Ancillary studies, including viral testing (Core and Non-core)

Reason/Evidentiary Support

In resource-limited practices (or when only extremely limited biopsy samples are available that preclude further testing etc.) where p16/HPV (oropharynx) or Epstein-Barr Virus (EBV) (nasopharynx) testing cannot be performed, staging and treatment of patients will be inherently different.[1] The American Joint Committee on Cancer (AJCC) and the Union for International Cancer Control (UICC) recommend that oropharyngeal squamous cell carcinomas that cannot be tested for p16/HPV be regarded and treated as HPV-negative. This recommendation should be followed for the completion of the International Collaboration on Cancer Reporting (ICCR) dataset.

Given that most HPV-related oropharyngeal squamous cell carcinomas are nonkeratinizing morphologically, arise deep in the tonsillar parenchyma, have cystic nodal metastases, and may have particular clinical features such as arising in non-smokers who are younger than typical head and neck squamous cell carcinomas, certain patients can be strongly suspected as having HPV-related tumours. In particular, nonkeratinizing histologic morphology, present in 50-60% of oropharyngeal squamous cell carcinoma, correlates very well with positive HPV status.[2] However, prediction of HPV status by such surrogate marker and clinical grounds is less reliable than direct p16/HPV testing.[3] Thus, when determining optimal treatment for patients, local practices must carefully exercise their own judgment and decide on what grounds they can classify patients as (likely) HPV-related in their populations.

It is now well established that HPV plays a pathogenic role in a large subset of oropharyngeal squamous cell carcinomas.[4,5] A smaller subset of nasopharyngeal carcinomas is related to transcriptionally active high risk HPV.

HPV-positive oropharyngeal carcinoma represents a unique squamous cell carcinoma type with proven more favourable prognosis than for HPV-negative tumours.[6] Staging of these patients is now different than for HPV-negative tumours and treatment differences are emerging.

There are many methods for testing HPV status with p16 immunohistochemistry emerging as a simple, thoroughly validated prognostic marker in oropharyngeal squamous cell carcinoma (SCC).[7] The most commonly used criterion for positivity as a surrogate marker moderate to intense nuclear and cytoplasmic staining in 70% or more of the tumour cells, which is the recommended cutoff for these guidelines,[8] with the caveat that the correlation with HPV status is not 100%.[9,10] The combination of p16 immunohistochemistry with nonkeratinizing morphology is very strongly associated with transcriptionally-active high risk HPV in the oropharynx.[2] HPV specific tests include in situ hybridization for DNA, PCR for HPV-DNA, RT-PCR for HPV-mRNA, and in situ hybridization for mRNA. There is no consensus on the best methodology for HPV testing but the World Health Organization (WHO), AJCC, UICC, and a College of American Pathologists Expert Panel have all recommended p16 immunohistochemistry. Additional HPV-specific testing is performed at the discretion of the pathologist.

The new WHO Blue Book terms squamous cell carcinomas of the oropharynx simply as HPV-positive or HPV-negative.[11,12] However, they specifically note that p16 immunohistochemistry alone (with

appropriate criteria for a positive versus negative test) is a suitable surrogate marker. They recommend the terminology HPV-positive even if only p16 is performed.

EBV is associated with the nonkeratinizing types of nasopharyngeal carcinomas in the vast majority of patients. The most reliable detection method for EBV is in situ hybridization for EBV encoded early RNA (EBER) present in cells latently infected by EBV, and is recommended because it is a modestly strong favourable prognostic marker and because it is confirmation of the tumour having a nasopharyngeal association.[13] A subset of patients with nasopharyngeal carcinoma are related to transcriptionally-active high risk HPV.[14-16] Most of these tumours are described as nonkeratinizing differentiated using the WHO terminology. They are EBV (EBER) negative and p16 positive. Testing for HPV/p16 in EBV negative nonkeratinizing carcinomas, however, is at the discretion of the local practice. It may be indicated in routine clinical practice to help alert the clinician that this may be an oropharyngeal primary tumour that is secondarily involving the nasopharynx and not because the HPV is of proven prognostic benefit in such tumours.[14-16]

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