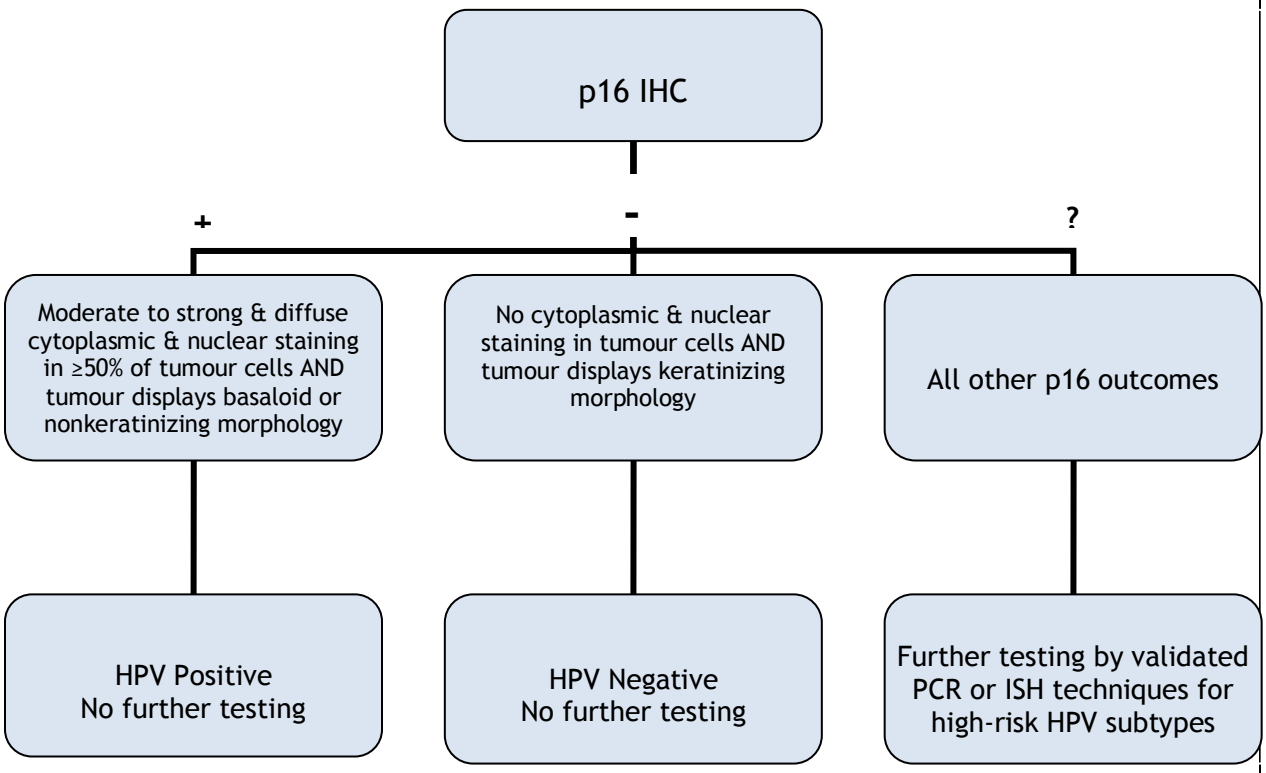
RECOMMENDATION 3

It is recommended that HPV status in oropharyngeal SCC be initially determined using immunohistochemical (IHC) staining for p16.

IHC staining for p16 can be considered positive when the following three criteria are met:

- cytoplasmic and nuclear staining
- staining is moderate to strong and diffuse
- staining is present in at least 70%* of tumour cells (*See Section 4 for explanation)

A validated polymerase chain reaction (PCR) or in situ hybridization (ISH) technique for high-risk HPV subtypes may be necessary to confirm p16 results in selected cases according to the following algorithm:



Summary of Key Evidence for Recommendation 3

- The above recommendations are based on a comparison of HPV diagnostic testing methods published in the literature. Thirteen retrospective cohort studies (14-26) were included in this guideline. The evidence suggests that, in patients with OPSCC, the performance of the three main techniques – PCR-based amplification, DNA ISH, and p16 IHC – is comparable.
  - PCR amplification of HPV DNA showed a sensitivity of 97% and specificity of 87%
  - DNA ISH showed a sensitivity that ranged from 83% to 93% and a specificity that ranged from 88% to 100%
  - IHC staining for p16 showed a sensitivity and specificity that ranged from 89% to 100% and 38% to 94%, respectively

Technical Considerations for Recommendation 3
While it is not possible to make evidence-based recommendations regarding the minimum set of criteria requiring adherence in a pathology laboratory with respect to HPV testing at this time, the following guidance is offered based on expert opinion and a consensus process by members of the Head and Neck DSG:

- Analysis should be performed on sections from paraffin blocks or unstained slides cut at 4 microns
- In cases of metastatic disease, where a core biopsy may not be a possibility, all efforts should be made to obtain enough tissue with FNA to prepare cell blocks.

