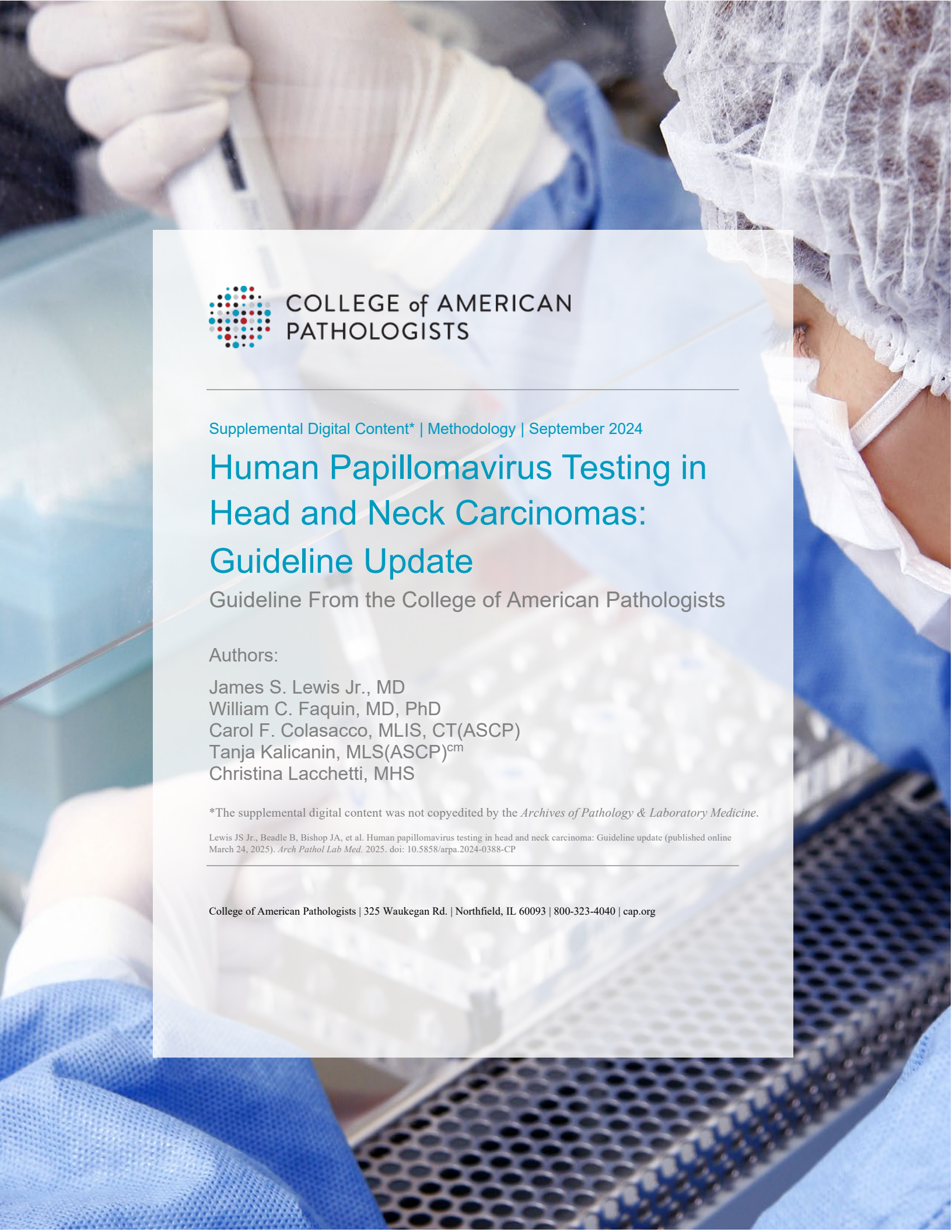

COLLEGE of AMERICAN PATHOLOGISTS

Supplemental Digital Content* | Methodology | September 2024

Human Papillomavirus Testing in Head and Neck Carcinomas: Guideline Update

Guideline From the College of American Pathologists

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GUIDELINE DEVELOPMENT METHODS

Panel Composition

The College of American Pathologists (CAP) convened an expert panel (EP) and advisory panel (AP) consisting of members with experience and expertise in head and neck pathology. Members included practicing pathologists and experts in surgical, medical, and radiation oncology, and a contracted methodologist. The CAP approved the appointment of the project co-chairs and panel members.

The roles of each panel are described in the Evidence-based Guideline Development Methodology Manual (Methodology Manual).

Conflict of Interest (COI) Policy

Prior to acceptance on the expert or advisory panel, potential members completed the CAP conflict of interest (COI) disclosure process, whose policy and form require disclosure of material financial interest in, or potential for benefit of significant value from, the guideline's development or its recommendations 24 months prior through the time of publication. The potential members completed the COI disclosure form, listing any relationship that could be interpreted as constituting an actual, potential, or apparent conflict. A complete description of the COI policy is available in the online Methodology Manual.

Everyone was required to disclose conflicts prior to beginning and continuously throughout the project's timeline. EP members' disclosed conflicts are listed in the appendix of the manuscript. The CAP provided funding for the administration of the project; no industry funds were used in the development of the guideline. All panel members volunteered their time and were not compensated for their involvement, except for the contracted methodologist.

Project Scope and Outcomes of Interest

The EP approved the following scope to assess evidence published since the release of the original guideline[1] and to update evidence-based recommendations for human papillomavirus (HPV) testing in head and neck carcinomas. The outcomes of interest were reviewed and finalized prior to the literature review.

Outcomes of Interest:

Overall Survival (OS)

Disease related

- Disease-specific survival
- Disease-free survival (DFS)
- Progression-free survival (PFS)
- Recurrence-free survival (RFS)

Diagnostic Test Characteristics

- Sensitivity
- Specificity
- Positive Predictive Value (PPV)
- Negative Predictive Value (NPV)
- Concordance
- Observer variability

Systematic Evidence Review

The objective of the systematic evidence review was to identify articles that provided data to inform the recommendations. If of sufficient quality, findings from this review would provide an evidence-base to support the recommendations of the guideline. The scope of the systematic evidence review and the key questions (KQs) with the Population, Intervention, Comparator, Outcome(s) (PICO) elements were established by the EP in consultation with the methodologist prior to beginning the literature search.

Detailed key questions including the PICO is included in Supplemental Table 1.

Search and Selection

Detailed literature searches were constructed using controlled vocabulary and keywords for concepts derived from the PICO elements defined at the onset of the project based upon the key questions. Initial searches were run on July 6, 2021, in Ovid MEDLINE (Wolters Kluwer Health, Philadelphia, PA) and Embase.com (Elsevier, Amsterdam, Netherlands), and rerun on August 2, 2023 to capture literature published since initial searches were run. Searches were also completed in Cochrane Library (John Wiley & sons, Inc., Hoboken, NJ), relevant organization's websites, guideline repositories (eg, Guidelines International Network Library, ECRI Guidelines Trust, Trip Medical Database), and clinical trial registries to identify unindexed (grey) literature. All search results were deduplicated using reference management software following published methods.[2] The literature search strategies and Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) diagram[3] are included as Supplemental Figures 1 and 2. The detailed search strings for Ovid MEDLINE and Embase.com are included as Supplemental Figure 2.

Selection at all levels was based on the predetermined inclusion/exclusion criteria which are detailed in the manuscript.

Data Extraction & Management

The data elements from an included article/document were extracted by one reviewer into standard data formats and tables developed using the systematic review database software, DistillerSR (Evidence Partners Inc., Ottawa, Canada); a second reviewer confirmed accuracy and completeness. Any discrepancies in data extraction were resolved by discussion between the co-chairs and the methodologist. A bibliographic database was established in EndNote (Thomson Reuters, Carlsbad, CA) to track all literature identified and reviewed during the study.

Assessing Quality and Risk of Bias

An assessment of the quality of the evidence was performed for all retained studies following application of the inclusion and exclusion criteria. Using this method, studies deemed to be of low quality would not be excluded from the systematic review but would be retained, and their methodological strengths and weaknesses discussed where relevant. To define an overall study quality rating for each included study, validated study-type specific tools were used to assess the risk of bias, plus additional important quality features were extracted. Specific details for each study type are outlined below.

- Systematic Reviews (SRs) and Meta-analyses questions were assessed as per the Assessing the Methodological Quality of Systematic Reviews (AMSTAR) 8 tool.[4]
- All observational studies were assessed using the Risk of Bias in Non-randomized Studies of Intervention (ROBINs-I) tool.[5]

In the following sections, the quantity of the evidence as determined by the number of studies that met our inclusion criteria and were retained, the evidence type as determined by study design, the quality of that evidence as determined by the quality assessment, and its consistency are all reported, both as individual studies and in totality, statement by statement. Definitions of the certainty of evidence is presented in Supplemental Table 2.

A total of 235 studies comprised the final body of studies included in the SER. Supplemental Figure 1 displays the results of the literature review. All articles were available as discussion or background references. All members of the EP participated in developing draft recommendations, reviewing open comment feedback, finalizing and approving the final recommendations, and writing/editing of the manuscript.

For further explanation of the Quality Assessment and the ROB assessment, refer to the Evidence-based Guideline Methodology Manual.

Evidence-to-Decision Framework

In addition to the panel discussion of the net benefits and harms for each guideline statement, the EP members rated each recommendation using the GRADE evidence-to-decision framework. This provides a systematic mechanism to document panel members' judgement for each of the recommendations. [6]

Open Comment Period and Organizational Review

A public, open access comment period was held from August 14 through September 1, 2023, on the CAP Web site for any interested stakeholder to provide feedback on the draft recommendations. Sixteen draft statements, two demographic questions, and three questions to assess feasibility were posted for peer review. An announcement was sent to the following societies deemed to have interest.

