COLLEGE of AMERICAN PATHOLOGISTS
Laboratory Quality Solutions

Supplemental Digital Content* | Methodology | May 2020

# Collection and Handling of Thoracic Small Biopsy and Cytology Specimens for Ancillary Studies

Authors:
Lesley Souter, PhD
Sinchita Roy-Chowdhuri, MD, PhD
Christopher Gilbert, DO
Brooke Billman, MLIS
Nicole E. Thomas, MPH, CT(ASCP)

*The Supplemental Digital Content was not copyedited by *Archives of Pathology & Laboratory Medicine*.

## METHODS USED TO PRODUCE THE GUIDELINE

### Panel Composition

The College of American Pathologists (CAP) along with its collaborators, the American College of Chest Physicians (CHEST), Association for Molecular Pathology (AMP), American Society of Cytopathology (ASC), American Thoracic Society (ATS), Pulmonary Pathology Society (PPS), Papanicolaou Society of Cytopathology (PSC), Society of Interventional Radiology (SIR), and Society of Thoracic Radiology (STR), convened an expert panel (EP) consisting of 10 pathologists, two pulmonologists, four radiologists, and one cytotechnologist with expertise in pulmonology, pulmonary pathology, molecular pathology, cytopathology, microbiology, radiology, cytotechnology, and a methodologist consultant to develop an evidence-based guideline to address collection and handling of thoracic small biopsy and cytology specimens for ancillary testing. The CAP approved the appointment of the project co-chairs and panel members. The EP members performed the systematic evidence review, drafted the recommendations, evaluated the public comments, revised the recommendations and contributed to the manuscripts.

An advisory panel (AP) of 11 pathologists, three pulmonologists, two radiologists, and one cytotechnologist also helped in the development of the guideline. The role of the AP members was to provide guidance and feedback on the scope and key questions for the literature search, vet the draft guideline statements prior to the public comment period, and to review and provide feedback for the manuscript and supplemental digital content (SDC).

### Conflict of Interest (COI) Policy

Prior to acceptance on the expert or advisory panel, potential members completed the collaborative conflict of interest (COI) disclosure process, whose policy and form (effective March 2016) require disclosure of material financial interest in, or potential for benefit of significant value from, the guideline's development or its recommendations 36 months prior through 12 months post-publication. The potential members completed the COI disclosure form, listing any relationship that could be interpreted as constituting an actual, potential, or apparent conflict. Each potential expert panel member's disclosures were assessed by a COI review committee and categorized as:

No Relevant Conflicts of Interest**:** Individuals with no relevant COI are approved for full participation including determining the scope and questions to be addressed, reviewing and discussing the evidence, formulating and grading recommendations, voting on recommendations, and writing the document. Research funding that is free of direct or indirect industry funding or control, such as that provided by a government program or a non-profit organization that does not receive industry funding and uses an award mechanism and oversight that is independent of industry, is not regarded to be a conflict of interest. Service on a data and safety monitoring board for such research is also not regarded as a conflict of interest. Finally, industry funded research unrelated to the content of the *Joint Recommendations* is not regarded as a conflict of interest.

Manageable Conflicts of Interest: Individuals with manageable conflicts must disclose their conflicts to the whole guideline panel. They may participate in discussions about the evidence, but must excuse themselves or be recused from decision-making, including formulating, voting on, writing, and grading recommendations related to their COI (i.e., recommendations addressing a product of the commercial entity with which they have a relationship or addressing a product of a competitor of the commercial entity with which they have a relationship). COI that require management include:

a. Research funding from an industry grant that is paid to the participant's institution and related to the content of the *Recommendations*;

b. Research funding from a government program or non-profit organization that receives funding from industry with business interests in the content of the *Recommendations*;
c. Participation on a data and safety monitoring board concerned with research that is relevant to the content of the *Recommendations* and is funded by an industry with business interests in the content of the *Recommendations*, or by a government program or non-profit organization that receives funding from industry with business interests in the content of the *Recommendations*.
d. Participation in scientific advisory board or consultant activities that are exclusively scientific in nature (i.e., does not involve any activities that could be perceived as promotional) related to the subject matter of the *Recommendations.*
e. Participation in industry-funded research, scientific advisory committees, consulting roles, non-promotional speaking engagements, or expert testimony on matters that are unrelated to the subject matter of the *Recommendations*, but the company involved is known to have business interest in the subject matter;
f. Delivery of non-promotional talks in which the speaker has full control of the content and is either unpaid or paid by a third party that is responsible for ensuring that the event is free of influence of relevant industry (i.e. if the event has industry financial support, all planning and content must be free of industry influence, and any payment of expenses and honoraria must occur through a third party, such as the medical society or institution sponsoring the event, or an event manager acceptable to them, rather than directly by a commercial entity with an interest in guideline subject matter or its agent);
g. Professional roles or activities (i.e., roles and activities performed as part of an individual's profession, whether reimbursed or not) that place an individual in a position to personally gain or lose depending upon the recommendations.

Disqualifying Conflicts of Interest: Disqualifying conflicts of interest include the following:
a. Any current professional relationship with or investment in a company involved in the manufacture or distribution of tobacco products.
b. A direct financial relationship with a relevant commercial entity that has an interest in the content of the Recommendations, exclusive of the research, data safety monitoring board activities, and scientific advisory board and consultant activities noted above. Such direct financial relationships include the following, whether paid to or held by the individual directly or issued to another entity at the direction of the individual (such as to a panelist's institution):
    i. Payment of wages, consulting fees, honoraria, or other payments (in cash, in stock or stock options, or in kind) by a relevant company as compensation for the individual's services or expertise, exclusive of the research and data safety monitoring board activities noted above. Examples of such services are: participation on scientific advisory committees or consulting that is, in full or in part, promotional in nature; non-continuing medical education (CME) speaking engagements and inclusion in speaker bureaus where control of material is held by industry; expert testimony on matters related to guideline content provided on behalf of a relevant company or a law firm representing a relevant company; employment by a relevant commercial entity (such as a relevant pharmaceutical or medical device company or a third party payer exclusive of commercial laboratory employment that has financial interests in the content of the Recommendations).
    ii. Investments in relevant companies by the panelist or the panelist's spouse or life partner (exclusive of general mutual funds).
c. A patent or other intellectual property that is relevant to the Recommendations' subject matter and has resulted or could result in payments to the panelist or the panelist's institution.

All panel members were required to disclose conflicts prior to beginning and continuously throughout the project's timeline.

Disclosures of interest judged by the oversight group as manageable conflicts are listed in the manuscript. Appendix 1 in the manuscript also includes a table of all disclosed interest of the expert panel members during the development of the guideline for complete transparency.

## Funding
The CAP provided funding for the administration of the project; no industry funds were used in the development of the guideline.

## Systematic Evidence Review (SER)
The objective of the SER was to identify articles of sufficient quality that would provide data to inform the recommendations. The scope of the SER and the key questions (KQs) were established by the EP and AP in consultation with the methodologist prior to beginning the literature search.

## Search and Selection
A comprehensive literature search was performed in PubMed on 3/30/2017. The search was limited to 1/1/2007- 3/3/2017. An additional search was performed in Embase.com with the same search date parameters. The databases searches used indexed terms and keywords for the concepts of thoracic abnormalities; specimen procurement methods; laboratory tests; and methodological, analytical, and procedural variables. Search results were limited to English language and the Cochrane search filter for humans was applied.[1] A publication filter to exclude letters, commentaries, editorials, case reports, and conference abstracts was added. Results of both searches were combined, and duplicate references were removed. A literature search refresh was completed in PubMed and Embase.com on 5/15/2018 and again 4/30/2019. Both search strategies can be found in Supplemental Figure 1.

A search for grey (unindexed) literature included a review of the ClinicalTrials.gov, Cochrane Library, Guidelines International Network, National Guideline Clearinghouse, Trip search engine, University of York Centre for Reviews and Dissemination- PROSPERO, and applicable U.S. and international organizational websites.

## Study Selection Criteria
Eligible Study Designs
Included study types: clinical practice guidelines, systematic reviews with meta-analyses, systematic reviews, randomized controlled trials, nonrandomized controlled trials, uncontrolled trials, observational studies, non-comparative studies. Excluded studies: follow-up studies, qualitative studies, mixed methods studies, time series, narrative reviews, consensus documents, letters, comments, editorials, meeting abstracts.

Inclusion Criteria
Published studies were included if they met each of the following criteria:

1. Prospectively or retrospectively evaluated either:
    a. Effective protocols for collection of effusions/pleural fluids, fine needle aspirate (FNA), endobronchial ultrasound guided-FNA (EBUS-FNA), core needle biopsy (CNB), and touch preparation specimens to optimize ancillary testing
    b. Effective protocols for handling and processing of effusions/pleural fluids, fine needle aspirate (FNA), endobronchial ultrasound guided-FNA (EBUS-FNA), core needle biopsy (CNB), and touch preparation specimens to optimize ancillary testing
2. Enrolled patients with suspected or undiagnosed thoracic abnormality including:
    a. Lung cancer
    b. Extrathoracic metastatic disease
    c. Sarcoidosis
    d. Thymoma
    e. Granuloma

   f. Spindle cell neoplasm
   g. Infections including tuberculosis, pneumocystis, fungus (aspergillus)
   Note: Extrathoracic metastatic disease and hematologic processes were excluded from this review as CAP is currently addressing hematolymphoid processes in another guideline
3. Included one of the outcomes of interest as outlined in the section below.
4. Peer-reviewed full-text articles.

Exclusion Criteria

1. Studies enrolling patients with specimen type of brushes, washes, bronchoalveolar lavages, unsatisfactory specimens, including lung specimens that have undergone ablation.
2. Studies of less than 30 samples or patients per study arm.
3. Studies of an excluded study type design as defined above.

### Outcomes of Interest

The EP deemed the following as outcomes of interest: diagnostic yield, specimen adequacy/sufficiency for ancillary testing, cellularity, volume of specimen, number of specimens collected, diagnostic concordance, ancillary testing success rates, number of procedures, and complications.

### 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.

### Literature Review and Analysis

The EP met 15 times through teleconference webinars from August 4, 2015, through May 23, 2019. Additional work was completed via electronic mail. The panel met in person March 18, 2017, December 2, 2017, and March 9-10, 2018 to review evidence from the systematic review and draft recommendations.

The EP formed the following key questions (KQs) for which to base the literature search:

1. With regard to each of the specimen types of interest (effusions/pleural fluids, FNA, endobronchial ultrasound guided-FNA [EBUS-FNA], CNBs, what evidence is available to determine the most effective protocols for sample collection, including:
   a. immediate handling of the specimen, including:
      - how the needle biopsy is expelled from needle
      - selection of the appropriate media
   b. the number (minimum and maximum) of passes needed to ensure that the laboratory can obtain adequate materials for diagnostic testing
   c. the impact of rapid onsite evaluation (ROSE) on adequacy, quality, and triage of specimens

2. With regard to each of the specimen types, the preparations created, and the tests to be performed, what evidence is available to determine the most effective methods for the handling and processing of specimens, including:
   a. the selection of appropriate media (e.g., liquids, tissue clot coagulum, fixed or unfixed) and the priority by disease
   b. the optimal ischemic time (i.e., time between tissue removal and initiation of fixation)

3. With regard to the various pathologic testing methods, what evidence is available to support an algorithm(s) for selection of specimens and sequence of testing, under defined circumstances?

All EP members participated in the systematic evidence review (SER): title-abstract screening, full-text review, and data extraction. A dual review was performed for each study and in each phase of the SER; the co-chairs and/or methodologist adjudicated all conflicts. A literature refresh was also conducted, where studies also underwent dual review. A total of 100 studies comprised the final body of studies included in the SER. Supplemental Figure 2 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.

## Peer Review

A public, open access comment period was held from May 23 through June 15, 2018 on the CAP Web site www.cap.org for any interested stakeholder to provide feedback on the draft recommendations. Seventeen draft recommendations, two demographic questions, three questions to assess feasibility, and one area to capture general comments were posted for feedback. An announcement was sent to the following societies deemed stakeholders:

**Medical Societies**

- American Association for Clinical Chemistry (AACC)
- American Association of Pathologists' Assistants (AAPA)
- American College of Chest Physicians (CHEST)
- American College of Radiology (ACR)
- American Society for Clinical Oncology (ASCO)
- American Society for Clinical Pathology (ASCP)
- American Society for Cytopathology (ASC)
- American Society of Cytotechnologists (ASCT)
- American Thoracic Society (ATS)
- Association for Molecular Pathology (AMP)
- 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)
- Cardiovascular and Interventional Radiological Society of Europe (CIRSE)
- European Federation of Cytology Societies (EFCS)
- European Society for Medical Oncology (ESMO)
- European Society of Pathology
- International Academy of Pathology (IAP)
- International Association for the Study of Lung Cancer (IASLC)
- International Academy of Cytology (IAC)
- National Comprehensive Cancer Network (NCCN)
- Papanicolaou Society of Cytopathology (PSC)
- Pulmonary Pathology Society (PPS)
- Radiological Society of North America (RSNA)
- Royal College of Pathologists
- Society for Thoracic Radiology (STR)

- Society of Interventional Radiology (SIR)
- United States & Canadian Academy of Pathology (USCAP)

**Government**

- US Food and Drug Administration (FDA)
- Centers for Medicare & Medicaid Services (CMS)
- Veteran's Affairs (VA) and Department of Defense (DOD)
- Centers for Disease Control and Prevention (CDC), Division of Laboratory Programs, Standards, and Services
- CDC, Centers for Medicare and Medicaid Services (CMS)
- NIH, Division of Cancer Treatment and Diagnosis
- NIH, National Health Council
- European Medicines Agency
- National Institute for Health and Care Excellence (NICE) (UK)

**Patient Advocacy Groups**

- Alpha-1 Foundation
- LAM Foundation
- American Lung Association
- Free to Breathe (formerly: National Lung Cancer Partnership)
- Lung Cancer Foundation of America (LCFA)
- Uniting Against Lung Cancer / Lung Cancer Research Foundation
- Addario Lung Cancer Medical Institute (ALCMI "Alchemy")
- Bonnie J. Addario Lung Cancer Foundation (ALCF)
- Caring Ambassadors Lung Cancer Program
- Dusty Joy Foundation
- Free Me From Lung Cancer
- Global Lung Cancer Coalition
- International Thoracic Oncology Nursing Forum
- Lung Cancer Alliance
- Lungevity Foundation
- Roy Castle Lung Cancer Foundation
- Women Against Lung Cancer in Europe

Two hundred eighty-seven individuals participated in the comment period. "Agree," "Disagree," and "Does not pertain to my area of expertise or practice" responses were captured for every proposed recommendation. The website also received 469 written comments. Ten draft recommendations had less than 10% disagreement, five had between 10-20% disagreement, and two had 20-30% disagreement. Each EP member was assigned one to two draft statement for which they reviewed the comments and presented them to the entire panel for group discussion. After consideration of the comments, seven draft recommendations were maintained with the original language and ten were revised. Resolution of all changes was obtained by majority consensus of the panel using nominal group technique (discussion at an in-person meeting, rounds of teleconference webinars, email discussion and multiple edited recommendations) amongst the panel members. The final recommendations were agreed upon by the EP with a formal vote. The panel considered laboratory efficiency and feasibility throughout the entire

considered judgment process. Fifty-seven and fifty-eight hundredths percent (57.58% [114 of 198]) responded that the entire guideline was feasible, 40.91% (81 of 198) responded that parts of it were feasible, and 1.52% (3 of 198) responded that none of it was feasible. Neither formal cost analysis nor cost effectiveness models were performed.