Justification for Recommendation 3
The current evidence suggests that PCR, DNA ISH, and IHC staining are all comparable. With no unequivocal evidence exclusively supporting any particular scheme, the Head & Neck Disease Site Group believes this scheme is practical and simple, and it minimizes the impact of testing on available pathology resources and is appropriate until such time as further evidence becomes available. The Head & Neck DSG acknowledges that the algorithm may be considered controversial by some, but it is believed to address the proficiencies that are most readily available in laboratories across the province.

Qualifying Statements for Recommendation 3

- The Head & Neck DSG considers quality assurance and quality control in HPV-status testing to be paramount. As such, all testing should be carried out in licensed and accredited laboratories, and test results should be interpreted by experienced pathologists/scientists. Laboratories need to follow proper quality control and participate in external proficiency testing to ensure test accuracy. Further discussion of specific quality and proficiency parameters necessary for individual laboratories performing HPV-status testing is beyond the scope of this guideline.
- Qualitative HPV PCR assay detection alone should be avoided
- The above recommendations do not apply to samples from dental procedures.

FUTURE RESEARCH
Insufficient data currently exist to assess the prognostic benefit of HPV positivity in SCC of the larynx, hypopharynx, nasopharynx, sinonasal tract and oral cavity. There is evidence in the literature to suggest that the prevalence of HPV in these subsites may be higher than originally believed. Meta-analyses (1,27,28) report a pooled prevalence in the oral cavity and the larynx as high as 40% and 24%, respectively. Lip and oral cavity, pharynx, larynx, nasopharynx and lymph nodes combined have a reported pooled HPV prevalence of 32%. Such values warrant further prospective local data collection via the creation of a provincial patient registry to establish the prevalence of HPV-associated SCC and to clarify the prognosis associated with HPV positivity in these patients. This will ensure the acquisition and availability of evidence upon which future clinical decisions can be based.

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Updating

All PEBC documents are maintained and updated
as described in the PEBC Document Assessment and Review Protocol.

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REFERENCES

1. Ragin CC, Taioli E. Survival of squamous cell carcinoma of the head and neck in relation to human papillomavirus infection: review and meta-analysis. Int J Cancer. 2007;121(8):1813-20.
2. Begum S, Gillison ML, Ansari-Lari MA, Shah K, Westra WH. Detection of Human Papillomavirus in Cervical Lymph Nodes: A Highly Effective Strategy for Localizing Site of Tumor Origin. Clin Cancer Res. 2003;9(17):6469-75.
3. Begum S, Gillison ML, Nicol TL, Westra WH. Detection of human papillomavirus-16 in fine-needle aspirates to determine tumor origin in patients with metastatic squamous cell carcinoma of the head and neck. Clin Cancer Res. 2007;13(4):1186-91.
4. Begum S, Westra WH. Basaloid squamous cell carcinoma of the head and neck is a mixed variant that can be further resolved by HPV status. Am J Surg Pathol. 2008;32:1044-50.
5. Compton AM, Moore-Medlin T, Herman-Ferdinandez L, Clark C, Caldito GC, Wang XI, et al. Human papillomavirus in metastatic lymph nodes from unknown primary head and neck squamous cell carcinoma. Otolaryngol Head Neck Surg. 2011;145(1):51-7.
6. Desai PC, Jaglal MV, Gopal P, Ghim SJ, Miller DM, Farghaly H, et al. Human papillomavirus in metastatic squamous carcinoma from unknown primaries in the head and neck: A retrospective 7 year study. Exper Molec Pathol. 2009;87(2):94-8.
7. El-Mofty SK, Zhang MQ, Davila RM. Histologic identification of human papillomavirus (HPV)-related squamous cell carcinoma in cervical lymph nodes: a reliable predictor of the site of an occult head and neck primary carcinoma. Head Neck Pathol. 2008;2(3):163-8.
8. Hoffmann M, Gottschlich S, Gorogh T, Lohrey C, Schwarz E, Ambrosch P, et al. Human papillomaviruses in lymph node neck metastases of head and neck cancers. Acta Oto-Laryngologica. 2005;125 (4):415-21.
9. Jannapureddy S, Cohen C, Lau S, Beitler JJ, Siddiqui MT. Assessing for primary oropharyngeal or nasopharyngeal squamous cell carcinoma from fine needle aspiration of cervical lymph node metastases. Diagn Cytopathol. 2010;38(11):795-800.
10. Park JM, Jung CK, Choi YJ, Lee KY, Kang JH, Kim MS, et al. The use of an immunohistochemical diagnostic panel to determine the primary site of cervical lymph node metastases of occult squamous cell carcinoma. Human Pathol. 2010;41(3):431-7.
11. Weiss D, Koopmann M, Rudack C. Prevalence and impact on clinicopathological characteristics of human papillomavirus-16 DNA in cervical lymph node metastases of head and neck squamous cell carcinoma. Head Neck. 2011;33(6):856-62.
12. Zhang MQ, El-Mofty SK, Davila RM. Detection of human papillomavirus-related squamous cell carcinoma cytologically and by in situ hybridization in fine-needle aspiration biopsies of cervical metastasis: a tool for identifying the site of an occult head and neck primary. Cancer. 2008;114(2):118-23.
13. Park GC, Lee M, Roh JL, Yu MS, Choi SH, Nam SY, et al. Human papillomavirus and p16 detection in cervical lymph node metastases from an unknown primary tumor. Oral Oncol. 2012;48(12):1250-6
14. Agoston ES, Robinson SJ, Mehra KK, Birch C, Semmel D, Mirkovic J, et al. Polymerase chain reaction detection of HPV in squamous carcinoma of the oropharynx. Am J Clin Pathol. 2010;134(1):36-41.
15. Lewis JS, Jr., Thorstad WL, Chernock RD, Haughey BH, Yip JH, Zhang Q, et al. P16 positive oropharyngeal squamous cell carcinoma:an entity with a favorable prognosis regardless of tumor HPV status. Am J Surg Pathol. 2010;34(8):1088-96.
16. Singhi AD, Westra WH. Comparison of human papillomavirus in situ hybridization and p16 immunohistochemistry in the detection of human papillomavirus-associated head and neck cancer based on a prospective clinical experience. Cancer. 2010;116(9):2166-73.