Medical societies:

- Association for Molecular Pathology (AMP)
- American Society for Clinical Pathology (ASCP)
- American Society for Investigative Pathology (ASIP)
- American Society of Cytopathology (ASC)
- American Society for Clinical Oncology (ASCO)
- American Head and Neck Society (AHNS)
- American Joint Committee on Cancer (AJCC)
- American College of Radiology (ACR)
- American Radium Society (ARS)
- American Society for Radiation Oncology (ASTRO)
- American Cancer Society (ACS)
- American Dental Association (ADA)
- American Academy of Oral and Maxilofacial Pathology (AAOMP)
- American Association for Clinical Chemistry (AACC)
- Australia New Zealand Head and Neck Cancer Society (ANZHNCS)
- American Academy of Otolaryngology–Head and Neck Surgery (AAO-HNS)
- American Board of Otolaryngology (ABO)
- American Broncho-Esophagological Association
- Association of Community Cancer Centers (ACCC)
- Association of Directors of Anatomic and Surgical Pathology (ADASP)
- Association of Pathology Chairs (APC)
- Canadian Association of Pathologists (CAP-APC)
- European Society for Medical Oncology (ESMO)
- European Society of Pathology (ESP)
- International Academy of Pathology (IAP)
- National Comprehensive Cancer Network (NCCN)
- North American Society of Head and Neck Pathology (NASHNP)
- Papanicolaou Society of Cytopathology (PSC)
- Quality Initiative in Interpretive Pathology (QIIP) Canadian Partnership Against Cancer

- Radiological Society of North America (RSNA)
- Royal College of Pathologists of Australasia (RCPA) - Structured Pathology Reporting Committee for head and neck
- Sociedade Brasileira de Patologia (Brazilian Society of Pathology)
- Society to Improve Diagnoses in Medicine (SIDM)
- The American Laryngological, Rhinological and Otological Society, Inc. (The Triological Society)
- United States & Canadian Academy of Pathology (USCAP)
- World Health Organization (WHO)

Patient advocacy groups:

- American Cancer Society
- Cancer Leadership Council
- Cancer Research and Prevention Foundation (formerly Prevent Cancer Foundation)
- Global Resource for Advancing Cancer Education
- Partnership Against Cancer American Cancer Society
- UICC Global Cancer Control Community (Union for International Cancer Control)
- Head and Neck Cancer Alliance

Government and other stakeholders:

- US Food and Drug Administration (FDA)
- Centers for Medicare & Medicaid Services (CMS)
- Centers for Disease Control and Prevention (CDC)
- Veteran's Affairs (VA) and Department of Defense (DOD)
- European Medicines Agency (EMEA)

"Agree" and "Disagree" responses were captured for every proposed recommendation and good practice statement. The EP reviewed all the comments. Resolution of all changes was obtained by majority consensus of the panel using nominal group technique (discussions on teleconferences) amongst the panel members. The final recommendations were approved by the EP. Neither formal cost analysis nor cost effectiveness models were performed.

Organizational review was instituted to review and approve the guideline. An independent review panel (IRP) representing the Council on Scientific Affairs was assembled to review and approve the guideline for the CAP. The IRP was masked to the expert panel and vetted through the COI process.

Dissemination Plans

The CAP hosts a resource page which includes a link to the manuscript and supplement; a summary of the recommendations, algorithm, a teaching PowerPoint (Microsoft Corporation, Redmond, WA), and a frequently asked question (FAQ) document, with other additional tools such as webinar recordings as applicable. The guideline is promoted and presented at various society meetings and distributed to the societies listed in the peer review.

Recommendation Statements

For each guideline statement designated a recommendation, a summary of the studies, and benefits and harms are included below. Supplemental Tables 3 and 4 are the risk of bias/quality assessment for systematic reviews/meta-analyses and observational studies respectively.

Statement 1. Pathologists should perform high-risk human papillomavirus (HR-HPV) testing on all patients with newly diagnosed oropharyngeal squamous cell carcinoma (OPSCC), including all histologic subtypes. This testing may be performed on the primary

tumor or on a regional lymph node metastasis when the clinical findings are consistent with an oropharyngeal primary.

Statement 1 is supported by a total of 114 studies comprising two meta-analyses[7,8] and 112 observational studies.[9-120] Four of these observational studies were assessed to be at low risk of bias,[13,16,69,120] 106 at a moderate risk,[9-11,14,15,17-68,70-80,82-119] and two at a high risk of bias.[12,81] High risk of bias elements included confound and selection.[12,81] Although the aggregate risk of bias across the evidence base was serious, the certainty of evidence was upgraded to high for all outcomes based on a large body of evidence showing strong and consistent clinical benefits in patients with HPV-associated versus HPV-independent OPSCC. See Supplemental Table 5 for the certainty of evidence assessment.

The routine use of HR-HPV status in patients with OPSCC is unique amongst cancer care, being established for use in patient care in the oropharynx more than for any other cancer type in the body. The laboratory tests for HR-HPV status, including surrogate marker p16 immunohistochemistry (IHC), are available in most pathology laboratories around the world, including, increasingly, high-risk HPV ribonucleic acid (RNA) in situ hybridization (ISH). The HPV-specific tests are also available from larger reference laboratories, so could be sent out as needed. As such, the capability and expertise to perform the testing is largely already in place. If an institution had to set up one or more of these tests as a laboratory developed test, the risks would include incurring the expenses and validating/verifying performance prior to offering the test and as an ongoing function. As long as the proper tests are used and are interpreted correctly, there are no expected harms for testing patients, as the entire medical community now understands and utilizes HR-HPV status properly. The benefits of testing are proper classification, staging, prognostication, and treatment, and the ability for patients to go on any of the numerous ongoing prospective clinical trials seeking to optimize care and outcomes for patients with HPV-associated OPSCC.

Statement 2. For oropharyngeal tissue specimens (ie, non-cytology), including regional lymph nodes with metastatic squamous cell carcinoma (SCC) and clinical findings consistent with an oropharyngeal primary, pathologists should perform HR-HPV testing by surrogate marker p16 IHC. In certain scenarios HPV-specific testing should be performed: a) in geographic regions with a low prevalence of HR-HPV associated OPSCC b) when p16 immunostaining is equivocal (50-70% staining or when staining is extensive but weak) c) when there is a discrepancy between p16 staining and morphology, d) for large, multisite tumors overlapping the oropharynx, e) when specimens are from a non-tonsillar, non-base of tongue oropharyngeal site, and f) when required by clinical trials. Additional HPV-specific testing may be done at the discretion of the pathologist and/or treating clinician.

Statement 2 is supported by a total of 74 studies, including three systematic reviews[7,121,122] and 71 observational studies.[12,14,17,23,26-29,32-36,39,43-45,47,49-52,55,56,59-61,63,66,74,76,77,79,84,86,88,90-95,97-99,101-103,107,111,112,114,120,123-140] The three systematic reviews[7,121,122] and 64 observational studies[12,14,17,23,26-29,32-36,39,43-45,47,49-52,55,56,59-61,63,66,74,76,77,79,84,86,88,90-95,97-99,101-103,107,111,112,114,120,123,127,128,130-132,134,137-140] considered clinical outcomes, and 35 observational studies[12,17,27,28,34,35,43,44,49,55,56,60,77,90-93,98,99,102,103,107,111,124-126,128,129,132-137,139] considered testing outcomes. The included systematic reviews were assessed as high[122] and intermediate[7,121] quality. None of the reviews reported on using publication status as an inclusion criterion or provided a list of both included and excluded studies. The observational studies were all assessed as being at a moderate risk of bias, with the exception of two which were deemed to be at a high risk of bias[12,125] based on risks in confounding, selection, classification, performance and detection domains. The aggregate risk of bias of the evidence base was serious and the evidence was upgraded to high based on a large body of evidence showing strong and consistent

clinical benefits and testing outcomes in patients with p16-positive versus p16-negative OPSCC. See Supplemental Table 6 for the certainty of evidence assessment.

HR-HPV status can be assessed directly, with HPV-specific tests, or indirectly, using morphology and surrogate marker p16 IHC. There is low interobserver variability in the performance and interpretation of these tests. [141] Given the utility and performance of p16 IHC, with a 70% nuclear and cytoplasmic staining cutoff, it is still a very practical and useful test with very high negative predictive value. However, we recognize that there are many scenarios where it is inadequate as a standalone test in OPSCC patients. Thus, there are six situations in which we recommend HPV-specific testing for patients with p16 positive tumor, and the simple way to think about it is "if the scenario or specimen of a p16 positive or negative tumor does not line up perfectly with an HPV-associated or -independent tumor, HPV-specific testing should be undertaken".

The major risk of this approach is that some patients with p16 positive tumors, but who are actually HPV-independent, will be wrongly assumed HPV-associated, resulting in treatment that may not be sufficient or appropriate for them. With all of the various "caveats" we lay out, this should "fence in" p16 immunostaining to help avoid this from occurring with regularity. The benefit is that almost every laboratory has access to p16 immunostaining in house and the various clones, tests, and interpretation show great consistency and reproducibility with the stringent 70% nuclear and cytoplasmic staining criteria are applied. Having to do HPV-specific testing in a significant subset of patients will result in costs for doing those tests in house or for sending them to a reference laboratory and in delays in reporting of final HPV status.

Statement 3. For tissue specimens, when p16 IHC is indicated, pathologists should report it as positive (and as a surrogate for HR-HPV) when there is at least 70% nuclear and cytoplasmic expression with at least moderate to strong intensity.

Statement 3 is supported by 27 observational studies. [15,25,27,34,45,49,56,77,90-92,98,125,132,133,137,142-152] The studies were assessed to be at low, [147,151] moderate, [15,25,27,34,45,49,56,77,90-92,98,132,133,137,142-146,148-150,152] or high[125] risk of bias based on retrospective acquisition of samples in all studies plus individual moderate risk of confounding, [15,25,27,34,45,49,56,77,90-92,98,125,132,133,137,145,148,149] selection, [15,25,27,34,45,49,56,90-92,98,125,132,133,137,142,148-150,152] performance, [125] and detection[15,25,27,34,45,49,56,77,91,92,98,125,132,133,137,145,146,148-150,152] bias. The aggregate risk of bias of the evidence base was serious and the evidence was upgraded to high based on a strong association and consistency between criteria for positive test and testing outcomes. See Supplemental Table 7 for the certainty of evidence assessment.

This guideline statement, same as in the initial evidence-based guideline, impacts all p16 IHC testing and is based on a very large amount of data from the initial 2018 guideline and from this literature update. The data strongly shows the performance and reproducibility of this assessment as a surrogate of transcriptionally-active high-risk HPV. There are no special resources required to implement this guideline statement, as almost everyone already has access to performing p16 in house. There are no anticipated risks to interpreting p16 in this way and the benefits are the optimal performance of p16 immunostaining as a surrogate of high-risk HPV when interpreted in this manner.

Statement 4. Pathologists should routinely perform HR-HPV testing on sinonasal SCC.