An independent review panel (IRP) was assembled to review and approve the guideline on behalf of the CAP Council on Scientific Affairs. The IRP was masked to the EP and to each other and were vetted through the COI process.

### Risk of Bias Assessment Methods

A risk of bias assessment was performed for all retained studies following application of the inclusion and exclusion criteria. Using this method, studies deemed 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. 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.

Clinical practice guidelines (CPGs)

- The following attributes were considered as per the AGREE II[2] tool using a 7-point scale:
  1. Scope and purpose
  2. Stakeholder involvement
  3. Rigor of development
  4. Clarity of presentation
  5. Applicability
  6. Editorial independence

Systematic Reviews (SRs) and Meta-analyses (MAs)

- The following questions were assessed as per the Assessing the Methodological Quality of Systematic Reviews (AMSTAR[3]) tool using yes, no, or unclear:
  1. Was an ‘a priori’ design provided?
  2. Was there duplicate study selection and data extraction?
  3. Was a comprehensive literature search performed?
  4. Was the status of publication (i.e. grey literature) used as an inclusion criterion?
  5. Was a list of studies (included and excluded) provided?
  6. Were the characteristics of the included studies provided?
  7. Was the scientific quality of the included studies assessed and documented?
  8. Was the scientific quality of the included studies used appropriately in formulating conclusions?
  9. Were the methods used to combine the findings of studies appropriate?
  10. Was the likelihood of publication bias assessed?
  11. Was the conflict of interest included?
- Additional assessed items included and were assessed as yes, no, or unclear:
  1. Reporting of funding sources.

Randomized Control Trials (RCTs)

- The following domains were assessed using the Cochrane Risk of Bias tool[4] using low risk, unclear risk, and high risk:
  1. Random sequence generation (selection bias)
  2. Allocation concealment (selection bias)
  3. Blinding of participants and personnel (performance bias)
  4. Blinding of outcome assessment (detection bias – patient-reported outcomes)
  5. Incomplete outcome data (attrition bias)
  6. Selective outcome reporting (reporting bias)
- Additional assessed items included and were assessed as yes, no, or unclear:
  1. Validated and reliable measures used

2. Adequately powered statistical analysis
3. Reporting of funding sources
4. Industry funding

Prospective cohort studies (PCS), retrospective cohort studies (RCS), and case-control studies (CCS)

- The following domains were assessed using the ROBINS-I tool[5] using low risk, moderate risk, serious risk, critical risk, or unclear:
    1. Confounding
    2. Patient selection (selection bias)
    3. Intervention classification (performance bias)
    4. Deviation from intended intervention (performance bias)
    5. Missing data (reporting bias)
    6. Outcome measurements (detection bias)
    7. Selection of reported outcomes (detection bias)
- Additional assessed items included and were assessed as yes, no, or unclear:
    1. Adequately powered statistical analysis
    2. Reporting of funding sources
    3. Industry funding

### Quality Assessment Results

A total of 218 studies were included in our systematic review with the evidentiary base supporting the recommendation statements consisting of 100 studies. This body of evidence comprised one meta-analysis, three systematic reviews, three RCTs, 34 PCSs, and 59 RCSs. In the following sections, the number of studies that met our inclusion criteria and were retained, the evidence type as determined by study design, the aggregate quality of that evidence as determined by the risk of bias assessment of the individual studies and consistency of results are reported. Aggregate risk of bias across studies informing a statement are defined as not serious, serious, very serious, or extremely serious.

Following the risk of bias assessment, each guideline statement was given a grade for quality of evidence. Quality of evidence is graded as High, Moderate, Low, Very Low, or Insufficient, based on our confidence in how closely the reported estimate of effect reflects the true effect (Supplemental Table 1). During the development process of this guideline, the CAP transitioned to following Grading of Recommendation Assessment, Development and Evaluation (GRADE) methodology. Although it was too late in development to use GRADE for this guideline, future updated will reflect this change. In order to make the transition more seamless, GRADE definitions for quality of evidence were adopted in this version of the guideline.

### Assessing the Strength of Recommendations

Development of recommendations required that the panel review the identified evidence and make a series of key judgments:

1. What are the significant findings related to each KQ or outcome? Determine any regulatory requirements and/or evidence that support a specific action.
2. What is the overall quality of evidence supporting each KQ or outcome?
3. What is the strength of each recommendation? Supplemental Table 2 describes the ratings for the strength of recommendation, based on the quality of evidence and the likelihood that further studies will change the conclusions. Recommendations not supported by evidence (i.e., evidence was missing or insufficient to permit a conclusion to be reached) were made based on consensus expert opinion. Another potential consideration is the likelihood that additional studies will be conducted that fill gaps in knowledge.

4. What is the net balance of benefits and harms? The panel used the Evidence to Decision Framework[6] to frame, discuss, and document their decisions for each recommendation.

## Strength of Recommendation, Quality of Evidence, Risk of Bias Assessment, and Summary of the Benefits and Harms by Guideline Statement

**Statement 1. *Strong Recommendation*. – Endobronchial ultrasound guided transbronchial needle aspiration (EBUS TBNA) may be used, if available, for initial evaluation (diagnosis, staging, identification of recurrence/metastasis) of mediastinal and hilar lymph nodes, as well as centrally located parenchymal lesions visible with endobronchial ultrasound.**
The quality of evidence is *moderate* to support this guideline statement. Refer to Supplemental Table 3a and 3b for the quality assessment results of the 15 studies[7-21] informing this statement. A meta-analysis[18] at low risk of bias focused on the ability of EBUS TBNA to provide an adequate sample for ancillary testing. The remaining 14 studies carried an aggregate serious risk of bias and included, 11 RCSs[7-13,15-17,21] and three prospective studies.[14,19,20] As a consequence of their retrospective design, all retrospective studies[7-13,15-17,21] suffered from selection bias, additional individual studies carried a risk of bias in performance domains,[13] reporting domains,[8,11-13,15-17,21] and detection domains,[7-13,15-17,21] while five studies did not report funding source.[8,9,13,15,16] The PCSs were limited by risk of selection bias in all,[14,19,20] reporting bias in two,[19,20] and detection bias in one.[14] None of the studies were found to have methodological flaws that would raise concerns about the findings.

There is balance of effects between the benefits of using EBUS TBNA in these locations and the harms. The panel believes this statement may be acceptable to key stakeholders and will be feasible to implement in some institutions.

**Statement 2. *Recommendation.* – When performing EBUS TBNAs, 19-, 21-, or 22-gauge needles may be used.**
The quality of evidence is *low*. Refer to Supplemental Table 4a and 4b for the quality assessment results of nine studies[22-30] included in the evidentiary base of statement 2. An included SR[24] carried a low risk of bias with the only limitation being unclear reporting surrounded whether the study used dual study selection and data extraction. Additionally, two RCTs,[26,28] two PCSs,[22,27] and four RCS[23,25,29,30] were published after the search date of the SR and included to inform this statement. The aggregate risk of bias across these studies was very serious. An additional prospective cohort study,[22] which did not evaluate needle gauge in relation to sufficiency for ancillary testing but did report on the outcome, was included as indirect evidence for this statement and was limited by a serious risk of bias. None of the studies were found to have methodological flaws that would raise concerns about the findings.

Studies from the systematic review showed that 19-, 21-, and 22-gauge needles attribute diagnostic accuracy and yield sufficient specimen cellularity for diagnosis in EBUS TBNAs. The panel recognizes however, that other needle gauges may also be employed successfully. The panel believes the guideline statement to be feasible and acceptable to most key stakeholders.

***Statement 3. Recommendation*. – When performing EBUS TBNA, ROSE should be utilized, if available.**
The quality of evidence is *moderate*. The evidence base informing this statement comprised one SR,[24] one RCT,[31] six PCSs,[20,32-36] and 13 RCSs.s[10,13,15,37-46] Refer to Supplemental Table 5a,5b for the quality assessment results of all included studies. The aggregate risk of bias across the 21 studies was serious based on poor methodology reporting,[32] and risk of selection bias,[32-34,36] detection bias,[32-34,36] performance bias,[35] and reporting bias[32,34,36] in the PCSs. The RCSs carried risk of selection bias inherent in retrospective studies, in addition to detection bias,[10,13,15,38,40-46] performance bias,[13,46], reporting bias[13,15,38-41,45,46] and no reporting of funding source.[13,15,38-41,44,45] None of the studies were found to have methodological flaws that would raise concerns about the findings.

Although the feasibility of this recommendation varies based on available resources at individual centers, the panel believes that the benefits of using ROSE outweigh the harms. In general, ROSE potentially helps maximize diagnostic yield, minimizes complications, and helps triage specimens more readily. ROSE requires dedicated cytology personnel and inter-department coordination. It also adds additional cost depending on who provides the assessment (cytotechnologist versus pathologist). Nonetheless, the panel believes the guideline statement to be acceptable, especially to stakeholders with ROSE services in place.

**Statement 4. *Recommendation*. – To achieve optimal diagnostic yield, when performing EBUS TBNA without ROSE, the bronchoscopist should perform at a minimum three and up to five passes, if technical and clinically feasible. When performing with ROSE, clinical judgment should be used to assess the number of passes needed. Additional passes may be required for ancillary studies.**

The quality of evidence is *moderate*. Refer to Supplemental Table 6 for the risk of bias assessment results of six studies[24,31,47-50] included in the evidentiary base for this statement. Studies carried an aggregate serious risk of bias and included one SR,[24] one RCT,[31] three PCSs,[47,49,50] and one RCS.[48] The RCT[31] suffered from selection bias, performance bias and detection bias, while the PCSs were limited by risk of reporting bias,[47] selection bias,[49] or performance bias.[50] In addition to high risk of selection bias, the retrospective cohort study[48] also suffered from a risk of detection bias and did not report on funding sources. None of the studies were found to have methodological flaws that would raise concerns about the findings.

Although the panel recommends the use of ROSE, it is understood that its use may not always be feasible. Based on a lack of data indicating increased adverse events with multiple EBUS TBNA passes, the recommended intervention of multiple passes favors benefits of obtaining sufficient material for ancillary testing over the harms. The panel believes this guideline statement to be acceptable to key stakeholders.

**Statement 5. When performing transthoracic needle procedures, ROSE should be used for adequacy assessment, if available. If performing CNB, without concurrent FNA, touch preparations may be used for adequacy assessment, if available. - *Strong Recommendation for the use of ROSE for adequacy assessment; Recommendation for the use of touch preparations without concurrent FNA.***

The quality of evidence for the use of ROSE is *moderate*, while the quality of evidence for the use of a touch preparation is *low*. Refer to Supplemental Table 7 for the risk of bias assessment results of all eight studies[24,36,44,48,51-54] included in the evidentiary base. The seven studies reporting on diagnostic yield and/or adverse events using ROSE carried a serious aggregate risk of bias. Included studies were limited by their risk of bias in relation to patient selection,[36,44,48,52,54] performance,[54] detection,[36,48,54] and reporting,[36,54] as well as a lack of reported funding.[36,44,48] Evidence supporting the use of touch preps for adequacy assessment comprises one PCS[53] and one RCS[52] with an aggregate very serious risk of bias. None of the studies were found to have methodological flaws that would raise concerns about the findings.

Although the feasibility of this recommendation varies based on available resources at individual institutions, the panel believes that the benefits of using ROSE outweigh the harms. In instances where CNBs are performed without concurrent FNA, the panel believes that touch preparations can provide value. The panel recognizes, however, that the adequacy of touch preparations can be variable, and as such, some stakeholders may not find them to be acceptable for their practice.

**Statement 6. *Recommendation*. – When performing transthoracic needle procedures, needle size should be determined by the operator and technique. For transthoracic FNAs, needles as small as 25 gauge may be used. For CNBs, needles as small as 20 gauge may be used.**

The quality of evidence is *low*. Refer to Supplemental Table 8 for the risk of bias assessment results of all 12 studies[53,55-65] informing this statement. Evidence supporting this statement is based on studies that evaluated adequacy using various needle gauges and studies that reported on adverse events using various needle gauges. Of the two studies reporting on tissue adequacy following specimen collection, one study was an MA,[56] while the other study was a RCS.[55] Although based on a SR, the MA[56] did not include an a prior design, duplicate study selection or data extraction, a complete list of included and excluded studies, or quality assessment of the included studies. Studies reporting on adverse events included six PCSs,[53,59,61-64] and five RCSs.[55,57,58,60,65] These studies were limited by their risk of bias to selection bias,[53,57-65] performance bias,[53,64] detection bias,[53,58,60,61,63,65] and reporting bias,[53,57,58,60-63,65] as well as a lack of reported funding.[53,58,61-65] None of the studies were found to have methodological flaws that would raise concerns about the findings.