17. Shi W, Kato H, Perez-Ordonez B, Pintilie M, Huang S, Hui A, et al. Comparative prognostic value of HPV16 E6 mRNA compared with in situ hybridization for human oropharyngeal squamous carcinoma. J Clin Oncol. 2009;27(36):6213-21.
18. Smeets SJ, Hesselink AT, Speel EJM, Haesevoets A, Snijders PJF, Pawlita M, et al. A novel algorithm for reliable detection of human papillomavirus in paraffin embedded head and neck cancer specimen. Int J Cancer. 2007;121(11):2465-72.
19. Jordan RC, Lingen MW, Perez-Ordonez B, He X, Pickard R, Koluder M, Jiang B, Wakely P, Xiao W, Gillison ML. Validation of Methods for Oropharyngeal Cancer HPV Status Determination in US Cooperative Group Trials. Am J Surg Pathol. 2012;36(7):945-54.
20. Schache AG LT, Risk JM, Filia A, Jones TM, Sheard J, Woolgar JA, et al. Evaluation of human papilloma virus diagnostic testing in oropharyngeal squamous cell carcinoma: sensitivity, specificity, and prognostic discrimination. Clin Cancer Res. 2011;17(19):6262-71.
21. Kuo KT HC, Lin CH, Kuo LT, Huang SH, Lin MC. The biomarkers of human papillomavirus infection in tonsillar squamous cell carcinoma-molecular basis and predicting favorable outcome. Modern Pathol. 2008;21(4):376-86.
22. Bishop JA, Ma XJ, Wang H, Luo Y, Illei PB, Begum S, et al. Detection of transcriptionally active high-risk HPV in patients with head and neck squamous cell carcinoma as visualized by a novel E6/E7 mRNA in situ hybridization method. Am J Surg Pathol. 2012 December;36(12):1874-82.
23. Hoffmann M, Tribius S, Quabius ES, Henry H, Pfannenschmidt S, Burkhardt C, et al. HPV DNA, E6iI-mRNA expression and p16INK4A immunohistochemistry in head and neck cancer - How valid is p16INK4A as surrogate marker? Cancer Letters. 2012 01 Oct;323(1):88-96.
24. Pannone G, Rodolico V, Santoro A, Lo Muzio L, Franco R, Botti G, et al. Evaluation of a combined triple method to detect causative HPV in oral and oropharyngeal squamous cell carcinomas: p16 immunohistochemistry, consensus PCR HPV-DNA, and in situ hybridization. Infectious Agents Cancer. 2012;7(1).
25. Evans MF, Matthews A, Kandil D, Adamson CS-C, Trotman WE, Cooper K. Discrimination of 'driver' and 'passenger' HPV in tonsillar carcinomas by the polymerase chain reaction, chromogenic in situ hybridization, and p16(INK4a) immunohistochemistry. Head Neck Pathol. 2011;5(4):344-8. PubMed PMID: 21786153.
26. Klussmann JP, Weissenborn SJ, Wieland U, Dries V, Eckel HE, Pfister HJ, et al. Human papillomavirus-positive tonsillar carcinomas: A different tumor entity? Med Microbiol Immunol. 2003;192(3):129-32.
27. Termine N, Panzarella V, Falaschini S, Russo A, Matranga D, Lo Muzio L, et al. HPV in oral squamous cell carcinoma vs head and neck squamous cell carcinoma biopsies: a meta-analysis (1988-2007). Ann Oncol. 2008;19(10):1681-90.
28. Kreimer AR, Clifford GM, Boyle P, Franceschi S. Human papillomavirus types in head and neck squamous cell carcinomas worldwide: a systematic review. Cancer Epidemiol Biomarkers Prev. 2005;14(2):467-75.