Statement 4 is supported by eight observational studies. [148,153-159] All included studies were limited by a risk of selection and classification bias, plus individual studies were further limited by risk of bias in attrition. [154,157] The aggregate risk of bias across the entire evidence base was serious and the overall certainty of evidence was moderate. See Supplemental Table 8 for the certainty of evidence assessment.

Because sinonasal SCC is much less common than OPSCC, and because rates of HPV-positivity are lower, it has taken a much longer time for quality data to accumulate. Studies are smaller and all retrospective but almost all show the same pattern, that HPV-associated SCC patients have improved disease-free and overall survival, independent of other variables. [153,158,160-163] The panel feels that the data is sufficient to justify routine testing and that it must include HPV-specific testing, with the clinical use of this information to follow. The potential harms are increased test application costs and pathologist time for the patients, which may not change clinical management for some time. It is also a possible harm that surgeons and oncologists may use this additional information to alter patient care outside of the standard of care in treating these patients with the potential for harm (under or overtreatment). Potential benefits are that patients will have better prognosis education and, at the margins of patient care decisions, the information be used to more specifically treat them and manage their follow up care.

Statement 5. When testing a sinonasal SCC specimen for HR-HPV, pathologists should test directly for transcriptionally-active HR-HPV (RNA in situ hybridization [ISH]); positivity for the surrogate marker p16 IHC may be used to screen tumors for confirmatory HPV-specific testing.

Statement 5 is supported by 6 retrospective observational studies. [148,153,155,156,158,159] All studies were assessed to have a moderate risk of bias, based on risk of bias in selection and classification. The aggregate risk of bias for the evidence base was serious and the overall certainty of the evidence was moderate. See Supplemental Table 8 for the certainty of evidence assessment.

Because the rates of HPV-association in sinonasal tumors are lower than OPSCC, being approximately 25 to 30% in the United States, p16 retains high negative predictive value but much lower positive predictive value. [163,164] Thus, HPV-specific testing must be utilized (either for all p16 positive patients or, if RNA in situ hybridization is used as a standalone test) to confirm patients' tumors as HPV-associated. The potential harm is that p16 IHC and HPV-specific tests are "additional" work and have cost and expense of application in routine practice. HPV-specific testing, particularly HPV RNA ISH, is not available to every laboratory so it would have to be sent out to a reference laboratory, increasing time and costs. The benefits of applying HPV-specific testing across the board is that patients will not be misclassified based on p16 alone.

Statement 6. Pathologists should routinely perform HR-HPV testing on patients with metastatic SCC of unknown primary in a cervical lymph node.

Statement 6 is supported by a total of eight studies comprising one systematic review/meta-analysis[165] and seven observational studies. [104,145,166-170] The systematic review received a score of 9 out of a possible 11 points on the AMSTAR. Observational studies were assessed to be at a low[166,170] and intermediate[104,145,167-169] risk of bias based on risk of bias in confounding, [145,168] selection, [168,169] classification, [104,145,169] and detection[104,145,167-169] domains. Although the aggregate risk of bias across the evidence base was serious, the evidence was upgraded to high based on evidence from meta-analyses showing strong and consistent clinical benefits in HPV-associated versus HPV-independent patients with cancer of unknown primary (CUP). See Supplemental Table 9 for the certainty of evidence.

Level II/III metastases, in the absence of an obvious primary site, are already considered to be likely oropharyngeal without HPV testing, but HR-HPV testing further solidifies this so that patient care can proceed appropriately. High-risk HPV positivity has been shown to be prognostic in patients with metastatic SCC of unknown primary, and the 8th edition American Joint Committee on Cancer (AJCC) [171] staging considers HPV-associated metastatic SCC in cervical lymph nodes where no primary is identified as T0 oropharyngeal SCC.

This recommendation is slightly different than from 2018 in that we recommend testing of metastatic SCC of unknown primary wherever it occurs in the neck, not just levels II and III, but as in recommendation 7, we recommend the consistent use of HPV-specific testing. The potential harms of this guideline recommendation are increased workload and expense of the testing, potentially without substantial clinical benefit, something particularly affecting smaller laboratories and lower resource parts of the world. Sending testing to reference laboratories increases expense and turnaround times. The benefits of testing are the more accurate knowledge of a patient's tumor and its etiology, which helps provide appropriate staging, treatment (including search in the oropharynx for the primary tumor), and eligibility for clinical trials.

Statement 7. For tissue specimens (ie, non-cytology) from patients presenting with metastatic SCC of unknown primary in a cervical lymph node, pathologists should perform HPV-specific testing or surrogate marker p16 IHC, followed by HPV-specific testing for p16 positive tumors. An explanatory note on the significance of a positive HPV result is recommended.

Statement 7 is supported by one observational study that evaluated the diagnostic test characteristics of non-cytology tissue specimens from patients presenting with metastatic SCC of unknown primary in a cervical lymph node. [145] The risk of bias was assessed to be serious, and the overall certainty of evidence was moderate. See Supplemental Table 10 for the certainty of evidence.

HPV-specific testing is important in neck surgical pathology specimens. Although the initial 2018 guideline used an approach of "p16+appropriate location (II or III) in the neck+nonkeratinizing morphology" to help diagnosing an HPV-associated metastatic SCC of unknown primary, the American Society of Clinical Oncology (ASCO) panel disagreed, recommending HPV-specific testing for all metastatic SCC of unknown primary patients. [145] The revised recommendation now does just that, taking away other criteria. The harms of this approach are that laboratories must have access to doing the appropriate HPV-specific test(s) and patients and laboratories will bear the cost. If tests must be sent to a reference laboratory, additional costs and delays in diagnosis will occur. The benefit of this approach is that all patients with metastatic SCC of unknown primary will have an accurate determination of HR-HPV status and be correctly classified. Patients with metastatic skin or lung SCC to the neck, which are frequently p16 positive, will not be misclassified as possible oropharyngeal SCC and needless surgery can be avoided. The additional benefit here is that pathologists need not know the location of the involved lymph node in order to know if to test or what test to perform.

Statement 8. Pathologists should not routinely perform HR-HPV testing on patients with primary oral cavity, laryngeal, nasopharyngeal, or hypopharyngeal SCCs of the head and neck for prognostic purposes.
Note: **HR-HPV testing in nasopharyngeal SCCs can be used at the discretion of the pathologist and/or treating clinician.**

Statement 8 was informed by a total of three systematic reviews[172-174] and 45 observational studies. [140,144,146,150,152,159,175-213] The three systematic reviews[172-174] and 14 observational studies[146,150,152,176,181,182,187,192-196,211,212] evaluated the performance of HPV testing and 43 investigated clinical outcomes[140,144,146,150,152,159,175-180,182-210,212,213] in non-oropharyngeal squamous cell carcinomas. The included systematic reviews were assessed as high quality, each scoring a nine out of a total 11 points. The reviews lost points for not using grey literature and failing to provide a list of both included and excluded studies. The observational studies were assessed to be at a low[181,183-185,187,191,192,200,203,205,206,208] and moderate[140,144,146,150,152,159,175-180,182,186,188-190,193-199,201,202,204,207,209-213] risk of bias. The aggregate risk of bias across these studies was serious and, although studies vary considerably based on non-oropharyngeal site, sample size, and specimen type, studies are remarkably consistent in lack of statistically significant clinical and testing

outcomes. As such, certainty of evidence was upgraded to high. See Supplemental Table 11 for the certainty of evidence.

The initial 2018 guideline[1] systematic review did not support routine testing for HR-HPV in non-OP primary tumors of the head and neck, and this update reviewed a substantial amount of new data which supports this conclusion, with the exception of the sinonasal tract. For patients with SCC of the oral cavity, larynx, nasopharynx, and hypopharynx, there is not a clear prognostic benefit for HR-HPV positivity. A significant amount of this testing is occurring in these patients, based on practice experience, but this is not recommended. The benefits of this are in saving money, time, and laboratory resources, in addition to savings for patients. Testing in these patients is potentially misleading and could result in inadequate treatment based on the assumption that the occasional HPV-associated patients have better treatment response and prognosis. The benefits of not testing are to avoid risk of this type of harm. The potential harm is that a subset of HPV-associated patients, particularly those with nasopharyngeal tumors, may have prognostically favorable, more treatment responsive tumors that could be treated with less morbidity. Better future research is needed.

Statement 10. Pathologists should perform HR-HPV testing on head and neck fine needle aspirations (FNA) of nodal SCC samples from all patients with: (a) clinical findings of an oropharyngeal or sinonasal primary or (b) metastatic SCC of unknown primary.

Statement 10 was informed by two observational studies[167,169] which investigated overall and disease-free survival. The included studies were assessed to be at a low[167] and moderate[169] risk of bias, based on retrospective acquisition of samples plus moderate risk of confounding, classification, and detection bias. Although the aggregate risk of bias across the evidence base was serious, the evidence was upgraded based on a strong association in clinical outcomes in FNAs of nodal SCC samples from all patients, with clinical findings of an oropharyngeal or sinonasal primary, or with metastatic SCC of unknown primary.

Since HPV-associated head and neck squamous cell carcinomas are commonly first detected by FNA sampling, there is a benefit of performing HR-HPV testing on FNA specimens to establish diagnosis, tumor staging, therapy, and patient prognosis. Most laboratories receiving head and neck FNAs have the resources needed to provide HR-HPV testing on these specimens because they are already commonly done on cervical cytology specimens. If this guideline recommendation were to be implemented, proper validation for head and neck FNA specimens would be required. The resources to implement this recommendation are minimal since it leverages equipment in the laboratory and expertise of the laboratory personnel.

Statement 11. For FNA specimens, pathologists should perform HPV-specific testing.
Note: **In selected circumstances p16 IHC can be performed instead of HPV-specific testing. If the result of HR-HPV testing on the FNA sample is negative, testing should be performed on tissue if it becomes available.**

Statement 11 was informed by 30 observational studies[142,167,214-241] that evaluated the performance of HPV testing in FNA specimens. The included studies were assessed as low,[167,232,241] intermediate,[142,214-227,229-231,233-237,239,240] and high[228,238] risk of bias. The aggregate risk of bias of the evidence base was serious but the evidence was upgraded based on a strong association and consistency in HR-HPV testing outcomes in FNAs of nodal SCC samples from all patients with primary oropharyngeal or sinonasal tumors, or with metastatic SCC of unknown primary. See Supplemental Table 12 for the certainty of evidence for Statements 10 and 11.