The panel weighed the harms of using a large gauge needle against the benefits of obtaining sufficient material for ancillary testing with smaller needle gauges as there were no studies that directly compared needle gauges in terms of adequacy for ancillary testing nor adverse events. The panel believes that the guideline statement is feasible and acceptable to key stakeholders.

**Statement 7. *Recommendation.* - When performing transthoracic FNA without CNB, the proceduralist should obtain multiple passes, if technically and clinically feasible, and should attempt to collect sufficient material for a tissue block (i.e., cell block, tissue clot).**
The quality of evidence is *low*. Refer to Supplemental Table 9 for the risk of bias assessment results of five studies that evaluated diagnostic yield and adequacy outcomes using multiple passes,[36,54,66-68] two studies reported on adverse events of multiple passes,[36,54] and eight studies reporting on diagnostic yield and adequacy when a tissue block was created.[53,69-75] The aggregate risk of bias across the 13 studies was very serious. The five studies reporting on diagnostic yield and adequacy for multiple passes included three PCSs[36,67,68] and two RCSs.[54,66] Of the studies reporting on multiple passes, one prospective cohort study[36] and one retrospective cohort study[68] also reported on adverse events. All studies reporting on diagnostic outcomes suffered from risk of bias in relation to patient selection,[36,54,66-68] additionally four studies were limited by detection bias,[36,54,66,67] four contained missing data,[36,54,66,68] and two did not report on funding source.[36,66] Of the eight studies reporting on the creation of tissue blocks, three were PCSs[53,71,76] and five were RCSs. [69,70,72,73,75] Included studies were limited by their risk of bias in relation to patient selection, [53,69-75] performance,[53,72] detection,[53,70,72-74] and reporting,[53,69-74] as well as a lack of reported funding.[53,73,74] None of the studies were found to have methodological flaws that would raise concerns about the findings.

Based on a lack of data indicating increased adverse events with multiple passes, the recommended intervention of multiple passes favors benefits of obtaining sufficient material for ancillary testing over the harms. The panel believes this statement to be feasible and acceptable to key stakeholder. As this is regular practice in many institutions, the guidance should also be highly implementable.

**Statement 8. *Recommendation*. - To achieve optimal diagnostic yield when performing transthoracic CNBs, the proceduralist should attempt to obtain a minimum of three core samples, if technically and clinically feasible. Additional samples may be required for ancillary studies.**
The quality of evidence is *low*. Refer to Supplemental Table 10 for complete risk of bias assessment results of one RCS[77] supporting this statement. The study was limited by a risk of study selection bias, as well as a risk of detection bias.

Based on a low rate of adverse events with collection of three of more samples, the benefits of obtaining sufficient material for ancillary testing outweighs the harms. The panel believes this guideline statement to be feasible as this is regular practice in some institutions. Additionally, the guidance is believed to be acceptable to key stakeholders.

**Statement 9. *Recommendation* - If performing bronchoscopy for the investigation of peripheral pulmonary lesions that are difficult to reach with conventional bronchoscopy, image-guidance adjuncts may be used, if local expertise and equipment are available.**
The quality of evidence is *low*. Refer to Supplemental Table 11 for the risk of bias assessment results of PCS[78] and RCS,[79] both of which informed this statement. The aggregate risk of bias was very serious based on risk of selection bias and detection bias in both studies.[78,79] Neither study was found to have methodological flaws that would raise concerns about the findings.

The benefits of increased diagnostic yield with image-guidance are large and outweigh the very small harms of using image-guidance adjuncts. The panel believes this guideline statement to be feasible and acceptable to key stakeholders.

**Statement 10. When performing transbronchial needle aspirates, ROSE should be used for adequacy assessment, if available. If performing transbronchial forceps biopsies, without concurrent FNA, touch preparations may be used for adequacy assessment, if available. – *Recommendation for the use of ROSE for adequacy assessment; Expert Consensus Opinion for the use of touch preparations.***
The quality of evidence supporting the use of ROSE is *moderate,* while the quality of evidence supporting the use of touch preparations is *very low*. Refer to Supplemental Table 12 for the risk of bias assessment results of all studies informing this statement. The evidence base for the use of ROSE comprises seven studies,[80-86] including one SR,[80] three PCSs,[84-86] and three RCSs.[81-83] The aggregate risk of bias was serious and included cohort studies were limited by their risk of bias in relation to patient selection,[81-84,86] performance,[82,83,86] detection,[82-84,86] and reporting,[82-86] as well as a lack of reported funding.[82,86] Evidence supporting the use of touch preparations for adequacy assessment comprises one PCS,[53] which was limited by risk of bias related to patient selection, performance, detection and reporting bias domains. None of the studies were found to have methodological flaws that would raise concerns about the findings.

Although the feasibility of this recommendation varies based on available resources at individual institutions, the benefits of using ROSE outweigh the harms. The panel believes this statement to be acceptable to key stakeholders. The panel recognizes, however, that the adequacy of touch preparations can be variable, and as such, some stakeholders may not find them acceptable for their practice.

**Statement 11. *Expert Consensus Opinion.* – When collecting pleural fluid for a suspected diagnosis of malignancy, the proceduralist should send as much fluid volume as reasonably attainable for cytologic evaluation and ancillary studies.**
The quality of evidence is *insufficient*. Refer to Supplemental Table 13 for the risk of bias assessment results of studies informing this statement. Although three PCSs[87-89] and four RCSs[90-93] reported on minimum volume of fluid required for a diagnosis, there was too much heterogeneity within the identified studies upon which to base an evidence-based statements. Additionally, no studies reported on an upper limit of fluid. None of the studies were found to have methodological flaws that would raise concerns about the findings.

Although a few studies addressed pleural fluid volume, the panel believes the benefit of sending "more" fluid to the cytology laboratory outweighs the harms of sending "less." Implementing this guideline statement affords the laboratory greater flexibility to perform more tests. Minor costs exist for sending larger amounts of fluids (cost of collection bags) but is relatively miniscule. Laboratory staff will also have to discard excess fluid.

**Statement 12. *Strong Recommendation.* – Cytology specimens (smears, cell blocks, liquid based cytology), may be used for ancillary studies if supported by adequate validation studies.**
The quality of evidence is *low*. Refer to Supplemental Table 14 for the risk of bias assessment results of all 10 studies[39,42,69,70,94-99] used to inform this statement. The aggregate risk of bias across the 10 studies was very serious. Of the 10 studies, two RCSs reported on the use of

cytology specimens for IHC,[95,99] one PCS[96] and one RCS[99] reported on the use of cytology specimens for FISH, and seven RCSs reported on cytology specimens when conducting molecular analysis.[39,42,69,70,94,97,98] Included studies were limited by risk of bias in selection,[39,42,69,70,94-96,98,99] performance,[94,95,97-99] detection,[42,70,99] and reporting[39,70,94,95,98] domains. None of the studies were found to have methodological flaws that would raise concerns about the findings.

The benefits of using cytology specimens for ancillary testing outweigh any perceived harms from testing in these specimens. The panel believes that implementation of this guidance will result in resource savings and that the statement is both feasible and acceptable to key stakeholders.

**Statement 13. Recommendation - CNB specimens collected for ancillary studies should be fixed in 10% neutral buffered formalin.**
The quality of evidence is *very low*. Refer to Supplemental Table 15 for the risk of bias assessment results of studies included in the evidentiary base for this statement. Although no study directly compared the use of one fixative with another, four studies[59,100-102] reporting on adequacy for ancillary testing or successful ancillary testing following fixation with 10% neutral buffered formalin comprise the indirect evidence supporting this statement. Included studies carried and aggregate very serious risk of bias and were limited by their risk of bias in relation to patient selection,[59,100-102] detection,[100,101] and reporting[100,101] domains, as well as a lack of reported funding.[100,102] None of the studies were found to have methodological flaws that would raise concerns about the findings.

Ten-percent neutral buffered formalin renders CNB specimens adequate for molecular testing and is generally the standard of practice. As such, the benefits of using it outweigh the harms and most stakeholders will have already be implementing this guideline statement.

**Statement 14. *Recommendation.* – When performing bronchoscopy for the investigation of tuberculosis, endobronchial ultrasonography may be used to increase the diagnostic yield of bronchoalveolar lavage and transbronchial biopsy.**
The quality of evidence is *low*. Refer to Supplemental Table 16 for complete quality assessment of the one RCS[103] informing this statement. The study was limited by risk of selection bias, performance bias, reporting bias, and detection bias. The study did not contain methodological flaws that would raise concerns about its findings.

Many hospitals already employ EBUS TBNA outside the investigation of tuberculosis so using it for this purpose is an extension of resources already in place. A comparative study from the systematic review show increased diagnostic yield and decreased time to diagnosis when compared to convention bronchoscopy. The panel believes that stakeholders will find the guideline statement acceptable.

**Statement 15. *Recommendation.* – When performing EBUS TBNA for the evaluation of intrathoracic granulomatous lymphadenopathy with the suspicion of tuberculosis, specimens should be collected for cytology, microbiology (mycobacterial smear and culture), and *Mycobacterium tuberculosis*-polymerase chain reaction (TB-PCR) evaluation, if available.**
The quality of evidence is *low*. Refer to Supplemental Table 17 for the quality assessment results of three RCSs[104-106] identified to inform this statement. Studies were limited by a very serious aggregate risk of bias and carried risk of bias in patient selection,[104-106] performance,[104] detection,[104-106] and reporting domains,[104-106] as well as a lack of reported funding.[104-106] None of the studies were found to have methodological flaws that would raise concerns about the findings.

The benefits of implementing the guideline statement allow for a more thorough diagnostic workup, providing complementary information to inform a diagnosis. Evidence from the

systematic review confirm that the combination of cytology, microbiology and TB-PCR increases diagnostic yield. In addition, as certain tests provide diagnostic information faster than others, sending specimens to all three laboratories can assure the appropriate triage of patients. Sending specimens for various evaluations will incur costs and might not be feasible in lower income settings.

**Statement 16. *Recommendation.* – When collecting pleural fluid for diagnosis of extrapulmonary tuberculosis, specimens should be submitted for microbiology culture studies for mycobacteria using liquid media protocol.**
The quality of evidence is *low.* Refer to Supplemental Table 18 for complete quality assessment of the one PCS[76] informing this statement. The PCS suffered from risk of reporting and detection bias but contained no methodological flaws that would raise concerns about the findings.

Liquid media protocol is recommended for its high analytic sensitivity and ability to render timely identification of patients with tuberculosis. This is supported by both evidence from the systematic review and in various recommendations from other professional medical societies and laboratory standard documents.

## Dissemination Plans

The CAP plans to host a Collection and Handling of Thoracic Small Biopsy and Cytology Specimens for Ancillary Studies web page which will include a link to the manuscript and supplement; a summary of the recommendations, a PowerPoint slide deck (Microsoft Corporation, Redmond, WA), a frequently asked question (FAQ) document, and an infographic. The guideline will be promoted and presented at various society meetings.

**Supplemental Table 1: Grades for Quality of Evidence**

| Grade | Definition | Aggregate Risk of Bias |
|---|---|---|
| High | We are very confident that the true effect lies close to that of the estimate of effect | Low Risk |
| Moderate | We are moderately confident in the effect estimate: The true effect is likely to be close to the estimate of effect, but there is a possibility that it is substantially different | Moderate/Serious Risk |
| Low | Our confidence in the effect estimate is limited: The true effect may be substantially different from the estimate of effect | Serious/Critical Risk |
| Very Low | We have very little confidence in the effect estimate: The true effect is likely substantially different from the estimate of effect | Critical Risk |
| Insufficient | Evidence is insufficient to discern net effect. Any estimate of effect is very uncertain. | Insufficient evidence and expert panel uses formal consensus process to reach recommendation |

Adapted from *J Clin Epidemiol*, 64(4), Balshem H, Helfand M, Schunemann HJ, et al. GRADE guidelines: 3. Rating the quality of evidence, p. 401-406, copyright 2011, with permission from Elsevier.[107]

**Supplemental Table 2: Grades for Strength of Recommendations**

| Designation | Recommendation | Rationale |
|---|---|---|
| **Strong Recommendation** | Recommend For or Against a particular practice (Can include “must” or “should”) | Supported by High or Moderate quality of evidence and clear benefit that outweighs any harms |
| **Recommendation** | Recommend For or Against a particular practice (Can include “should” or “may”) | Some limitations in quality of evidence (Moderate – Very |

| | | |
|---|---|---|
| | | Low), balance of benefits and harms, values, or costs but panel concludes that there is sufficient evidence to inform a recommendation |
| **Expert Consensus Opinion** | Recommend For or Against a particular practice (Can include “should” or “may”) | Serious limitations in quality of evidence (Very Low or Insufficient), balance of benefits and harms, values or costs, but panel consensus is that a statement is necessary |
| **No Recommendation** | No Recommendation For or Against a particular practice | Insufficient evidence to provide a recommendation, balance of benefits and harms, values or costs |

Derived from Andrews et al.[108]

**Supplemental Table 3a. Risk of Bias Assessment Results for Statement 1**

| Systematic Reviews | | | Cohort Studies | | PCS | | |
|---|---|---|---|---|---|---|---|
| Study | | Labarca[18] 2018 | Study | | Nosotti[14] 2009 | Nair[19] 2018 | Chaiyakul[20] 2018 |
| AMSTAR Assessment | A priori design | Y | ROBINS-I Assessment | Confounding | MR | MR | MR |
| | Duplicate study selection and data extraction | Y | | Patient selection | MR | MR | MR |
| | Comprehensive literature search | Y | | Intervention classification | LR | LR | LR |
| | Publication status as inclusion criterion | N | | Deviation from intended intervention | LR | LR | LR |
| | List of included and excluded studies | Y | | Missing data | MR | MR | MR |
| | Characteristics of included studies | Y | | Outcome measurements | LR | LR | MR |
| | Study quality assessment conducted | Y | | Selection of reported outcomes | MR | LR | LR |
| | Quality assessment used in formulating conclusions | Y | | Overall Risk of Bias | MR | MR | MR |
| | Appropriate methods to combine findings | Y | Adequately powered | | Y | Y | Y |
| | Publication bias assessment | N | Reported funding sources | | N | Y | Y |
| | Conflict of interest reported | Y | Industry funded | | U | N | N |
| Reported funding sources | | Y | Study Quality | | Int-Low | Int | Int |
| Study Quality | | High | | | | | |

Abbreviations: Int, intermediate; Int-Low, intermediate-low; LR, low risk; MR, moderate risk; N, no; PCS, prospective cohort study; U, unclear/unsure; Y, yes.