The performance of p16 IHC on cell blocks has proven to be poor.[217,218,220] Given the important clinical and prognostic implications of classifying a metastatic carcinoma of unknown primary as

either HPV-associated or HPV-independent, HPV-specific testing is recommended by the panel for FNA specimens. The benefit of this guideline statement is that the specificity of the test result will be high while also preserving the sensitivity of the test. Although HPV-specific testing may, in some cases, be modestly more expensive and technically demanding than p16 IHC, most laboratories will have access to HPV testing methodologies already in use for cervical cytology specimens. In addition, HPV-specific testing on cell block material can use the same HPV-specific testing used for surgical pathology specimens. The statement provides flexibility for the choice of HPV-specific test used. For laboratories where HPV-specific testing is not possibly, p16 IHC is accepted, but repeat testing should be performed on surgical pathology specimens if the cell block testing is negative.

Statement 12. For HPV specific testing, pathologists should utilize tests that exhibit optimal performance characteristics, such as RNA-ISH or deoxyribonucleic (DNA) polymerase chain reaction (PCR); and have adequate coverage of non-HPV16 high-risk types. DNA-ISH is not recommended.

Statement 12 was informed by three systematic reviews[121,173,174] and 17 observational studies[98,122,127,134,138,140,157,179,200,208,242-248] that evaluated the performance of HPV-specific testing and clinical outcomes, including OS, [98,121,122,127,134,138,140,157,179,200,208,242,244-248] DFS, [121,122,127,157,173,179,242] and PFS or DSS. [98,138,140,244] The included systematic reviews were assessed as high quality, scoring eight[121] or nine[173,174] out of a total 11 points. None of the systematic reviews reported on using grey literature nor did they list both included and excluded studies. The observational studies were assessed to be at a low[179,200,208,242,243,248] and moderate[98,121,127,134,138,140,157,244-247] risk of bias. The aggregate risk of bias of the evidence base was serious and the evidence was upgraded based on evidence from systematic reviews and observational studies showing a large and consistent estimate of the magnitude of effect. See Supplemental Table 13 for the certainty of evidence.

HPV-specific testing methodologies with optimal performance characteristics including RNA-ISH and DNA PCR are readily available to most laboratories either as an in-house test or as a send-out. The benefit of using such tests covering a broad range of HR-HPV types is that it helps to avoid false negative results which could negatively affect both patient management and prognostication. The cost of covering a broad range of HR-HPV types is low compared to the consequences of a false negative test result.

Supplemental Table 1: Key Questions (KQs) and Population, Intervention, Comparator, Outcomes (PICO) Elements

<table>
<tr><td colspan="3">KQ1a.
In patients with oropharyngeal squamous cell carcinoma (OPSCC), are clinical outcomes improved in HPV-associated carcinoma (RNA-ISH, RT-PCR, p16 + RNA-ISH, DNA PCR, DNA-ISH, p16 IHC alone) compared to HPV-independent carcinoma?</td></tr>
<tr><td colspan="3">Population</td></tr>
<tr><td colspan="3">Patients with primary OPSCC
• Non-tonsillar/non-Waldeyer ring
  ○ Uvula, soft palate, tonsillar pillar, posterior pharyngeal wall
• Waldeyer ring
  ○ Base of tongue, palatine tonsil</td></tr>
<tr><td>Intervention</td><td>Comparator</td><td>Outcomes</td></tr>
<tr><td>HPV-associated carcinoma</td><td>HPV-independent</td><td>Overall survival
Disease related</td></tr>
<tr><td colspan="3">KQ1b.</td></tr>
</table>

In patients with non-oropharyngeal squamous cell carcinoma (non-OPSCC), are clinical outcomes improved in HPV-associated carcinoma (RNA-ISH, RT-PCR, p16 + RNA-ISH, DNA PCR, DNA-ISH) compared to HPV-independent carcinoma?		
Population		
Patients with primary non-OPSCC • Sinonasal • Nasopharyngeal • Laryngeal/hypopharyngeal • Oral cavity		
Intervention	**Comparator**	**Outcomes**
HPV-associated carcinoma	HPV-independent	Overall survival Disease related
KQ1c. In patients with metastatic squamous carcinoma of unknown primary in the neck, are clinical outcomes improved in HPV-associated carcinoma (RNA-ISH, RT-PCR, p16 + RNA-ISH, DNA PCR, DNA-ISH) compared to HPV-independent carcinoma?		
Population		
Patients with metastatic squamous carcinoma of unknown primary in the neck		
Intervention	**Comparator**	**Outcomes**
HPV-associated carcinoma	HPV-independent	Overall survival Disease related
KQ1d. In patients with non-squamous head and neck carcinoma, are clinical outcomes improved in HPV-associated carcinoma (RNA-ISH, RT-PCR, p16 + RNA-ISH, RT-PCR, DNA PCR, DNA-ISH) compared to HPV-independent carcinoma?		
Population		
Patients with non-squamous head and neck carcinoma		
Intervention	**Comparator**	**Outcomes**
HPV-associated carcinoma	HPV-independent	Overall survival Disease related
KQ2a. In patients with newly diagnosed OPSCC including multi-site overlapping tumors, non-OPSCC, non-squamous head and neck carcinoma, and cervical nodal metastatic carcinomas of unknown and/or known primary do relevant outcomes differ based on the type(s) of HR-HPV testing performed?		
Population		
Patients with newly diagnosed OPSCC including multi-site overlapping tumors, non-OPSCC, non-squamous head and neck carcinoma and cervical nodal metastatic carcinomas of unknown and/or known primary		
Intervention	**Comparator**	**Outcomes**
RNA-ISH IHC p16 plus one confirmatory test (PCR for HPV DNA, ISH for HPV DNA, ISH for E6/E7 RNA)	IHC p16 alone	Overall survival Disease related Diagnostic test characteristics
KQ2b. What testing approach is best for fine needle aspiration (FNA) specimens compared to tissue-based HR-HPV testing?		
Population		
Patients with newly diagnosed OPSCC including multi-site overlapping tumors, non-OPSCC, non-squamous head and neck carcinoma and cervical nodal metastatic carcinomas of unknown and/or known primary		
Intervention	**Comparator**	**Outcomes**

FNA FNA testing with: • IHC p16 alone • ISH alone • Liquid-based tests • Specific combination of tests	Biopsy/tissue-based	Concordance Test correlation
KQ3. Does performance of specific tests or testing algorithms for HR-HPV differ based on specimen characteristics such as size, age, type of fixation, time-to-fixation and length of tissue fixation, the criteria/definition for a "positive" p16 IHC or ISH test result?		
Population		
HR-HPV tests		
Intervention	**Comparator**	**Outcomes**
• Specimen size, age • Percent neoplastic cellularity • Tissue fixation • Length of tissue fixation • Time-to-fixation • Antibody • Probes • Testing conditions and criteria	Acceptable "standard"	Test Correlation Concordance
KQ4. In patients with OPSCC, can HR-HPV status be used to determine if a cancer is a recurrence versus new primary?		
Population		
Patients with primary OPSCC		
Intervention	**Comparator**	**Outcomes**
HR-HPV positive	Known new primary tumor	-

Abbreviations: DNA, deoxyribonucleic acid; HPV, human papillomavirus; HR-HPV, high risk human papillomavirus; IHC, immunohistochemistry; ISH, in situ hybridization; KQ, key question; PCR, polymerase chain reaction; RNA, ribonucleic acid; RT-PCR, real time polymerase chain reaction

Supplemental Table 2: Grades for Certainty of Evidence[a]

Designation	Description
High	There is high confidence that available evidence reflects true effect. Further research is very unlikely to change the confidence in the estimate of effect.
Moderate	There is moderate confidence that available evidence reflects true effect. Further research is likely to have an important impact on the confidence in estimate of effect and may change the estimate.
Low	There is limited confidence in the estimate of effect. The true effect may be substantially different from the estimate of the effect.
Very Low	There is very little confidence in the estimate of effect. The true effect is likely to be substantially different from the estimate of effect. Any estimate of effect is very uncertain.

[a]Data derived from Grading of Recommendations, Assessment, Development, and Evaluation (GRADE) Working Group materials. [249]

Supplemental Table 3. Risk of Bias Assessment for Systematic Reviews/Meta-analyses

AMSTAR	Sedghizadeh et al, [7] 2016	Albers et al, [121] 2017	Ahmadi et al, [122] 2019	Ren et al, [165] 2019	Tham et al, [172] 2018	Wang et al, [173] 2020	Ahmadi et al, [174] 2018	Lassen et al, [8] 2018
A priori design	√	√	√	√	√	√	√	√
Duplicate study selection & data extraction	√	√	√	√	√	√	√	x
Comprehensive lit search performed	√	√	√	√	√	√	√	√
Grey lit used	x	x	x	√	x	x	x	√
List included & excluded studies	x	x	x	x	x	x	x	x
Characteristics of included studies provided	√	√	√	√	√	√	√	√
Quality assessed & documented	√	x	√	√	√	√	√	x
Quality used appropriately for conclusion	√	√	√	√	√	√	√	x
Methods to combine used appropriately	√	√	√	√	√	√	√	√
Publication bias assessed	x	√	√	x	√	√	√	x
COI	√	√	√	√	√	√	√	√
AMSTAR SCORE	**8**	**8**	**9**	**10**	**9**	**9**	**9**	**6**

Abbreviations: AMSTAR, Assessing the Methodological Quality of Systematic Reviews; COI, conflicts of interest

Supplemental Table 4. Risk of Bias Assessment for Observational Studies

Study	Risk of bias judgement: Confounding	Risk of bias judgement: Selection	Risk of bias judgement: Classification of interventions	Risk of bias judgement: Deviations from intended interventions	Risk of bias judgement: Missing data	Risk of bias judgement: Measurement of outcomes	Risk of bias judgement: Selection of reported result	Overall risk of bias judgement
Wei et al, [214] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Jouhi et al, [9] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Lilja-Fischer et al, [10] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Schneider et al, [11] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Hammarstedt et al, [12] 2021	High	High	Moderate	Moderate	Low	Moderate	Moderate	High
Lundberg et al, [13] 2016	Low	Low	Low	Low	Low	Low	Low	Low
Minami et al, [175] 2017	Low	Low	Low	Low	Low	Moderate	Low	Low
Tachibana et al, [176] 2019	Moderate	Moderate	Low	Low	Low	Low	Low	Moderate
Zhang et al, [177] 2016	Moderate	Moderate	Low	Low	Low	Low	Low	Moderate
Meccariello et al, [14] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Morbini et al, [123] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Baldassarri et al, [215] 2015	Moderate	Low	Moderate	Low	Low	Moderate	Low	Moderate
Meshman et al, [178] 2017	Moderate	Moderate	Moderate	Low	Low	Low	Low	Moderate
Suresh et al, [15] 2021	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Barasch et al, [16] 2016	Low	Low	Low	Low	Low	Moderate	Low	Low
Gondim et al, [124] 2016	Low	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Hongo et al, [153] 2021	Low	Moderate	Moderate	Low	Low	Low	Low	Moderate