**Supplemental Table 3b. Risk of Bias Assessment Results for Statement 1**

| Cohort Studies | | RCS | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Study | | Vaidya[7] 2016 | Wang[8] 2014 | Zhao[9] 2013 | Joseph[10] 2013 | Kennedy[12] 2010 | Gilbert[15] 2009 | Nakajima[16] 2008 | Gilbert[13] 2009 | Han[11] 2013 | Hu[21] 2018 | Nambirajan[17] 2019 |
| ROBINS-I Assessment | Confounding | MR | MR | MR | MR | MR | MR | MR | MR | MR | SR | MR |
| | Patient selection | CR | CR | CR | CR | CR | CR | CR | CR | CR | CR | CR |
| | Intervention classification | LR | LR | LR | LR | LR | LR | LR | MR | LR | LR | LR |
| | Deviation from intended intervention | LR | LR | LR | LR | LR | LR | LR | MR | LR | LR | LR |
| | Missing data | LR | MR | LR | LR | MR | SR | MR | MR | MR | CR | CR |
| | Outcome measurements | MR | SR | MR | MR | MR | MR | MR | MR | MR | SR | MR |
| | Selection of reported outcomes | MR | LR | LR | MR | LR | MR | LR | MR | MR | LR | MR |
| | Overall Risk of Bias | CR | CR | CR | CR | CR | CR | CR | CR | CR | CR | CR |
| Adequately powered | | Y | Y | N | Y | Y | Y | N | Y | Y | Y | Y |
| Reported funding sources | | Y | N | N | Y | Y | N | N | N | Y | Y | Y |
| Industry funded | | N | U | U | N | N | U | U | U | Y | N | N |
| Study Quality | | Low | Very Low | Very Low | Low | Low | Very Low | Very Low | Very Low | Very Low | Very Low | Very Low |

Abbreviations: CR, critical risk; LR, low risk; MR, moderate risk; N, no; RCS, retrospective cohort study; SR, serious risk; U, unclear/unsure; Y, yes.

**Supplemental Table 4a. Risk of Bias Assessment Results for Statement 2**

| Systematic Reviews | | | Randomized Controlled Trials | | | |
|---|---|---|---|---|---|---|
| Study | | Van der Heijden[24] 2014 | Study | | Wolters[26] 2019 | Dooms[28] 2018 |
| AMSTAR Assessment | A priori design | Y | Cochrane Risk of Bias | Random sequence generation | HR | LR |
| | Duplicate study selection and | U | | Allocation concealment | HR | LR |

| Systematic Reviews | | | Randomized Controlled Trials | | | |
|---|---|---|---|---|---|---|
| Study | | Van der Heijden[24] 2014 | Study | | Wolters[26] 2019 | Dooms[28] 2018 |
| | data extraction | | | | | |
| | Comprehensive literature search | Y | | Blinding – patients and conductors | HR | HR |
| | Publication status as inclusion criterion | Y | | Blinding – outcome assessors | HR | LR |
| | List of included and excluded studies | Y | | Complete outcome data | LR | LR |
| | Characteristics of included studies | Y | | Selective outcome reporting | LR | HR |
| | Study quality assessment conducted | Y | | Overall Risk of Bias | HR | IR |
| | Quality assessment used in formulating conclusions | Y | Validated and reliable measures | | Y | Y |
| | Appropriate methods to combine findings | Y | Adequately powered | | Y | Y |
| | Publication bias assessment | Y | Reported funding sources | | N | Y |
| | Conflict of interest reported | Y | Industry funded | | U | N |
| Reported funding sources | | Y | Study Quality | | Int | High-Int |
| Study Quality | | High | | | | |

Abbreviations: HR, high risk; Int, intermediate; IR, intermediate risk; LR, low risk; N, no; U, unclear/unsure; Y, yes.

**Supplemental Table 4b. Risk of Bias Assessment Results for Statement 2**

| Cohort Studies | | PCS | | RCS | | | |
|---|---|---|---|---|---|---|---|
| Study | | Pickering[27] 2018 | Jeyabalan[22] 2016 | Jeyabala[23] 2014 | Tyan[25] 2017 | Garrison[29] 2018 | Kinoshita[30] 2018 |
| ROBINS-I Assessment | Confounding | MR | MR | MR | MR | MR | MR |
| | Patient selection | SR | LR | CR | CR | CR | CR |
| | Intervention classification | LR | LR | LR | LR | LR | LR |
| | Deviation from intended intervention | MR | LR | LR | LR | LR | LR |
| | Missing data | MR | LR | LR | MR | MR | MR |
| | Outcome measurements | LR | LR | MR | MR | MR | MR |
| | Selection of reported outcomes | MR | LR | MR | LR | LR | LR |
| | Overall Risk of Bias | SR | MR | CR | CR | CR | CR |
| Adequately powered | | Y | Y | Y | Y | Y | Y |
| Reported funding sources | | Y | N | N | N | Y | Y |
| Industry funded | | N | U | U | U | N | N |
| Study Quality | | Int-Low | Int | Low | Low | Low | Low |

Abbreviations: CR, critical risk; Int, intermediate; LR, low risk; MR, moderate risk; N, no; PCS, prospective cohort study; SR, serious risk; RCS, retrospective cohort study; U, unclear/unsure; Y, yes.

**Supplemental Table 5a. Risk of Bias Assessment Results for Statement 3**

| Systematic Reviews | | | Randomized Controlled Trials | | | Cohort Studies | | PCS | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Study | | Van der Heijden[24] 2014 | Study | | Trisolini[31] 2015 | Study | | Mallya[32] 2015 | Cardoso[33] 2015 | Madan[34] 2014 | Pit[35] 2013 | Fassina[36] 2011 | Chaiyakul[20] 2018 |
| AMSTAR Assessment | A priori design | Y | Cochrane Risk of Bias Tool Assessment | Random sequence generation | HR | ROBINS-I Assessment | Confounding | LR | MR | MR | MR | MR | MR |
| | Duplicate study selection and data extraction | U | | Allocation concealment | HR | | Patient selection | MR | SR | MR | LR | SR | MR |
| | Comprehensive literature search | Y | | Blinding – patients and conductors | HR | | Intervention classification | LR | LR | LR | LR | LR | LR |
| | Publication status as inclusion criterion | Y | | Blinding – outcome assessors | HR | | Deviation from intended intervention | LR | LR | LR | SR | LR | LR |
| | List of included and excluded studies | Y | | Complete outcome data | HR | | Missing data | MR | LR | MR | LR | SR | MR |
| | Characteristics of included studies | Y | | Selective outcome reporting | LR | | Outcome measurements | LR | MR | MR | LR | MR | MR |
| | Study quality assessment conducted | Y | | Overall Risk of Bias | HR | | Selection of reported outcomes | MR | MR | LR | LR | MR | LR |
| | Quality assessment used in formulating conclusions | Y | Validated and reliable measures | | Y | | Overall Risk of Bias | MR | SR | MR | SR | SR | MR |
| | Appropriate methods to combine findings | Y | Adequately powered | | Y | Adequately powered | | Y | Y | N | Y | Y | Y |
| | Publication bias assessment | Y | Reported funding sources | | Y | Reported funding sources | | Y | N | N | N | N | Y |
| | Conflict of interest reported | Y | Industry funded | | N | Industry funded | | N | U | U | U | U | N |

| Systematic Reviews | | Randomized Controlled Trials | | Cohort Studies | PCS | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Study | Van der Heijden[24] 2014 | Study | Trisolini[31] 2015 | Study | Mallya[32] 2015 | Cardoso[33] 2015 | Madan[34] 2014 | Pit[35] 2013 | Fassina[36] 2011 | Chaiyakul[20] 2018 |
| Reported funding sources | Y | Study Quality | Int | Study Quality | Low | Int-Low | Int-Low | Int-Low | Int-Low | Int |
| Study Quality | High | | | | | | | | | |

Abbreviations: HR, high risk; Int, intermediate; Int-Low; intermediate-low; LR, low risk; MR, moderate risk; N, no; PCS, prospective cohort study; SR, serious risk; U, unclear/unsure; Y, yes.

**Supplemental Table 5b. Risk of Bias Assessment Results for Statement 3**

| Cohort Studies | | Retrospective Cohort Studies | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Study | | Mishra[46] 2016 | Izumo[37] 2016 | Schacht[38] 2016 | Thiryayi[39] 2016 | Hopkins[40] 2016 | Rokadia[41] 2016 | Rooper[42] 2016 | Guo[43] 2016 | Tachibana[44] 2013 | Joseph[10] 2013 | Griffin[45] 2011 | Gilbert[13] 2009 | Gilbert[15] 2009 |
| ROBINS-I Assessment | Confounding | MR | MR | MR | MR | MR | MR | MR | MR | MR | MR | MR | MR | MR |
| | Patient selection | SR | CR | CR | CR | CR | CR | CR | CR | CR | CR | CR | CR | CR |
| | Intervention classification | LR | LR | LR | LR | LR | LR | LR | LR | LR | LR | LR | MR | LR |
| | Deviation from intended intervention | MR | LR | LR | LR | LR | LR | LR | LR | LR | LR | LR | MR | LR |
| | Missing data | MR | LR | MR | MR | SR | CR | LR | LR | LR | LR | MR | MR | SR |
| | Outcome measurements | MR | LR | MR | LR | MR | SR | LR | MR | MR | MR | MR | MR | MR |
| | Selection of reported outcomes | MR | LR | LR | LR | MR | SR | MR | LR | LR | MR | MR | MR | MR |
| | Overall Risk of Bias | SR | CR | CR | CR | CR | CR | CR | CR | CR | CR | CR | CR | CR |
| Adequately powered | | N | Y | Y | Y | Y | Y | Y | Y | Y | Y | Y | Y | Y |
| Reported funding sources | | Y | Y | N | N | N | N | Y | Y | N | Y | N | N | N |
| Industry funded | | N | N | U | U | U | U | N | N | U | N | U | U | U |

| Cohort Studies | Retrospective Cohort Studies | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Study | Mishra[4]6 2016 | Izumo[3]7 2016 | Schacht[38] 2016 | Thiryayi[39] 2016 | Hopkins[40] 2016 | Rokadia[41] 2016 | Rooper[4]2 2016 | Guo[4]3 2016 | Tachibana[44] 2013 | Joseph[1]0 2013 | Griffin[4]5 2011 | Gilbert[1]3 2009 | Gilbert[1]5 2009 |
| Study Quality | Low | Low | Very Low | Low | Very Low | Very Low | Low | Low | Low | Low | Low | Very Low | Very Low |

Abbreviations: CR, critical risk; LR, low risk; MR, moderate risk; N, no; SR, serious risk; U, unclear/unsure; Y, yes.

**Supplemental Table 6. Risk of Bias Assessment Results for Statement 4**

| Systematic Reviews | | | Randomized Controlled Trials | | | Cohort Studies | | PCS | | | RCS |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Study | | Van der Heijden[24] 2014 | Study | | Trisolini[31]2015 | Study | | Sun[47] 2015 | Leong[49] 2017 | Oki[50] 2018 | Ecka[48] 2013 |
| AMSTAR Assessment | A priori design | Y | Cochrane Risk of Bias Tool Assessment | Random sequence generation | HR | ROBINS-I Assessment | Confounding | LR | LR | LR | MR |
| | Duplicate study selection and data extraction | U | | Allocation concealment | HR | | Patient selection | LR | MR | LR | CR |
| | Comprehensive literature search | Y | | Blinding – patients and conductors | HR | | Intervention classification | LR | LR | MR | LR |
| | Publication status as inclusion criterion | Y | | Blinding – outcome assessors | HR | | Deviation from intended intervention | LR | LR | LR | LR |
| | List of included and excluded studies | Y | | Complete outcome data | HR | | Missing data | MR | LR | LR | LR |
| | Characteristics of included studies | Y | | Selective outcome reporting | LR | | Outcome measurements | LR | LR | LR | LR |
| | Study quality assessment conducted | Y | | Overall Risk of Bias | HR | | Selection of reported outcomes | LR | LR | LR | MR |
| | Quality assessment used in formulating conclusions | Y | Validated and reliable measures | | Y | | Overall Risk of Bias | MR | MR | MR | CR |

| Systematic Reviews | | | Randomized Controlled Trials | | Cohort Studies | PCS | | | RCS |
|---|---|---|---|---|---|---|---|---|---|
| Study | | Van der Heijden[24] 2014 | Study | Trisolini[31]2015 | Study | Sun[47] 2015 | Leong[49] 2017 | Oki[50] 2018 | Ecka[48] 2013 |
| | Appropriate methods to combine findings | Y | Adequately powered | Y | Adequately powered | Y | Y | Y | Y |
| | Publication bias assessment | Y | Reported funding sources | Y | Reported funding sources | Y | Y | Y | N |
| | Conflict of interest reported | Y | Industry funded | N | Industry funded | N | N | N | U |
| Reported funding sources | | Y | Study Quality | Int | Study Quality | Int-Low | Int-Low | Int-Low | Low |
| Study Quality | | High | | | | | | | |

Abbreviations: CR, critical risk; HR, high risk; Int, intermediate; Int-Low; intermediate-low; LR, low risk; MR, moderate risk; N, no; PCS, prospective cohort study; RCS, retrospective cohort study; U, unclear/unsure; Y, yes.