Study	Risk of bias judgement: Confounding	Risk of bias judgement: Selection	Risk of bias judgement: Classification of interventions	Risk of bias judgement: Deviations from intended interventions	Risk of bias judgement: Missing data	Risk of bias judgement: Measurement of outcomes	Risk of bias judgement: Selection of reported result	Overall risk of bias judgement
Shinn et al, [17] 2021	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Lai et al, [18] 2016	Moderate	Low	Moderate	Low	Low	Moderate	Low	Moderate
Nauta et al, [19] 2018	Moderate	Moderate	Moderate	Low	Moderate	Moderate	Low	Moderate
Rasmussen et al, [20] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Toman et al, [21] 2017	Moderate	Moderate	Moderate	Low	Moderate	Moderate	Low	Moderate
Tatebe et al, [22] 2018	Moderate	Low	Moderate	Low	Low	Moderate	Low	Moderate
Ou et al, [23] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Abreu et al, [179] 2020	Low	Low	Moderate	Low	Low	Low	Low	Low
Guo et al, [125] 2019	Moderate	Moderate	High	Moderate	Low	Moderate	Low	High
Rollo et al, [126] 2020	Moderate	Moderate	Moderate	Low	Low	Low	Low	Moderate
Jiarpinitnun et al, [180] 2020	Low	Low	Moderate	Low	Low	Low	Low	Low
Sritippho et al, [181] 2016	Low	Low	Low	Low	Low	Low	Low	Low
Kano et al, [182] 2017	Moderate	Moderate	Low	Low	Low	Low	Low	Moderate
Seok et al, [24] 2020	Low	Low	Low	Low	Moderate	Moderate	Low	Moderate
Kwon et al, [127] 2017	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Meng et al, [25] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Gotz et al, [183] 2018	Low	Low	Low	Low	Low	Low	Low	Low
Craig et al, [26] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Gronhoj et al, [27] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate

Study	Risk of bias judgement: Confounding	Risk of bias judgement: Selection	Risk of bias judgement: Classification of interventions	Risk of bias judgement: Deviations from intended interventions	Risk of bias judgement: Missing data	Risk of bias judgement: Measurement of outcomes	Risk of bias judgement: Selection of reported result	Overall risk of bias judgement
Wagner et al, [28] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Wagner et al, [29] 2017	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Kwon et al, [30] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Beitler et al, [31] 2019	Moderate	Low	Moderate	Low	Low	Moderate	Low	Moderate
Biron et al, [216] 2016	Moderate	Low	Low	Low	Low	Moderate	Low	Moderate
Fakhry et al, [32] 2021	Low	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Freitag et al, [33] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Tian et al, [184] 2019	Low	Low	Low	Low	Low	Low	Low	Low
Ni et al, [185] 2019	Low	Low	Low	Low	Moderate	Low	Low	Low
Abi-Raad et al, [217] 2021	Moderate	Moderate	Moderate	Low	Low	Low	Low	Moderate
Buonocore et al, [218] 2019	Moderate	Moderate	Low	Low	Low	Moderate	Low	Moderate
Channir et al, [219] 2016	Moderate	Moderate	Moderate	Moderate	Low	Moderate	Low	Moderate
Wong et al, [220] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Wright et al, [221] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Yang et al, [222] 2019	Moderate	Moderate	Low	Low	Low	Moderate	Low	Moderate
Craig et al, [34] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Buexm et al, [35] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Hoffmann et al, [128] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Norregaard et al, [36] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Wang et al, [186] 2017	Low	Moderate	Low	Low	Low	Low	Low	Low
Xu et al, [37] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate

Study	Risk of bias judgement: Confounding	Risk of bias judgement: Selection	Risk of bias judgement: Classification of interventions	Risk of bias judgement: Deviations from intended interventions	Risk of bias judgement: Missing data	Risk of bias judgement: Measurement of outcomes	Risk of bias judgement: Selection of reported result	Overall risk of bias judgement
Maqsood et al, [38] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Wendt et al, [39] 2021	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Liu et al, [40] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Blatt et al, [187] 2021	Low	Low	Low	Low	Low	Low	Low	Low
Hashmi et al, [188] 2020	Low	Moderate	Low	Low	Low	Low	Low	Low
Nittala et al, [41] 2021	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Rollo et al, [223] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Moderate	Moderate
Song et al, [224] 2021	Moderate	Moderate	Moderate	Low	Low	Low	Moderate	Moderate
Modesto et al, [42] 2019	Moderate	Low	Moderate	Low	Low	Moderate	Low	Moderate
Abdelhakam et al, [225] 2021	Moderate	Moderate	Moderate	Low	Low	Moderate	Moderate	Moderate
Hou et al, [226] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Kapoor et al, [227] 2021	Moderate	Moderate	Low	Low	Low	Moderate	Low	Moderate
Weiss et al, [43] 2021	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Yamashita et al, [44] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Marklund et al, [45] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Beltz et al, [46] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Pettus et al, [228] 2017	High	High	Moderate	Moderate	Low	Moderate	Moderate	High
Wurdemann et al, [47] 2017	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Chi et al, [129] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Baumeister et al, [48] 2017	Low	Low	Moderate	Low	Low	Moderate	Low	Moderate
Broglie et al, [130] 2017	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Cohen et al, [154] 2020	Low	Moderate	Moderate	low	Moderate	Low	Low	Moderate
Dixon et al, [166] 2016	Low	Low	Low	Low	Low	Low	Low	Low

Study	Risk of bias judgement: Confounding	Risk of bias judgement: Selection	Risk of bias judgement: Classification of interventions	Risk of bias judgement: Deviations from intended interventions	Risk of bias judgement: Missing data	Risk of bias judgement: Measurement of outcomes	Risk of bias judgement: Selection of reported result	Overall risk of bias judgement
Driessen et al, [49] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Jiang et al, [189] 2016	Moderate	Low	Moderate	Low	Low	Low	Low	Moderate
Lorch et al, [50] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Mirabile et al, [51] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Molony et al, [52] 2017	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Patel et al, [53] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Ruuskanen et al, [190] 2019	Low	Low	Moderate	Low	Low	Low	Low	Low
Saiyed et al, [131] 2021	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Sivars et al, [229] 2017	Moderate	Moderate	Low	Low	Low	Moderate	Moderate	Moderate
Vital et al, [155] 2017	Low	Moderate	Moderate	Low	Low	Low	Low	Moderate
Zevallos et al, [54] 2016	Moderate	Moderate	Moderate	Low	Low	Low	Low	Moderate
Zhu et al, [191] 2019	Low	Low	Low	Low	Low	Low	Low	Low
Berdugo et al, [55] 2021	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Lewis et al, [56] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Thompson et al, [57] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Nakano et al, [132] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Randen-Brady et al, [133] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Xu et al, [142] 2016	Low	Moderate	Low	Low	Low	Low	Low	Moderate
Kida et al, [58] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Moderate	Moderate
Murthy et al, [143] 2016	Low	Low	Moderate	Low	Low	Low	Low	Low
Kiyuna et al, [192] 2019	Low	Low	Low	Low	Low	Low	Low	Low
Merlano et al, [144] 2016	Low	Low	Moderate	Low	Low	Low	Low	Low
Vivenza et al, [193] 2016	Low	Low	Moderate	Low	Low	Low	Low	Low
Abrahao et al, [194] 2018	Moderate	Low	Moderate	Low	Low	Low	Low	Moderate
Cheol Park et al, [167] 2017	Low	Low	Low	Low	Low	Moderate	Low	Low

Study	Risk of bias judgement: Confounding	Risk of bias judgement: Selection	Risk of bias judgement: Classification of interventions	Risk of bias judgement: Deviations from intended interventions	Risk of bias judgement: Missing data	Risk of bias judgement: Measurement of outcomes	Risk of bias judgement: Selection of reported result	Overall risk of bias judgement
Dogan et al, [59] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Garnaes et al, [60] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Huebbers et al, [61] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Nauta et al, [195] 2021	Low	Low	Moderate	Low	Low	Low	Low	Low
Ren et al, [62] 2020	Low	Moderate	Moderate	Low	Low	Low	Moderate	Moderate
Simon et al, [196] 2020	Low	Low	Low	Low	Low	Moderate	Low	Low
Wagner et al, [63] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Wang et al, [64] 2016	Low	Moderate	Moderate	Low	Low	Low	Low	Moderate
Yang et al, [134] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Mizumachi et al, [65] 2017	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Yamamoto et al, [66] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Wuerdemann et al, [67] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Caparrotti et al, [68] 2017	Moderate	Low	Moderate	Low	Low	Low	Low	Moderate
Ringash et al, [69] 2017	Low	Low	Low	Low	Moderate	Low	Low	Low
Wu et al, [70] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Ghantous et al, [197] 2018	Low	Low	Moderate	Low	Low	Low	Low	Moderate
Bryant et al, [198] 2018	Moderate	Low	Moderate	Low	Low	Low	Low	Moderate
Fakhry et al, [242] 2019	Low	Low	Low	Low	Low	Low	Low	Low
Kharytaniuk et al, [168] 2016	Moderate	Moderate	Low	Low	Low	Moderate	Low	Moderate
Skillington et al, [71] 2016	Moderate	Moderate	Moderate	Low	Low	Low	Low	Moderate
Lee et al, [72] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
O'Neill et al, [73] 2021	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Hewavisenti et al, [74] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Ambulos et al, [135] 2016	Moderate	Moderate	Moderate	Low	Low	Low	Low	Moderate
Cho et al, [169] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Descamps et al, [75] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate

Study	Risk of bias judgement: Confounding	Risk of bias judgement: Selection	Risk of bias judgement: Classification of interventions	Risk of bias judgement: Deviations from intended interventions	Risk of bias judgement: Missing data	Risk of bias judgement: Measurement of outcomes	Risk of bias judgement: Selection of reported result	Overall risk of bias judgement
Janecka-Widla et al, [76] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Ko et al, [199] 2017	Low	Moderate	Low	Low	Low	Low	Low	Low
Chakravarthy et al, [200] 2016	Low	Low	Low	Low	Low	Low	Low	Low
Rosenthal et al, [77] 2016	Moderate	Low	Moderate	Low	Low	Moderate	Low	Moderate
Sato et al, [78] 2019	Moderate	Moderate	Moderate	Low	Low	Low	Low	Moderate
Argirion et al, [79] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Guerendiain et al, [136] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Takes et al, [230] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Ren et al, [145] 2019	Moderate	Low	Moderate	Low	Low	Moderate	Low	Moderate
Palve et al, [201] 2018	Low	Low	Low	Low	Low	Moderate	Low	Low
Kao et al, [80] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Masoud Rahbari et al, [81] 2016	High	High	Moderate	Low	Low	Moderate	Low	High
Huho et al, [231] 2018	Moderate	Moderate	Moderate	Low	Moderate	Moderate	Low	Moderate
Baschnagel et al, [202] 2017	Moderate	Low	Low	Low	Low	Low	Low	Low
Gurin et al, [82] 2020	Moderate	Moderate	Low	Low	Low	Low	Low	Moderate
Gurin et al, [83] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Lai et al, [203] 2017	Low	Low	Low	Low	Low	Low	Low	Low
Lu et al, [84] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Singh et al, [146] 2017	Low	Low	Low	Low	Low	Moderate	Low	Low
Cohen et al, [232] 2017	Low	Low	Low	Low	Low	Low	Low	Low
Sivarajah et al, [85] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Gargano et al, [233] 2021	Moderate	Moderate	Moderate	Low	Low	Low	Low	Moderate
Han et al, [234] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Manucha et al, [235] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Wilson et al, [236] 2021	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Wilson et al, [237] 2021	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate

Study	Risk of bias judgement: Confounding	Risk of bias judgement: Selection	Risk of bias judgement: Classification of interventions	Risk of bias judgement: Deviations from intended interventions	Risk of bias judgement: Missing data	Risk of bias judgement: Measurement of outcomes	Risk of bias judgement: Selection of reported result	Overall risk of bias judgement
Kwon et al, [86] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Jang et al, [238] 2020	High	High	High	Moderate	Low	Moderate	Moderate	High
Boeker et al, [170] 2021	Low	Low	Low	Low	Low	Low	Low	Low
Chowdhury et al, [156] 2017	Low	Moderate	Moderate	Low	Low	Low	Low	Moderate
Dronkers et al, [87] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Yin et al, [88] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Kiessling et al, [89] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Schlussel Markovic et al, [157] 2020	Low	Moderate	Moderate	Low	Moderate	Low	Low	Moderate
Cobzeanu et al, [147] 2020	Low	Low	Low	Low	Low	Low	Low	Low
Augustin et al, [90] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Hongo et al, [148] 2021	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Shelton et al, [91] 2017	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Schiavetto et al, [92] 2021	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Cierpikowski et al, [204] 2021	Low	Low	Low	Low	Low	Moderate	Low	Low
Svajdler et al, [158] 2020	Low	Moderate	Moderate	Low	Low	Low	Low	Moderate
Yang et al, [205] 2018	Low	Low	Low	Low	Low	Low	Low	Low
Tsuchida et al, [149] 2017	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Dok et al, [93] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Gronhoj et al, [94] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Hong et al, [95] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Huang et al, [243] 2017	Low	Low	Low	Low	Low	Moderate	Low	Low
Kumar et al, [96] 2017	Moderate	Moderate	Low	Low	Low	Low	Low	Moderate
Larsen et al, [137] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Yang et al, [206] 2016	Low	Low	Low	Low	Low	Low	Low	Low
Fanetti et al, [97] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate

Study	Risk of bias judgement: Confounding	Risk of bias judgement: Selection	Risk of bias judgement: Classification of interventions	Risk of bias judgement: Deviations from intended interventions	Risk of bias judgement: Missing data	Risk of bias judgement: Measurement of outcomes	Risk of bias judgement: Selection of reported result	Overall risk of bias judgement
Lu et al, [98] 2023	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Broglie et al, [99] 2017	Low	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Chatfield-Reed et al, [244] 2020	Moderate	Moderate	Moderate	Low	Moderate	Moderate	Low	Moderate
D'Souza et al, [207] 2016	Low	Moderate	Low	Low	Low	Low	Low	Low
El-Salem et al, [239] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Gronhoj et al, [100] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Hernandez et al, [208] 2019	Low	Low	Low	Low	Low	Low	Low	Low
Hong et al, [101] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Hughes et al, [209] 2019	Low	Moderate	Low	Low	Low	Moderate	Low	Moderate
Lacau St Guily et al, [138] 2017	Low	Moderate	Moderate	Low	Moderate	Moderate	Low	Moderate
Mashiana et al, [240] 2021	Moderate	Moderate	Moderate	Low	Low	Low	Low	Moderate
Mena et al, [102] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Mirghani et al, [103] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Schneider et al, [210] 2020	Low	Moderate	Low	Low	Low	Low	Low	Low
Schroeder et al, [104] 2020	Low	Low	Moderate	Low	Low	Moderate	Low	Moderate
Taberna et al, [211] 2016	Low	Moderate	Low	Low	Low	Moderate	Low	Moderate
Zafereo et al, [212] 2016	Low	Low	Low	Low	Low	Moderate	Low	Low
Girardi et al, [105] 2020	Moderate	Moderate	Moderate	Low	Moderate	Moderate	Low	Moderate
Grisar et al, [106] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Xu et al, [107] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Kaka et al, [108] 2017	Moderate	Moderate	Low	Low	Low	Low	Low	Moderate
Sanchez Barrueco et al, [150] 2019	Low	Moderate	Low	Low	Low	Moderate	Low	Moderate
Chernesky et al, [241] 2018	Low	Low	Low	Low	Low	Low	Low	Low
Clark et al, [109] 2019	Low	Moderate	Low	Low	Low	Moderate	Low	Moderate
Hernandez et al, [151] 2016	Low	Low	Low	Low	Low	Low	Low	Low

Study	Risk of bias judgement: Confounding	Risk of bias judgement: Selection	Risk of bias judgement: Classification of interventions	Risk of bias judgement: Deviations from intended interventions	Risk of bias judgement: Missing data	Risk of bias judgement: Measurement of outcomes	Risk of bias judgement: Selection of reported result	Overall risk of bias judgement
Satgunaseelan et al, [152] 2016	Low	Moderate	Low	Low	Low	Low	Low	Low
Alexiev et al, [110] 2020	Moderate	Low	Moderate	Low	Low	Moderate	Low	Moderate
Meng et al, [245] 2020	Moderate	Moderate	Low	Low	Low	Moderate	Low	Moderate
Bozinovic et al, [139] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Lorenzatti Hiles et al, [111] 2021	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Ni et al, [246] 2019	Moderate	Moderate	Low	Low	Low	Moderate	Low	Moderate
Wang et al, [247] 2016	Moderate	Moderate	Low	Low	Low	Moderate	Low	Moderate
Hong et al, [112] 2016	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Jensen et al, [213] 2021	Low	Moderate	Low	Low	Low	Low	Low	Low
Linge et al, [248] 2018	Low	Moderate	Low	Low	Low	Moderate	Low	Low
Golusinski et al, [113] 2017	Moderate	Moderate	Moderate	Low	Moderate	Moderate	Low	Moderate
Chen et al, [114] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Del Mistro et al, [115] 2020	Moderate	Low	Moderate	Low	Low	Moderate	Low	Moderate
Kemnade et al, [116] 2020	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Poropatich et al, [117] 2019	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Carpen et al, [118] 2018	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Biesaga et al, [119] 2021	Moderate	Moderate	Moderate	Low	Low	Moderate	Low	Moderate
Mehanna et al, [120] 2023	Low	Low	Low	Low	Low	Moderate	Low	Low
Lifsics et al, [140] 2023	Moderate	Moderate	Low	Low	Low	Low	Low	Moderate
Wu et al, [159] 2022	Moderate	Moderate	Moderate	Low	Moderate	Moderate	Low	Moderate

Supplemental Table 5. Certainty of Evidence Assessment for Statement 1

Outcome	Number of Studies	Design	Aggregate Risk of Bias	Inconsistency	Indirectness	Imprecision	Other	Importance	Certainty of Evidence Grade for Outcome	Overall Certainty of Evidence Grade for Statement
OS	102	2 SRs, 100 Observational	Serious	Not serious	Not serious	Not serious	None	Critical	High[a]	High[a]
DFS	36	1 SR, 35 Observational	Serious	Not serious	Not serious	Not serious	None	Critical	High[a]	
PFS	19	2 SRs, 17 Observational	Serious	Not serious	Not serious	Not serious	None	Critical	High[a]	
DSS	19	1 SR, 18 Observational	Serious	Not serious	Not serious	Not serious	None	Critical	High[a]	
Other clinical outcomes	30	2 SRs, 28 Observational	Serious	Not serious	Not serious	Not serious	None	Critical	High[a]	

[a]Certainty of evidence was upgraded to high for all outcomes based on a large body of evidence showing strong and consistent clinical benefits in patients with HPV-associated versus HPV-independent OPSCC.
Abbreviations: DFS, disease-free survival; DSS, disease-specific survival; OS, overall survival; PFS, progression-free survival

Supplemental Table 6. Certainty of Evidence Assessment for Statement 2

Outcome	Number of Studies	Design	Aggregate Risk of Bias	Inconsistency	Indirectness	Imprecision	Other	Importance	Certainty of Evidence Grade for Outcome	Overall Certainty of Evidence Grade for Statement
OS	67	3 SRs, 64 Observational	Serious	Not serious	Not serious	Not serious	None	Critical	High	High[a]
DFS	22	3 SRs, 19 Observational	Serious	Not serious	Not serious	Not serious	None	Critical	High	
PFS	12	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	Moderate	
Sen/Spec	25	Observational	Serious	Serious	Not serious	Not serious	None	Critical	Moderate	
PPV/NPV	20	Observational	Serious	Serious	Not serious	Not serious	None	Critical	Moderate	
Concordance	18	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	Moderate	

[a]Certainty of evidence was upgraded to high based on a large body of evidence showing strong and consistent clinical benefits and testing outcomes in patients with p16-positive versus p16-negative OPSCC.
Abbreviations: DFS, disease-free survival; DSS, disease-specific survival; NPV, negative predictive value; OS, overall survival; PFS, progression-free survival; PPV, positive predictive value; sen, sensitivity; spec, specificity, SR, systematic review

Supplemental Table 7. Certainty of Evidence Assessment for Statement 3

Outcome	Number of Studies	Design	Aggregate Risk of Bias	Inconsistency	Indirectness	Imprecision	Other	Importance	Certainty of Evidence Grade for Outcome	Overall Certainty of Evidence Grade for Statement
Testing outcomes & criteria for positive test	27	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	High	High[a]

[a]Certainty of evidence was upgraded to high based on a strong association and consistency between criteria for positive test and testing outcomes.