**Supplemental Table 7. Risk of Bias Assessment Results for Statement 5**

| Systematic Reviews | | | Cohort Studies | | PCS | | | RCS | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Study | | Van der Heijden[24] 2014 | Study | | Mfokazi[51] 2016 | Fassina[36] 2011 | Chang[53] 2008 | Ecka[48] 2013 | Tachibana[44] 2013 | Coley[52] 2015 | Welborn[54] 2018 |
| AMSTAR Assessment | A priori design | Y | ROBINS-I Assessment | Confounding | LR | MR | MR | MR | MR | MR | MR |
| | Duplicate study selection and data extraction | U | | Patient selection | LR | SR | SR | CR | CR | CR | CR |
| | Comprehensive literature search | Y | | Intervention classification | LR | LR | MR | LR | LR | LR | LR |
| | Publication status as inclusion criterion | Y | | Deviation from intended intervention | LR | LR | MR | LR | LR | LR | MR |
| | List of included and excluded studies | Y | | Missing data | LR | SR | MR | LR | LR | LR | MR |
| | Characteristics of included studies | Y | | Outcome measurements | LR | MR | MR | LR | MR | LR | MR |
| | Study quality assessment conducted | Y | | Selection of reported outcomes | LR | MR | LR | MR | LR | LR | LR |

| Systematic Reviews | | | Cohort Studies | | PCS | | | RCS | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Study | | Van der Heijden[24] 2014 | Study | | Mfokazi[51] 2016 | Fassina[36] 2011 | Chang[53] 2008 | Ecka[48] 2013 | Tachibana[44] 2013 | Coley[52] 2015 | Welborn[54] 2018 |
| | Quality assessment used in formulating conclusions | Y | | Overall Risk of Bias | LR | SR | SR | CR | CR | CR | CR |
| | Appropriate methods to combine findings | Y | Adequately powered | | Y | Y | Y | Y | Y | Y | Y |
| | Publication bias assessment | Y | Reported funding sources | | Y | N | N | N | N | Y | Y |
| | Conflict of interest reported | Y | Industry funded | | N | U | U | U | U | N | N |
| Reported funding sources | | Y | Study Quality | | High-Int | Int-Low | Int-Low | Low | Low | Low | Low |
| Study Quality | | High | | | | | | | | | |

Abbreviations: CR, critical risk; High-Int, high-intermediate; Int-Low; intermediate-low; LR, low risk; MR, moderate risk; N, no; PCS, prospective cohort study; RCS, retrospective cohort study; SR, serious risk; U, unclear/unsure; Y, yes.

**Supplemental Table 8. Risk of Bias Assessment Results for Statement 6**

| Systematic Reviews | | | Cohort Studies | | PCS | | | | | | RCS | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Study | | DiBardino[56] 2015 | Study | | Konjengbam[64] 2014 | Tam[59] 2013 | D' Alessandro[61] 2007 | Konofaos[62] 2006 | Chang[53] 2008 | Uzun[63] 2017 | Lalji[55] 2015 | Chen[58] 2014 | Khan[60] 2012 | Aktas[65] 2015 | Jaconi[57] 2015 |
| AMSTAR Assessment | A priori design | N | ROBINS-I Assessment | Confounding | MR | SR | MR | MR | MR | MR | MR | MR | MR | MR | MR |
| | Duplicate study selection and data extraction | N | | Patient selection | MR | SR | MR | SR | SR | CR | CR | CR | CR | CR | CR |
| | Comprehensive literature search | Y | | Intervention classification | LR | LR | LR | LR | MR | LR | LR | LR | LR | LR | LR |
| | Publication status as inclusion criterion | N | | Deviation from intended intervention | MR | LR | LR | LR | MR | LR | LR | LR | LR | LR | LR |

| Systematic Reviews | | Cohort Studies | | PCS | | | | | | RCS | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Study | DiBardino[56] 2015 | Study | Konjengbam[64] 2014 | Tam[59] 2013 | D' Alessandro[6] 1 2007 | Konofaos[62] 2006 | Chang[53] 2008 | Uzun[63] 2017 | Lajji[55] 2015 | Chen[58] 2014 | Khan[60] 2012 | Aktas[65] 2015 | Jaconi[57] 2015 |
| List of included and excluded studies | N | Missing data | LR | LR | MR | MR | MR | SR | LR | SR | SR | SR | CR |
| Characteristics of included studies | Y | Outcome measurements | LR | LR | MR | LR | MR | MR | LR | SR | SR | MR | LR |
| Study quality assessment conducted | N | Selection of reported outcomes | LR | LR | MR | LR | LR | MR | LR | MR | MR | MR | LR |
| Quality assessment used in formulating conclusions | N | Overall Risk of Bias | MR | SR | MR | SR | SR | CR | CR | CR | CR | CR | CR |
| Appropriate methods to combine findings | N | Adequately powered | N | Y | Y | Y | Y | Y | Y | Y | N | Y | Y |
| Publication bias assessment | N | Reported funding sources | N | Y | N | N | N | N | N | Y | N | N | Y |
| Conflict of interest reported | Y | Industry funded | U | N | U | U | U | U | U | N | U | U | N |
| Reported funding sources | N | Study Quality | Int-Low | Int-Low | Int-Low | Int-Low | Int-Low | Low | Low | Low | Very Low | Very Low | Very Low |
| Study Quality | Int-Low | | | | | | | | | | | | |

Abbreviations: CR, critical risk; Int-Low; intermediate-low; LR, low risk; MR, moderate risk; N, no; PCS, prospective cohort study; RCS, retrospective cohort study; SR, serious risk; U, unclear/unsure; Y, yes.

**Supplemental Table 9. Risk of Bias Assessment Results for Statement 7**

| Cohort Studies | | PCS | | | | | | RCS | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Study | | Fassina[36] 2011 | Diacon[67] 2007 | Chang[53] 2008 | Ugurluoglu[74] 2015 | Dotson[68] 2019 | Son[71] 2015 | Halloush[66] 2007 | Baum[69] 2017 | Treece[70] 2016 | Grunes[75] 2016 | Welborn[54] 2018 | Wang[72] 2015 | Schneider[73] 2015 |
| ROBINS-I Assessment | Confounding | MR | MR | MR | MR | MR | MR | MR | MR | MR | MR | MR | MR | MR |
| | Patient selection | SR | MR | SR | MR | SR | MR | CR | CR | CR | CR | CR | CR | CR |
| | Intervention classification | LR | LR | MR | LR | LR | LR | LR | LR | LR | LR | LR | MR | LR |
| | Deviation from intended intervention | LR | MR | MR | LR | LR | LR | LR | LR | LR | LR | MR | MR | LR |
| | Missing data | SR | LR | MR | CR | MR | MR | MR | LR | SR | LR | MR | MR | MR |
| | Outcome measurements | MR | SR | MR | MR | LR | MR | MR | LR | SR | MR | MR | SR | MR |
| | Selection of reported outcomes | MR | LR | LR | MR | LR | LR | MR | LR | CR | LR | LR | MR | MR |
| | Overall Risk of Bias | SR | SR | SR | CR | SR | MR | CR | CR | CR | CR | CR | CR | CR |
| Adequately powered | | Y | Y | Y | Y | Y | Y | Y | Y | N | Y | Y | Y | Y |
| Reported funding sources | | N | Y | N | N | Y | Y | N | Y | Y | Y | Y | Y | N |
| Industry funded | | U | N | U | U | N | N | U | N | N | N | N | N | U |
| Study Quality | | Int-Low | Int-Low | Int-Low | Low | Int-Low | Int-Low | Low | Low | Low | Low | Low | Very Low | Low |

Abbreviations: CR, critical risk; Int-Low; intermediate-low; LR, low risk; MR, moderate risk; N, no; PCS, prospective cohort study; RCS, retrospective cohort study; SR, serious risk; U, unclear/unsure; Y, yes.

**Supplemental Table 10. Risk of Bias Assessment Results for Statement 8**

| Cohort Studies | | RCS |
|---|---|---|
| Study | | Wehrschuetz[77] 2010 |
| ROBI | Confounding | MR |
| | Patient selection | CR |

| Cohort Studies | | RCS |
|---|---|---|
| Study | | Wehrschuetz[77] 2010 |
| | Intervention classification | LR |
| | Deviation from intended intervention | LR |
| | Missing data | MR |
| | Outcome measurements | MR |
| | Selection of reported outcomes | LR |
| | Overall Risk of Bias | CR |
| Adequately powered | | Y |
| Reported funding sources | | N |
| Industry funded | | U |
| Study Quality | | Low |

Abbreviations: CR, critical risk; LR, low risk; MR, moderate risk; N, no; RCS, retrospective cohort study; U, unclear/unsure; Y, yes.

**Supplemental Table 11. Risk of Bias Assessment Results for Statement 9**

| Cohort Studies | | PCS | RCS |
|---|---|---|---|
| Study | | Sanchez-Font[78] 2014 | Balbo[79] 2013 |
| ROBINS-I Assessment | Confounding | MR | MR |
| | Patient selection | MR | CR |
| | Intervention classification | LR | LR |
| | Deviation from intended intervention | LR | LR |
| | Missing data | LR | LR |
| | Outcome measurements | MR | MR |
| | Selection of reported outcomes | MR | LR |
| | Overall Risk of Bias | MR | CR |
| Adequately powered | | Y | N |
| Reported funding sources | | N | N |
| Industry funded | | U | U |

| Cohort Studies | PCS | RCS |
|---|---|---|
| Study | Sanchez-Font[78] 2014 | Balbo[79] 2013 |
| Study Quality | Int-Low | Very Low |

Abbreviations: CR, critical risk; Int-Low; intermediate-low; LR, low risk; MR, moderate risk; N, no; PCS, prospective cohort study; RCS, retrospective cohort study; U, unclear/unsure; Y, yes.

**Supplemental Table 12. Risk of Bias Assessment Results for Statement 10**

| Systematic Reviews | | | Cohort Studies | | PCS | | | | RCS | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Study | | Mondoni[80] 2016 | Study | | Conti[86] 2016 | Sindhwani[84] 2013 | Diacon[85] 2010 | Chang[53] 2008 | Madan[81] 2016 | Chen[82] 2015 | Loo[83] 2014 |
| AMSTAR Assessment | A priori design | Y | ROBINS-I Assessment | Confounding | MR | MR | MR | MR | MR | MR | MR |
| | Duplicate study selection and data extraction | U | | Patient selection | MR | MR | LR | SR | CR | CR | CR |
| | Comprehensive literature search | Y | | Intervention classification | SR | LR | LR | MR | LR | CR | MR |
| | Publication status as inclusion criterion | N | | Deviation from intended intervention | MR | LR | LR | MR | LR | MR | LR |
| | List of included and excluded studies | Y | | Missing data | MR | MR | MR | MR | LR | SR | SR |
| | Characteristics of included studies | Y | | Outcome measurements | MR | MR | LR | MR | LR | CR | SR |
| | Study quality assessment conducted | Y | | Selection of reported outcomes | LR | MR | LR | LR | LR | MR | MR |
| | Quality assessment used in formulating conclusions | N | | Overall Risk of Bias | SR | MR | MR | SR | CR | CR | CR |
| | Appropriate methods to combine findings | Y | Adequately powered | | Y | N | Y | Y | Y | Y | N |
| | Publication bias assessment | N | Reported funding sources | | N | N | Y | N | Y | N | N |
| | Conflict of interest reported | Y | Industry funded | | U | U | N | U | N | U | U |
| Reported funding sources | | N | Study Quality | | Int-Low | Int-Low | Int | Int-Low | Low | Very Low | Very Low |
| Study Quality | | High-Int | | | | | | | | | |

Abbreviations: CR, critical risk; High-Int, high-intermediate; Int-Low; intermediate-low; LR, low risk; MR, moderate risk; N, no; PCS, prospective cohort study; RCS, retrospective cohort study; SR, serious risk; U, unclear/unsure; Y, yes.

**Supplemental Table 13. Risk of Bias Assessment Results for Statement 11**

| Cohort Studies | | PCS | | | RCS | | | |
|---|---|---|---|---|---|---|---|---|
| Study | | Swiderek[87] 2010 | Abouzgheib[88] 2009 | Buckley[89] 2008 | Wu[90] 2017 | Rooper[91] 2014 | Thomas[92] 2011 | DeMaio[93] 2019 |
| ROBINS-I Assessment | Confounding | MR | MR | MR | MR | MR | MR | MR |
| | Patient selection | MR | MR | MR | CR | CR | CR | CR |
| | Intervention classification | LR | LR | LR | LR | LR | LR | LR |
| | Deviation from intended intervention | LR | LR | LR | LR | LR | LR | LR |
| | Missing data | MR | LR | LR | LR | LR | LR | MR |
| | Outcome measurements | MR | MR | LR | LR | LR | LR | LR |
| | Selection of reported outcomes | LR | LR | LR | LR | MR | LR | LR |
| | Overall Risk of Bias | MR | MR | MR | CR | CR | CR | CR |
| Adequately powered | | Y | Y | Y | Y | N | Y | Y |
| Reported funding sources | | Y | N | N | Y | Y | N | Y |
| Industry funded | | N | U | U | N | N | U | N |
| Study Quality | | Int | Int-Low | Int | Low | Low | Low | Low |

Abbreviations: CR, critical risk; HR, high risk; Int, intermediate; Int-Low; intermediate-low; LR, low risk; MR, moderate risk; N, no; PCS, prospective cohort study; RCS, retrospective cohort study; SR, serious risk; U, unclear/unsure; Y, yes.

**Supplemental Table 14. Risk of Bias Assessment Results for Statement 12**

| Cohort Studies | | PCS | Retrospective Cohort Studies | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Study | | Minca[96] 2014 | Zhang[99] 2015 | Lee[95] 2016 | Florentine[94] 2015 | Thiryayi[39] 2016 | Rooper[42] 2016 | Baum[69] 2017 | Treece[70] 2016 | Wu[98] 2014 | Ozluk[97] 2017 |
| ROBINS-I Assessment | Confounding | MR | MR | MR | MR | MR | MR | MR | MR | MR | MR |
| | Patient selection | SR | CR | CR | CR | CR | CR | CR | CR | CR | CR |
| | Intervention classification | MR | MR | LR | LR | LR | LR | LR | LR | LR | MR |
| | Deviation from intended intervention | LR | LR | LR | LR | LR | LR | LR | LR | LR | LR |

| Cohort Studies | | PCS | Retrospective Cohort Studies | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Study | | Minca[96] 2014 | Zhang[99] 2015 | Lee[95] 2016 | Florentine[94] 2015 | Thiryayi[39] 2016 | Rooper[42] 2016 | Baum[69] 2017 | Treece[70] 2016 | Wu[98] 2014 | Ozluk[97] 2017 |
| | Missing data | MR | LR | SR | MR | MR | LR | LR | SR | MR | LR |
| | Outcome measurements | LR | LR | MR | MR | LR | LR | LR | SR | MR | LR |
| | Selection of reported outcomes | LR | LR | LR | MR | LR | MR | LR | CR | MR | LR |
| | Overall Risk of Bias | MR | CR | CR | CR | CR | CR | CR | CR | CR | CR |
| Adequately powered | | Y | Y | N | Y | Y | Y | Y | N | Y | Y |
| Reported funding sources | | Y | N | N | Y | N | Y | Y | Y | Y | Y |
| Industry funded | | Y | U | U | N | U | N | N | N | N | N |
| Study Quality | | Int-Low | Low | Very Low | Low | Low | Low | Low | Low | Low | Very Low |

Abbreviations: CR, critical risk; Int-Low; intermediate-low; LR, low risk; MR, moderate risk; N, no; PCS, prospective cohort study; SR, serious risk; U, unclear/unsure; Y, yes.

**Supplemental Table 15. Risk of Bias Assessment Results for Statement 13**

| Cohort Studies | | Prospective Cohort Studies | | Retrospective Cohort Studies | |
|---|---|---|---|---|---|
| Study | | Wu[102] 2016 | Tam[59] 2013 | Hsiao[100] 2013 | Tian[101] 2017 |
| ROBINS-I Assessment | Confounding | LR | SR | MR | MR |
| | Patient selection | MR | SR | CR | CR |
| | Intervention classification | LR | LR | LR | LR |
| | Deviation from intended intervention | LR | LR | LR | LR |
| | Missing data | LR | LR | MR | MR |
| | Outcome measurements | LR | LR | MR | MR |
| | Selection of reported outcomes | LR | LR | LR | LR |
| | Overall Risk of Bias | MR | SR | CR | CR |
| Adequately powered | | Y | Y | Y | Y |
| Reported funding sources | | N | Y | Y | Y |

| Cohort Studies | Prospective Cohort Studies | | Retrospective Cohort Studies | |
|---|---|---|---|---|
| Study | Wu[102] 2016 | Tam[59] 2013 | Hsiao[100] 2013 | Tian[101] 2017 |
| Industry funded | U | N | N | N |
| Study Quality | Int | Int-Low | Low | Low |

Abbreviations: CR, critical risk; Int, intermediate; Int-Low; intermediate-low; LR, low risk; MR, moderate risk; N, no; SR, serious risk; U, unclear/unsure; Y, yes.

**Supplemental Table 16. Risk of Bias Assessment Results for Statement 14**

| Cohort Studies | | RCS |
|---|---|---|
| Study | | Lin[103] 2009 |
| ROBINS-I Assessment | Confounding | MR |
| | Patient selection | CR |
| | Intervention classification | LR |
| | Deviation from intended intervention | MR |
| | Missing data | SR |
| | Outcome measurements | LR |
| | Selection of reported outcomes | MR |
| | Overall Risk of Bias | CR |
| Adequately powered | | Y |
| Reported funding sources | | Y |
| Industry funded | | N |
| Study Quality | | Low |

Abbreviations: CR, critical risk; LR, low risk; MR, moderate risk; N, no; RCS, retrospective cohort study; Y, yes.

**Supplemental Table 17. Risk of Bias Assessment Results for Statement 15**

| Cohort Studies | | RCS | | |
|---|---|---|---|---|
| Study | | Kiral[105] 2015 | Eom[104] 2015 | Thangakunam[106] 2017 |
| ROBINS-I Assessment | Confounding | MR | MR | MR |
| | Patient selection | CR | SR | CR |

| Cohort Studies | | RCS | | |
|---|---|---|---|---|
| Study | | Kiral[105] 2015 | Eom[104] 2015 | Thangakunam[106] 2017 |
| | Intervention classification | LR | LR | LR |
| | Deviation from intended intervention | LR | MR | LR |
| | Missing data | MR | SR | MR |
| | Outcome measurements | LR | MR | SR |
| | Selection of reported outcomes | MR | MR | SR |
| | Overall Risk of Bias | CR | SR | CR |
| Adequately powered | | N | Y | Y |
| Reported funding sources | | N | N | N |
| Industry funded | | U | U | U |
| Study Quality | | Low | Low | Very Low |

Abbreviations: CR, critical risk; LR, low risk; MR, moderate risk; N, no; RCS, retrospective cohort study; SR, serious risk; U, unclear/unsure; Y, yes.

**Supplemental Table 18. Risk of Bias Assessment Results for Statement 16**

| Cohort Studies | | PCS |
|---|---|---|
| Study | | Wang[76] 2016 |
| ROBINS-I Assessment | Confounding | MR |
| | Patient selection | LR |
| | Intervention classification | LR |
| | Deviation from intended intervention | LR |
| | Missing data | MR |
| | Outcome measurements | MR |
| | Selection of reported outcomes | LR |
| | Overall Risk of Bias | MR |

| Cohort Studies | PCS |
|---|---|
| Study | Wang[76] 2016 |
| Adequately powered | Y |
| Reported funding sources | Y |
| Industry funded | N |
| Study Quality | Int |

Abbreviations: Int, intermediate; LR, low risk; MR, moderate risk; N, no; PCS, prospective cohort study; Y, yes

## Supplemental Figure 1:

**PubMed Search String:**
("Biopsy, Needle"[Mesh] OR "Minimally Invasive Surgical Procedures"[Mesh:noexp] OR "Image-Guided Biopsy"[Mesh] OR "Pleural Effusion"[Mesh] OR "core biopsies"[tw] OR "core biopsy"[tw] OR "skinny needle"[tw] OR CNB[tw] OR "core needle"[tw] OR cytopuncture[tw] OR cytopunctures[tw] OR EBUS[tw] OR "endobronchial ultrasound guided"[tw] OR "fine needle"[tw] OR FNA[tw] OR FNAB[tw] OR "Image-Guided Biopsies"[tw] OR "Image-Guided Biopsy"[tw] OR "needle aspirate"[tw] OR "needle aspirates"[tw] OR "needle aspiration"[tw] OR "needle biopsies"[tw] OR "needle biopsy"[tw] OR "needle core"[tw] OR "small biopsies"[tw] OR "small biopsy"[tw] OR "small tissue"[tw] OR "transbronchial needle aspiration"[tw] OR "transbronchial needle aspirations"[tw] OR effusion[tw] OR effusions[tw] OR (pleural[tw] AND (fluid[tw] OR fluids[tw])) OR "Touch imprint"[tw] OR "Touch imprints"[tw] OR "Touch imprinting"[tw] OR "Touch prep"[tw] OR "Touch preparation"[tw] OR "Touch preparations"[tw] OR "Touch preps"[tw] OR "Touch print"[tw] OR "Touch prints"[tw]) AND (("Thoracic Neoplasms"[Mesh:noexp] OR "Mediastinal Neoplasms"[Mesh] OR "Respiratory Tract Neoplasms"[Mesh:noexp] OR "Lung Neoplasms"[Mesh] OR "Tracheal Neoplasms"[Mesh] OR "Thymus Neoplasms"[Mesh] OR "Sarcoidosis, Pulmonary"[Mesh] OR "Solitary Pulmonary Nodule"[Mesh] OR "Mesothelioma"[Mesh] OR "Lung Diseases, Fungal"[Mesh] OR "Tuberculosis"[Mesh:noexp] OR "Tuberculosis, Pleural"[Mesh] OR "Tuberculosis, Pulmonary"[Mesh:noexp] OR PCP[tw] OR pneumocystis[tw] OR tuberculosis[tw] OR tuberculous[tw]) OR ((aspergilloma[tw] OR aspergilloses[tw] OR aspergillosis[tw] OR aspergillus[tw] OR blastomycoses[tw] OR blastomycosis[tw] OR "coccidioides immitis"[tw] OR coccidioides[tw] OR coccidioidomycoses[tw] OR coccidioidomycosis[tw] OR cryptococcoses[tw] OR cryptococcosis[tw] OR cryptococcus[tw] OR entomophthoramycoses[tw] OR entomophthoramycosis[tw] OR geotrichoses[tw] OR geotrichosis[tw] OR geotrichum[tw] OR histoplasma[tw] OR histoplasmoses[tw] OR histoplasmosis[tw] OR microspora[tw] OR microsporidia[tw] OR microsporidioses[tw] OR microsporidiosis[tw] OR mucorale[tw] OR mucorales[tw] OR mucormycoses[tw] OR mucormycosis[tw] OR mucoromycotina[tw] OR mycobacteria[tw] OR Mycobacterium[Mesh] OR mycobacterium[tw] OR Mycoses[Mesh] OR mycoses[tw] OR phycomycoses[tw] OR phycomycosis[tw] OR Scedosporiosis[tw] OR Scedosporium[tw] OR torulosis[tw] OR trichosporon[tw] OR trichosporonoses[tw] OR trichosporonosis[tw] OR zygomycoses[tw] OR zygomycosis[tw] OR Cancer[sb] OR "spindle cell"[tw] OR abnormalities[tw] OR abnormality[tw] OR lesion[tw] OR lesions[tw] OR mass[tw] OR masses[tw] OR nodule[tw] OR nodules[tw] OR Lymphadenopathy[Mesh] OR Sarcoidosis[Mesh:noexp] OR Adenopathies[tw] OR Adenopathy[tw] OR Lymphadenopathies[tw] OR Lymphadenopathy[tw] OR Sarcoidosis[tw] OR Sarcoidoses[tw] OR fungal[tw] OR Fungi[Mesh] OR fungus[tw] OR infection[tw] OR infections[tw]) AND (chest[tw] OR extrathoracic[tw] OR extra-thoracic[tw] OR Lung[tw] OR lungs[tw] OR mediastinal[tw] OR mesothelial[tw] OR mesothelium[tw] OR pleura[tw] OR pleural[tw] OR pulmonary[tw] OR respiratory[tw] OR thoracic[tw] OR thorax[tw] OR thymus[tw] OR trachea[tw] OR tracheal[tw] OR transthoracic[tw] OR trans-thoracic[tw]))) AND ("C.I. Fluorescent Brightening Agent 28"[Supplementary Concept] OR "Cytodiagnosis"[Mesh:noexp] OR "Cytological Techniques"[Mesh:noexp] OR "Fluorescent Antibody Technique, Direct"[Mesh] OR "DNA Mismatch Repair"[Mesh] OR "DNA Mutational Analysis"[Mesh] OR "Early Detection of Cancer"[Mesh] OR "Enzyme-Linked Immunosorbent Assay"[Mesh] OR "genomics"[Majr] OR "Genetic Testing"[Mesh] OR "Genotype"[Mesh:noexp] OR "Histocytochemistry"[Mesh:noexp] OR "Histocytological Preparation Techniques"[Mesh] OR "Immunohistochemistry"[Mesh] OR "Microbiological Techniques"[Mesh] OR "Molecular Diagnostic Techniques"[Mesh:noexp] OR "mucicarmine"[Supplementary Concept] OR "Pathology, Clinical/methods"[Mesh] OR "Pathology, Clinical/standards"[Mesh] OR "Pathology, Molecular/methods"[Mesh] OR "Pathology, Molecular/standards"[Mesh] OR "Pathology, Surgical/methods"[Mesh] OR "Pathology, Surgical/standards"[Mesh] OR "Polymerase Chain Reaction"[Mesh] OR Calcofluor-white[tw] OR culture[tw] OR cultures[tw] OR cytochemistry[tw] OR cytochemical[tw] OR cytodiagnosis[tw] OR cytogenetics[tw] OR cytology[tw] OR cytologies[tw] OR cytologic[tw] OR cytological[tw] OR cytopathology[tw] OR cytopathologic[tw] OR cytopathological[tw] OR cytopathologies[tw] OR