Supplemental Table 8. Certainty of Evidence Assessment for Statements 4 and 5

Outcome	Number of Studies	Design	Aggregate Risk of Bias	Inconsistency	Indirectness	Imprecision	Other	Importance	Certainty of Evidence Grade for Outcome	Overall Certainty of Evidence Grade for Statement
OS	8	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	Moderate	Moderate
DFS	3	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	Moderate	
DSS	1	Observational	Serious	NA	Not serious	Not serious	Single study	Critical	Moderate	
Sen/Spec	6	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	Moderate	

Abbreviations: DFS, disease-free survival; DSS, disease-specific survival; NA, not available; OS, overall survival; sen, sensitivity; spec, specificity

Supplemental Table 9. Certainty of Evidence Assessment for Statement 6

Outcome	Number of Studies	Design	Aggregate Risk of Bias	Inconsistency	Indirectness	Imprecision	Other	Importance	Certainty of Evidence Grade for Outcome	Overall Certainty of Evidence Grade for Statement
OS	8	1 SR, 7 Observational	Serious	Not serious	Not serious	Not serious	None	Critical	High	High[a]
DFS	3	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	Moderate	
PFS	1	Systematic Review/ Meta-analysis	Serious	NA	Not serious	Not serious	10 studies included in meta-analysis	Critical	High	

DSS	1	Observational	Serious	NA	Not serious	Not serious	Single study	Critical	Moderate	
Recurrence	2	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	Moderate	

[a]Certainty of evidence was upgraded to high based on evidence from meta-analyses showing strong and consistent clinical benefits in HPV-associated versus HPV-independent patients with carcinoma of unknown primary (CUP).
Abbreviations: DFS, disease-free survival; DSS, disease-specific survival; NA, not available; OS, overall survival; PFS, progression-free survival, SR, systematic review

Supplemental Table 10. Certainty of Evidence Assessment for Statement 7

Outcome	Number of Studies	Design	Aggregate Risk of Bias	Inconsistency	Indirectness	Imprecision	Other	Importance	Certainty of Evidence Grade for Outcome	Overall Certainty of Evidence Grade for Statement
Sen/Spec	1	Observational	Serious	NA	Not serious	Not serious	Single study	Critical	Moderate	Moderate
PPV/NPV	1	Observational	Serious	NA	Not serious	Not serious	Single study	Critical	Moderate	
Concordance	1	Observational	Serious	NA	Not serious	Not serious	Single study	Critical	Moderate	

Abbreviations: NA, not available; NPV, negative predictive value; PPV, positive predictive value; sen, sensitivity; spec, specificity

Supplemental Table 11. Certainty of Evidence Assessment for Statement 8

Outcome	Number of Studies	Design	Aggregate Risk of Bias	Inconsistency	Indirectness	Imprecision	Other	Importance	Certainty of Evidence Grade for Outcome	Overall Certainty of Evidence Grade for Statement
OS	39	3 SRs, 36 Observational	Serious	Not serious	Not serious	Not serious	None	Critical	High	High[a]
DFS	15	1 SR, 14 Observational	Serious	Not serious	Not serious	Not serious	None	Critical	High	
PFS	6	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	Moderate	
DSS	9	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	Moderate	
Sen/Spec	17	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	Moderate	
PPV/NPV	16	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	Moderate	

[a]Certainty of evidence was upgraded to high based on studies showing a lack of statistically significant differences in clinical and testing outcomes.
Abbreviations: DFS, disease-free survival; DSS, disease-specific survival; NPV, negative predictive value; OS, overall survival; PFS, progression-free survival; PPV, positive predictive value; sen, sensitivity; spec, specificity, SR, systematic review

Supplemental Table 12. Certainty of Evidence Assessment for Statement 10 and Statement 11

Outcome	Number of Studies	Design	Aggregate Risk of Bias	Inconsistency	Indirectness	Imprecision	Other	Importance	Certainty of Evidence Grade for Outcome	Overall Certainty of Evidence Grade for Statement
OS	2	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	Moderate	High[a]
DFS	2	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	Moderate	
Sen/Spec	29	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	Moderate	
PPV/NPV	21	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	Moderate	
Concordance/ Kappa	15	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	Moderate	

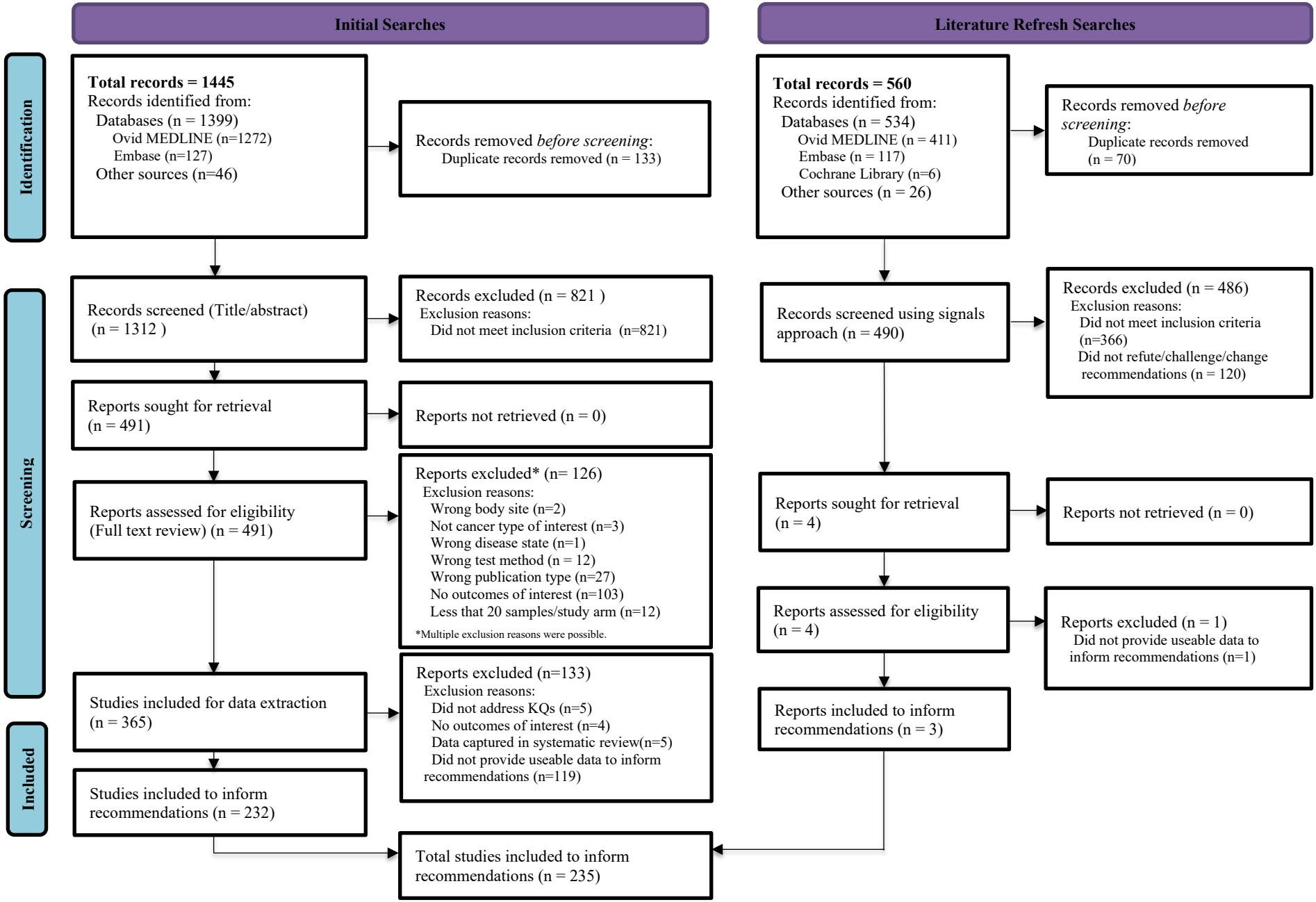
[a]Certainty of evidence was upgraded to high based on a strong association and consistency in clinical outcome and testing outcomes in fine needle aspirations of nodal SCC samples from all patients, with clinical findings of an oropharyngeal or sinonasal primary, or with metastatic SCC of unknown primary.
Abbreviations: DFS, disease-free survival; NPV, negative predictive value; OS, overall survival; PPV, positive predictive value; sen, sensitivity; spec, specificity

Supplemental Table 13. Certainty of Evidence Assessment for Statement 12

Outcome	Number of Studies	Design	Aggregate Risk of Bias	Inconsistency	Indirectness	Imprecision	Other	Importance	Certainty of Evidence Grade for Outcome	Overall Certainty of Evidence Grade for Statement
OS	19	3 SRs, 16 Observational	Serious	Not serious	Not serious	Not serious	None	Critical	High	High[a]
DFS	7	2 SRs, 5 Observational	Serious	Not serious	Not serious	Not serious	None	Critical	High	
PFS	1	Observational	Serious	NA	Not serious	Not serious	Single study	Critical	Moderate	
DSS	3	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	Moderate	
Other clinical outcomes	4	Observational	Serious	Not serious	Not serious	Not serious	None	Critical	Moderate	

[a]Certainty of evidence was upgraded based on a high correlation between diagnostic test characteristics and clinical outcomes.
Abbreviations: DFS, disease-free survival; DSS, disease-specific survival; NA, not available; OS, overall survival; PFS, progression-free survival, SR, systematic review

Supplemental Figure 1: Systematic Literature Review Flow Diagram



Adapted From: Page MJ, McKenzie JE, Bossuyt PM, Boutron I, Hoffmann TC, Mulrow CD, et al. The PRISMA 2020 statement: an updated guideline for reporting systematic reviews. *BMJ* 2021;372:n71. doi: 10.1136/bmj.n71. For more information, visit: http://www.prisma-statement.org/