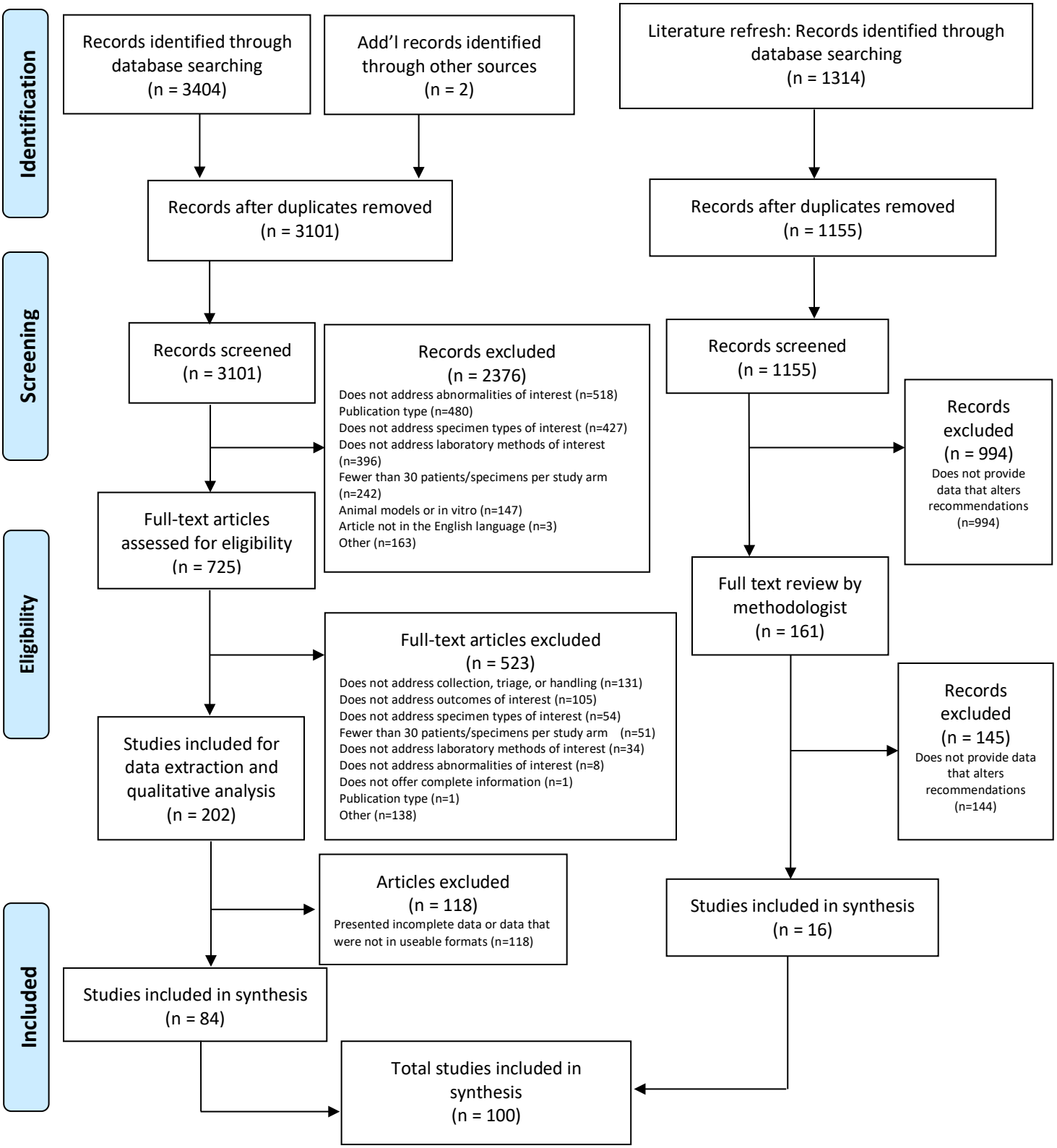
DFA[tw] OR "direct fluorescent antibody"[tw] OR FISH[tw] OR "Flow cytometry"[tw] OR "fluorescence in situ hybridization"[tw] OR histocytochemistry[tw] OR histocytochemical[tw] OR histopathology[tw] OR histopathologies[tw] OR histopathologic[tw] OR histopathological[tw] OR immunocytochemistry[tw] OR immunocytochemical[tw] OR immunohistochemistry[tw] OR immunohistochemical[tw] OR IHC[tw] OR Immunoperoxidase[tw] OR IPOX[tw] OR IPX[tw] OR microbiology[tw] OR microbiological[tw] OR mucicarmine[tw] OR "next-generation sequencing"[tw] OR NGS[tw] OR PCR[tw] OR "polymerase chain reaction"[tw] OR stain[tw] OR stains[tw] OR stained[tw] OR staining[tw] OR stainings[tw] OR ((histomolecular[tw] OR histo-molecular[tw] OR molecular[tw] OR mutation[tw] OR mutational[tw]) AND (analyses[tw] OR analysis[tw] OR biology[tw] OR detection[tw] OR method[tw] OR methods[tw] OR pathology[tw] OR profile[tw] OR profiles[tw] OR profiling[tw] OR studies[tw] OR study[tw] OR therapy[tw] OR therapies[tw] OR sub-type[tw] OR subtype[tw] OR sub-typing[tw] OR subtyping[tw] OR test[tw] OR tests[tw] OR testing[tw]))) AND ("Cold Ischemia"[Mesh] OR "Time Factors"[Mesh] OR "Specimen Handling"[Mesh:noexp] OR "Tissue Fixation"[Mesh] OR ischemia[tw] OR ischemic[tw] OR ischaemia[tw] OR ischaemic[tw] OR minute[tw] OR minutes[tw] OR time[tw] OR preanalytic[tw] OR pre-analytic[tw] OR preanalytical[tw] OR pre-analytical[tw] OR processing[tw] OR "collection media"[tw] OR "cell block"[tw] OR "cell blocks"[tw] OR fixation[tw] OR fixative[tw] OR fixatives[tw] OR AZF[tw] OR alcohol[tw] OR Cellient[tw] OR Cytolyt[tw] OR cytolyte[tw] OR formalin[tw] OR Hanks[tw] OR preservcyt[tw] OR RPMI[tw] OR saline[tw] OR surepath[tw] OR "sure path"[tw] OR thinprep[tw] OR "thin prep"[tw] OR air-dried[tw] OR air-dry[tw] OR air-drying[tw] OR cytospin[tw] OR liquid-based[tw] OR smear[tw] OR smears[tw] OR algorithm[tw] OR algorithms[tw] OR algorythm[tw] OR algorythm[tw] OR algorithmic[tw] OR triage[tw] OR triages[tw] OR triaged[tw] OR triaging[tw] OR protocol[tw] OR protocols[tw] OR flow-chart[tw] OR flowchart[tw] OR flow-charts[tw] OR flowcharts[tw] OR complication[tw] OR complications[tw] OR "failure rate"[tw] OR "failure rates"[tw] OR "rate of failure"[tw] OR adequate[tw] OR adequacy[tw] OR quality[tw] OR yield[tw] OR "analytic validity"[tw] OR "clinical validity"[tw] OR sterile[tw] OR fresh[tw] OR unpreserved[tw] OR un-preserved[tw] OR ((needle[tw] OR needles[tw]) AND (gauge[tw] OR gauges[tw] OR size[tw] OR sizes[tw] OR rinse[tw] OR rinsed[tw] OR rinses[tw] OR rinsing[tw])) OR ((number[tw]) AND (passes[tw] OR passages[tw] OR sites[tw])) OR ((sample[tw] OR samples[tw] OR specimen[tw] OR specimens[tw]) AND (collection[tw] OR collections[tw] OR handling[tw] OR handlings[tw])) OR ((Sequencing[tw] OR sequence[tw] OR order[tw]) AND (test[tw] OR tests[tw] OR testing[tw])) OR ((thick[tw] OR thickness) AND (cut[tw] OR slide[tw])) OR ((tumor[tw] OR tumour[tw] OR tumors[tw] OR tumours[tw]) AND (size[tw])) OR ROSE[tw] OR "rapid on-site"[tw] OR "rapid onsite"[tw]) AND ((English[Language]) AND ("2007"[Date - Publication] : "3000"[Date - Publication])) NOT ((animals[MeSH] NOT humans[MeSH]) OR "Case Reports"[Publication Type] OR "Comment"[Publication Type] OR "Editorial"[Publication Type] OR ("Letter"[Publication Type] NOT "Clinical Study"[Publication Type]))

**EMBASE Search String:**

(('biopsy needle'/exp OR 'minimally invasive procedure'/de OR 'image guided biopsy'/exp OR 'core biopsies' OR 'core biopsy' OR 'skinny needle' OR cnb OR cytopuncture OR cytopunctures OR ebus OR 'endobronchial ultrasound guided' OR 'fine needle' OR fna OR fnab OR 'image-guided biopsies' OR 'image-guided biopsy' OR 'needle aspirate' OR 'needle aspirates' OR 'needle aspiration' OR 'needle biopsies' OR 'needle biopsy' OR 'needle core' OR 'small biopsies' OR 'small biopsy' OR 'small tissue' OR 'transbronchial needle aspiration' OR 'transbronchial needle aspirations' OR 'pleura effusion'/exp OR 'pleural effusion' OR 'pleural effusions' OR (pleural NEAR/5 (fluid OR fluids)) OR (touch NEAR/1 (imprint OR imprints OR imprinting OR prep OR preparation OR preparations OR preps OR print OR prints))) AND ('neoplasms of the thorax and thoracic cavity'/de OR 'mediastinum tumor'/exp OR 'thorax tumor'/exp OR 'thymoma'/exp OR 'respiratory tract tumor'/de OR 'respiratory tract cancer'/de OR 'pleura tumor'/exp OR 'trachea tumor'/exp OR 'lung tumor'/exp OR 'lung sarcoidosis'/de OR 'lung nodule'/de OR 'mesothelioma'/de OR 'trachea cancer'/exp OR 'lung mycosis'/exp OR 'tuberculosis'/de OR 'tuberculous pleurisy'/de OR 'lung tuberculosis'/de OR pcp OR pneumocystis OR tuberculosis OR tuberculous OR ((aspergilloma OR aspergilloses OR aspergillosis OR aspergillus OR blastomycoses OR blastomycosis OR 'coccidioides immitis' OR coccidioides OR coccidioidomycoses OR coccidioidomycosis OR cryptococcoses OR cryptococcosis OR

cryptococcus OR entomophthoramycoses OR entomophthoramycosis OR geotrichoses OR geotrichosis OR geotrichum OR histoplasma OR histoplasmoses OR histoplasmosis OR microspora OR microsporidia OR microsporidioses OR microsporidiosis OR mucorale OR mucorales OR mucormycoses OR mucormycosis OR mucoromycotina OR mycobacteria OR 'mycobacterium'/exp OR 'mycosis'/exp OR mycoses OR phycomycoses OR phycomycosis OR scedosporiosis OR scedosporium OR toruloses OR torulosis OR trichosporon OR trichosporonoses OR trichosporonosis OR zygomycoses OR zygomycosis OR cancer OR cancerous OR carcinoma OR carcinomatosis OR carcinosarcoma OR carcinosis OR dysplasia OR leukemia OR lymphoma OR malignancy OR malignant OR neoplasia OR neoplasm OR neoplastic OR oncologic OR oncology OR tumor OR tumour OR 'spindle cell' OR abnormalities OR abnormality OR lesion OR lesions OR mass OR masses OR nodule OR nodules OR 'lymphadenopathy'/de OR 'mediastinum lymphadenopathy'/de OR 'lymph node metastasis'/exp OR 'generalized lymphadenopathy'/de OR 'immunoblastic lymphadenopathy'/de OR 'lymph node hyperplasia'/de OR 'sarcoidosis'/de OR adenopathies OR adenopathy OR lymphadenopathies OR lymphadenopathy OR sarcoidoses OR sarcoidosis OR fungal OR 'fungus'/exp OR fungus OR infection OR infections) AND (chest OR extrathoracic OR 'extra thoracic' OR lung OR lungs OR mediastinal OR mesothelial OR mesothelium OR pleura OR pleural OR pulmonary OR respiratory OR thoracic OR thorax OR thymus OR trachea OR tracheal OR transthoracic OR 'trans thoracic'))) AND ('cytodiagnosis'/de OR 'direct fluorescent antibody technique'/de OR 'genomics'/de OR genomic OR genomics OR 'mismatch repair'/de OR 'dna mutational analysis'/de OR 'early cancer diagnosis'/de OR 'enzyme linked immunosorbent assay'/de OR 'genetic screening'/exp OR 'genotype'/de OR 'cytochemistry'/exp OR 'microdissection'/de OR 'microtomy'/exp OR 'staining'/exp OR 'tissue preservation'/de OR 'immunohistochemistry'/exp OR 'microbiological examination'/de OR 'fungal examination'/exp OR 'molecular diagnosis'/de OR 'polymerase chain reaction'/exp OR 'calcofluor white' OR culture OR cultures OR cytochemistry OR cytochemical OR cytodiagnosis OR cytogenetics OR cytology OR cytologies OR cytologic OR cytological OR cytopathology OR cytopathologic OR cytopathological OR cytopathologies OR dfa OR 'direct fluorescent antibody' OR fish OR 'flow cytometry' OR 'fluorescence in situ hybridization' OR histocytochemistry OR histocytochemical OR histopathology OR histopathologies OR histopathologic OR histopathological OR immunocytochemistry OR immunocytochemical OR immunohistochemistry OR immunohistochemical OR ihc OR immunoperoxidase OR ipox OR ipx OR microbiology OR microbiological OR mucicarmine OR 'next-generation sequencing' OR ngs OR pcr OR 'polymerase chain reaction' OR stain OR stains OR stained OR staining OR stainings OR ((histomolecular OR 'histo-molecular' OR molecular OR mutation OR mutational) NEAR/3 (analyses OR analysis OR biology OR detection OR method OR methods OR pathology OR profile OR profiles OR profiling OR studies OR study OR therapies OR therapy OR 'sub type' OR subtype OR 'sub typing' OR subtyping OR test OR tests OR testing))) AND ('cold ischemia'/de OR 'time factor'/de OR 'specimen handling'/de OR 'tissue fixation'/de OR ischemia OR ischemic OR ischaemia OR ischaemic OR minute OR minutes OR time OR preanalytic OR 'pre analytic' OR preanalytical OR 'pre analytical' OR processing OR 'collection media' OR 'cell block' OR 'cell blocks' OR fixation OR fixative OR fixatives OR azf OR alcohol OR cellient OR cytolyte OR formalin OR hanks OR preservcyt OR rpmi OR saline OR surepath OR thinprep OR 'air dried' OR 'air dry' OR 'air drying' OR cytospin OR 'liquid based' OR smear OR smears OR algorithm OR algorithms OR algorythm OR algorithmic OR triage OR triages OR triaged OR triaging OR protocol OR protocols OR 'flow chart' OR flowchart OR complication OR complications OR 'failure rate' OR 'failure rates' OR 'rate of failure' OR adequate OR adequacy OR quality OR yield OR 'analytic validity' OR 'clinical validity' OR sterile OR fresh OR unpreserved OR 'un preserved' OR ((needle OR needles) NEAR/3 (gauge OR gauges OR size OR sizes OR rinse OR rinsed OR rinses OR rinsing)) OR (number NEAR/3 (passes OR passages OR sites)) OR ((sample OR specimen) NEAR/4 (collection OR collections OR handling OR handlings)) OR ((sequencing OR sequence OR order) NEAR/3 (test OR tests OR testing)) OR ((thick OR thickness) NEAR/3 (cut OR slide)) OR ((tumor OR tumour OR tumors OR tumours) NEAR/3 size) OR rose OR 'rapid on-site' OR 'rapid onsite') AND ([2007-2018]/py AND [english]/lim)) NOT ('conference abstract'/it OR 'conference paper'/exp OR 'case report'/exp OR 'case study'/exp OR 'editorial'/exp OR 'note'/exp OR ('letter'/exp NOT 'clinical study'/exp) OR ('animal'/exp NOT 'human'/exp) OR [medline]/lim)