Supplemental Figure 2: Database Search Strings

Ovid MEDLINE Search String:

Ovid MEDLINE(R) and Epub Ahead of Print, In-Process, In-Data-Review & Other Non-Indexed Citations, Daily and Versions(R) <1946 to July 06, 2021>

1 exp alphapapillomavirus/ 8200
2 Papillomavirus E7 proteins/ 2609
3 (HPV or HR-HPV or HPV-pos$ or HPV-associated or HPV-related).tw,kf. 45180
4 Human papillomavirus.tw,kf. 37622
5 High-risk HPV.tw,kf. 4747
6 High-risk human papillomavirus.tw,kf. 3080
7 human papillomavirus-associated.tw,kf. 642
8 human papillomavirus-related.tw,kf. 575
9 human papillomavirus-positive.tw,kf. 483
10 ((E6 or E7) and (oncoprotein$ or protein$)).tw,kf. 6811
11 1 or 2 or 3 or 4 or 5 or 6 or 7 or 8 or 9 or 10 56200
12 Human Papillomavirus DNA tests/ 544
13 Biopsy, Fine Needle/ 14050
14 DNA probes, HPV/ 1067
15 Immunohistochemistry/ 296985
16 Polymerase Chain Reaction/ 246026
17 Tissue Array Analysis/ 8686
18 In Situ Hybridization/ 50375
19 Cyclin-Dependent Kinase Inhibitor p16/ 8540
20 (CDKN2A or P16?INK?41 or P16 or RT?PCR or DNA?PCR or DNA?ISH or RNA?ISH or IHC or PCR or ISH).tw,kf. 586543
21 DNA probe$.tw,kf. 13106
22 DNA test$.tw,kf. 4560
23 Cyclin-dependent kinase inhibitor p16.tw,kf. 135
24 Immunohistochemistry.tw,kf. 201945
25 Polymerase chain reaction.tw,kf. 258951
26 Tissue array analysis.tw,kf. 84
27 situ hybridization.tw,kf. 95674
28 FNA.tw,kf. 10896
29 Fine needle aspiration.tw,kf. 29283
30 Liquid base$.tw,kf. 3719
31 12 or 13 or 14 or 15 or 16 or 17 or 18 or 19 or 20 or 21 or 22 or 23 or 24 or 25 or 26 or 27 or 28 or 29 or 30 1283631
32 11 and 31 17455
33 "Squamous Cell Carcinoma of Head and Neck"/ 7025
34 Palatal Neoplasms/ 3019
35 exp Salivary Gland Neoplasms/ 17932
36 Tongue Neoplasms/ 10372

37 Laryngeal Neoplasms/ 28048
38 exp Nose Neoplasms/ 17911
39 exp Pharyngeal Neoplasms/ 35468
40 Lymphatic Metastasis/ 92570
41 33 or 34 or 35 or 36 or 37 or 38 or 39 or 40 195193
42 OP?SCC.tw,kf. 1139
43 (Squamous cell carcinoma and (Oropharyngeal or Non-oropharyngeal)).tw,kf. 3500
44 Non?squamous cell carcinoma.tw,kf. 71
45 (Carcinoma$ or malignan$ or neoplas$ or cancer$ or tumor$ or tumour$ or metast$).tw,kf. 3611083
46 42 or 43 or 44 or 45 3611083
47 (Mouth or oral cavity or jaw).tw,kf. 138089
48 (salivary gland$ or lingual or sublingual or submandibular or Tongue).tw,kf. 113091
49 waldeyer$ ring.tw,kf. 644
50 (pharynx or pharyngeal or hypopharynx or hypopharyngeal or oropharynx or oropharyngeal or tonsil$ or palate or Uvula).tw,kf. 124646
51 (larynx or laryngeal or parotid or glottis or epiglottis).tw,kf. 96749
52 (nasal cavity or nasopharynx or nasopharyngeal or paranasal sinuses or sinonasal).tw,kf. 63922
53 47 or 48 or 49 or 50 or 51 or 52 471336
54 46 and 53 132622
55 41 or 54 257311
56 11 and 31 and 55 3054
57 limit 56 to yr="2016-current" 1393
58 limit 57 to english language 1350
59 58 not (animals/ not (animals/ and humans/)) 1344
60 limit 59 to (case reports or comment or editorial or letter) 72
61 59 not 60 1272

Embase Search String:

No. Query Results
#68 #66 AND #67 55
#67 [07-07-2021]/sd 2588953
#66 #62 NOT #65 289
#65 #63 NOT #64 23
#64 systematic:ti,ab,kw OR data:ti,ab,kw OR rationale:ti,ab,kw OR evidence:ti,ab,kw OR cohort:ti,ab,kw 8882198
#63 #62 AND [review]/lim 41
#62 #60 NOT #61 312
#61 #60 AND ([conference abstract]/lim OR [editorial]/lim OR [letter]/lim) 1064
#60 #58 NOT #59 1376
#59 #58 AND [medline]/lim 1693
#58 #55 NOT #56 AND [english]/lim 3069
#57 #55 NOT #56 3137
#56 #55 AND ([animal cell]/lim OR [animal experiment]/lim OR [animal model]/lim OR [animal tissue]/lim) 51

#55 #11 AND #31 AND #53 3188
#54 #32 AND #53 3188
#53 #39 AND #52 77920
#52 #44 AND #51 181724
#51 #45 OR #46 OR #47 OR #48 OR #49 OR #50 611425
#50 'nasal cavity':ti,ab,kw OR nasopharynx:ti,ab,kw OR nasopharyngeal:ti,ab,kw OR 'paranasal sinuses':ti,ab,kw OR sinonasal:ti,ab,kw 88332
#49 larynx:ti,ab,kw OR laryngeal:ti,ab,kw OR parotid:ti,ab,kw OR glottis:ti,ab,kw OR epiglottis:ti,ab,kw 124637
#48 pharynx:ti,ab,kw OR pharyngeal:ti,ab,kw OR hypopharynx:ti,ab,kw OR hypopharyngeal:ti,ab,kw OR oropharynx:ti,ab,kw OR oropharyngeal:ti,ab,kw OR tonsil*:ti,ab,kw OR palate:ti,ab,kw OR uvula:ti,ab,kw 167806
#47 'waldeyer* ring':ti,ab,kw 154
#46 'salivary gland*':ti,ab,kw OR lingual:ti,ab,kw OR sublingual:ti,ab,kw OR submandibular:ti,ab,kw OR tongue:ti,ab,kw 148097
#45 mouth:ti,ab,kw OR 'oral cavity':ti,ab,kw OR jaw:ti,ab,kw 178661
#44 #40 OR #41 OR #42 OR #43 5227264
#43 'non*squamous cell carcinoma':ti,ab,kw 106
#42 carcinoma*:ti,ab,kw OR malignan*:ti,ab,kw OR neoplas*:ti,ab,kw OR cancer*:ti,ab,kw OR tumor*:ti,ab,kw OR tumour*:ti,ab,kw OR metast*:ti,ab,kw 5227262
#41 'squamous cell carcinoma':ti,ab,kw AND (oropharyngeal:ti,ab,kw OR 'non oropharyngeal':ti,ab,kw) 5772
#40 'op*scc':ti,ab,kw 2046
#39 #33 OR #34 OR #35 OR #36 OR #37 OR #38 290567
#38 'lymphatic metastasis'/exp 163531
#37 'pharyngeal neoplasms'/exp 48920
#36 'tongue neoplasms'/exp 14263
#35 'salivary gland neoplasms'/exp 25058
#34 'palatal neoplasms'/exp 25201
#33 'squamous cell carcinoma of head and neck'/exp 34516
#32 #11 AND #31 27137
#31 #12 OR #13 OR #14 OR #15 OR #16 OR #17 OR #18 OR #19 OR #20 OR #21 OR #22 OR #23 OR #24 OR #25 OR #26 OR #27 OR #28 OR #29 OR #30 2253566
#30 ‘liquid base$‘:ti,ab,kw 73
#29 'fine needle aspiration':ti,ab,kw 42662
#28 'fna':ti,ab,kw 21956
#27 'situ hybridization':ti,ab,kw 118361
#26 'tissue array analysis':ti,ab,kw 151
#25 'polymerase chain reaction':ti,ab,kw 312211
#24 'immunohistochemistry':ti,ab,kw 339653
#23 'cyclin-dependent kinase inhibitor p16':ti,ab,kw 186
#22 'dna test*':ti,ab,kw 7125
#21 'dna probe*':ti,ab,kw 14311
#20 cdkn2a:ti,ab,kw OR p16*ink*41:ti,ab,kw OR p16:ti,ab,kw OR rt*pcr:ti,ab,kw OR dna*pcr:ti,ab,kw OR dna*ish:ti,ab,kw OR rna*ish:ti,ab,kw OR ihc:ti,ab,kw OR pcr:ti,ab,kw OR ish:ti,ab,kw 952155

#19 'cyclin-dependent kinase inhibitor p16'/exp 15371
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#17 'tissue array analysis'/exp 26593
#16 'polymerase chain reaction'/exp 1109490
#15 'immunohistochemistry'/exp 721821
#14 'dna probes, hpv'/exp 30103
#13 'biopsy, fine needle'/exp 40664
#12 'human papillomavirus dna tests'/exp 2457
#11 #1 OR #2 OR #3 OR #4 OR #5 OR #6 OR #7 OR #8 OR #9 OR #10 81652
#10 (e6:ti,ab,kw OR e7:ti,ab,kw) AND (oncoprotein$:ti,ab,kw OR protein$:ti,ab,kw) 9023
#9 'human papillomavirus-positive':ti,ab,kw 633
#8 'human papillomavirus-related':ti,ab,kw 709
#7 'human papillomavirus-associated':ti,ab,kw 808
#6 'high-risk human papillomavirus':ti,ab,kw 4071
#5 'high-risk hpv':ti,ab,kw 7271
#4 'human papillomavirus':ti,ab,kw 48537
#3 hpv:ti,ab,kw OR 'hr hpv':ti,ab,kw OR 'hpv pos*':ti,ab,kw OR 'hpv associated':ti,ab,kw OR 'hpv related':ti,ab,kw 66684
#2 'papillomavirus e7 proteins'/exp 3328
#1 'alphapapillomavirus'/exp 18214

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