**Supplemental Figure 2: Literature Review Flow Diagram**



Adapted from Moher D, Liberati A, Tetzlaff J, Altman DG; The PRISMA Group. Preferred reporting items for systematic reviews and meta-analysis: the PRISMA statement. PLoS Med. 2009;6(6):e1000097. doi:10.1371/journal.pmed1000097[109]

## REFERENCES


1. Lefebvre C, Manheimber E, Glanville J. Chapter 6: Searching for studies. In: Higgins JPT, Green S, eds. *Cochrane Handbook for Systematic Reviews of Interventions*. Version 5.1.0. Chichester, West Sussex, England: The Cochrane Collaboration; 2011. http://www.training.cochrane.org/handbook. Updated March 2011. Accessed February 27, 2020.
2. Brouwers MC, Kho ME, Browman GP, et al. AGREE II: advancing guideline development, reporting and evaluation in health care. *CMAJ.* 2010;182(18):E839-842. doi: 10.1503/cmaj.090449
3. Shea BJ, Grimshaw JM, Wells GA, et al. Development of AMSTAR: a measurement tool to assess the methodological quality of systematic reviews. *BMC Med Res Methodol.* 2007;7:10. doi: https://doi.org/10.1186/1471-2288-7-10
4. Higgins JP, Altman DG, Gotzsche PC, et al. The Cochrane Collaboration's tool for assessing risk of bias in randomised trials. *BMJ.* 2011;343:d5928. doi: 10.1136/bmj.d5928
5. Sterne JA, Hernan MA, Reeves BC, et al. ROBINS-I: a tool for assessing risk of bias in non-randomised studies of interventions. *BMJ.* 2016;355:i4919. doi: 10.1136/bmj.i4919
6. Alonso-Coello P, Schunemann HJ, Moberg J, et al. GRADE Evidence to Decision (EtD) frameworks: a systematic and transparent approach to making well informed healthcare choices. 1: Introduction. *BMJ.* 2016;353:i2016. doi: 10.1136/bmj.i2016
7. Vaidya PJ, Saha A, Kate AH, et al. Diagnostic value of core biopsy histology and cytology sampling of mediastinal lymph nodes using 21-gauge EBUS-TBNA needle. *J Cancer Res Ther.* 2016;12(3):1172-1177.
8. Wang H, D'Cruz C, Yam DC, Dilla LM, Tsang P. Learning curve for endobronchial ultrasoundguided transbronchial needle aspiration: experience of a community based teaching hospital. *Mol Med Rep.* 2014;10(5):2441-2446.
9. Zhao H, Xie Z, Zhou ZL, Sui XZ, Wang J. Diagnostic value of endobronchial ultrasound-guided transbronchial needle aspiration in intrapulmonary lesions. *Chin Med J (Engl).* 2013;126(22):4312-4315.
10. Joseph M, Jones T, Lutterbie Y, et al. Rapid on-site pathologic evaluation does not increase the efficacy of endobronchial ultrasonographic biopsy for mediastinal staging. *Ann Thorac Surg.* 2013;96(2):403-410.
11. Han SG, Yoo H, Jhun BW, et al. The role of endobronchial ultrasound-guided transbronchial needle aspiration in the diagnosis of recurrent non-small cell lung cancer after surgery. *Intern Med.* 2013;52(17):1875-1881.
12. Kennedy MP, Jimenez CA, Morice RC, et al. Factors influencing the diagnostic yield of endobronchial ultrasound-guided transbronchial needle aspiration. *J Bronchology Interv Pulmonol.* 2010;17(3):202-208.
13. Gilbert S, Wilson DO, Christie NA, et al. Endobronchial ultrasound as a diagnostic tool in patients with mediastinal lymphadenopathy. *Ann Thorac Surg.* 2009;88(3):896-900; discussion 901-892.
14. Nosotti M, Tosi D, Palleschi A, Ferrero S, Rosso L. Transbronchial needle aspiration under direct endobronchial ultrasound guidance of PET-positive isolated mediastinal adenopathy in patients with previous malignancy. *Surg Endosc.* 2009;23(6):1356-1359.

15. Gilbert S, Wilson DO, Christie NA, et al. Should endobronchial ultrasonography be part of the thoracic surgeon's armamentarium? *J Thorac Cardiovasc Surg.* 2009;137(2):413-418.
16. Nakajima T, Yasufuku K, Fujiwara T, et al. Endobronchial ultrasound-guided transbronchial needle aspiration for the diagnosis of intrapulmonary lesions. *J Thorac Oncol.* 2008;3(9):985-988.
17. Nambirajan A, Longchar M, Madan K, et al. Endobronchial ultrasound-guided transbronchial needle aspiration cytology in patients with known or suspected extra-pulmonary malignancies: a cytopathology-based study. *Cytopathology.* 2019;30(1):82-90.
18. Labarca G, Folch E, Jantz M, Mehta HJ, Majid A, Fernandez-Bussy S. Adequacy of samples obtained by endobronchial ultrasound with transbronchial needle aspiration for molecular analysis in patients with non-small cell lung cancer. systematic review and meta-analysis. *Ann Am Thorac Soc.* 2018;15(10):1205-1216.
19. Nair A, Haridas N, Ahmed S, Borkar PV. EBUS-TBNA-initial experience from a tertiary care center in southern India. *J Clin Diagn Res.* 2018;12(9):OC29-OC32.
20. Chaiyakul S. Endobronchial ultrasound-guided transbronchial needle aspiration for the diagnosis of central intrapulmonary lesions not visible by conventional bronchoscopy. *J Med Assoc Thai.* 2018;101(7):939-947.
21. Hu J, Li M, Xu L. Endobronchial ultrasound-guided transbronchial needle aspiration (EBUS-TBNA) in mediastinal lesions. *J of Int Transl Med.* 2018;6(2):76-79.
22. Jeyabalan A, Bhatt N, Plummeridge MJ, Medford AR. Adequacy of endobronchial ultrasound-guided transbronchial needle aspiration samples processed as histopathological samples for genetic mutation analysis in lung adenocarcinoma. *Mol Clin Oncol.* 2016;4(1):119-125.
23. Jeyabalan A, Shelley-Fraser G, Medford AR. Impact of needle gauge on characterization of endobronchial ultrasound-guided transbronchial needle aspiration (EBUS-TBNA) histology samples. *Respirology.* 2014;19(5):735-739.
24. van der Heijden EH, Casal RF, Trisolini R, et al. Guideline for the acquisition and preparation of conventional and endobronchial ultrasound-guided transbronchial needle aspiration specimens for the diagnosis and molecular testing of patients with known or suspected lung cancer. *Respiration.* 2014;88(6):500-517.
25. Tyan C, Patel P, Czarnecka K, et al. Flexible 19-gauge endobronchial ultrasound-guided transbronchial needle aspiration needle: first experience. *Respiration.* 2017;94(1):52-57.
26. Wolters C, Darwiche K, Franzen D, et al. A prospective, randomized trial for the comparison of 19-g and 22-g endobronchial ultrasound-guided transbronchial aspiration needles; introducing a novel end point of sample weight corrected for blood content. *Clin Lung Cancer.* 2019;20(3):e265-e273. doi: 10.1016/j.cllc.2019.02.019.
27. Pickering EM, Holden VK, Heath JE, Verceles AC, Kalchiem-Dekel O, Sachdeva A. Tissue acquisition during EBUS-TBNA: comparison of cell blocks obtained from a 19G versus 21G needle. *J Bronchology Interv Pulmonol.* 2019;26(4):237-244.
28. Dooms C, Vander Borght S, Yserbyt J, et al. A randomized clinical trial of flex 19g needles versus 22g needles for endobronchial ultrasonography in suspected lung cancer. *Respiration.* 2018;96(3):275-282.
29. Garrison G, Leclair T, Balla A, et al. Use of an additional 19-G EBUS-TBNA needle increases the diagnostic yield of EBUS-TBNA. *J Bronchology Interv Pulmonol.* 2018;25(4):269-273.
30. Kinoshita T, Ujiie H, Schwock J, et al. Clinical evaluation of the utility of a flexible 19-gauge EBUS-TBNA needle. *J Thorac Dis.* 2018;10(4):2388-2396.
31. Trisolini R, Cancellieri A, Tinelli C, et al. Randomized trial of endobronchial ultrasound-guided transbronchial needle aspiration with and without rapid on-site evaluation for lung cancer genotyping. *Chest.* 2015;148(6):1430-1437.
32. Mallya V, Kumar SP, Meganathan P, Shivkumar S, Mehta R. The utility of ROSE (rapid on-site evaluation) in endobronchial ultrasound (EBUS)-guided transbronchial needle aspiration (TBNA): is the picture rosy? *J Cytol.* 2015;32(4):230-233.

33. Cardoso AV, Neves I, Magalhaes A, Sucena M, Barroca H, Fernandes G. The value of rapid on-site evaluation during EBUS-TBNA. *Rev Port Pneumol (2006).* 2015;21(5):253-258.
34. Madan K, Mohan A, Ayub, II, et al. Initial experience with endobronchial ultrasound-guided transbronchial needle aspiration (EBUS-TBNA) from a tuberculosis endemic population. *J Bronchology Interv Pulmonol.* 2014;21(3):208-214.
35. Plit ML, Havryk AP, Hodgson A, et al. Rapid cytological analysis of endobronchial ultrasound-guided aspirates in sarcoidosis. *Eur Respir J.* 2013;42(5):1302-1308.
36. Fassina A, Corradin M, Zardo D, Cappellesso R, Corbetti F, Fassan M. Role and accuracy of rapid on-site evaluation of CT-guided fine needle aspiration cytology of lung nodules. *Cytopathology.* 2011;22(5):306-312.
37. Izumo T, Matsumoto Y, Sasada S, Chavez C, Nakai T, Tsuchida T. Utility of rapid on-site cytologic evaluation during endobronchial ultrasound with a guide sheath for peripheral pulmonary lesions. *Jpn J Clin Oncol.* 2016;47(3):221-225.
38. Schacht MJ, Toustrup CB, Madsen LB, Martiny MS, Larsen BB, Simonsen JT. Endobronchial ultrasound-guided transbronchial needle aspiration: performance of biomedical scientists on rapid on-site evaluation and preliminary diagnosis. *Cytopathology.* 2016;27(5):344-350.
39. Thiryayi SA, Rana DN, Narine N, Najib M, Bailey S. Establishment of an endobronchial ultrasound-guided transbronchial fine needle aspiration service with rapid on-site evaluation: 2 years experience of a single UK centre. *Cytopathology.* 2016;27(5):335-343.
40. Hopkins E, Moffat D, Parkinson I, et al. Cell block samples from endobronchial ultrasound transbronchial needle aspiration provide sufficient material for ancillary testing in lung cancer-a quaternary referral centre experience. *J Thorac Dis.* 2016;8(9):2544-2550.
41. Rokadia HK, Mehta A, Culver DA, et al. Rapid on-site evaluation in detection of granulomas in the mediastinal lymph nodes. *Ann Am Thorac Soc.* 2016;13(6):850-855.
42. Rooper LM, Nikolskaia O, Carter J, Ning Y, Lin MT, Maleki Z. A single EBUS-TBNA procedure can support a large panel of immunohistochemical stains, specific diagnostic subtyping, and multiple gene analyses in the majority of non-small cell lung cancer cases. *Hum Pathol.* 2016;51:139-145.
43. Guo H, Liu S, Guo J, et al. Rapid on-site evaluation during endobronchial ultrasound-guided transbronchial needle aspiration for the diagnosis of hilar and mediastinal lymphadenopathy in patients with lung cancer. *Cancer Lett.* 2016;371(2):182-186.
44. Tachibana K, Nakazato Y, Tsuchida S, et al. Immediate cytology improves accuracy and decreases complication rate in real-time computed tomography-guided needle lung biopsy. *Diagn Cytopathol.* 2013;41(12):1063-1068.
45. Griffin AC, Schwartz LE, Baloch ZW. Utility of on-site evaluation of endobronchial ultrasound-guided transbronchial needle aspiration specimens. *Cytojournal.* 2011;8:20. doi: 10.4103/1742-6413.90081
46. Mishra MM, Reynolds JP, Sturgis CD, Booth CN. Diagnosis of mediastinal lesions unassociated with lung carcinoma diagnosed by endobronchial ultrasound transbronchial needle aspiration (EBUS-TBNA). *J Am Soc Cytopathol.* 2016;5(4):189-195.
47. Sun J, Yang H, Teng J, et al. Determining factors in diagnosing pulmonary sarcoidosis by endobronchial ultrasound-guided transbronchial needle aspiration. *Ann Thorac Surg.* 2015;99(2):441-445.
48. Ecka RS, Sharma M. Rapid on-site evaluation of EUS-FNA by cytopathologist: an experience of a tertiary hospital. *Diagn Cytopathol.* 2013;41(12):1075-1080.
49. Leong TL, Christie M, Kranz S, et al. Evaluating the genomic yield of a single endobronchial ultrasound-guided transbronchial needle aspiration in lung cancer: meeting the challenge of doing more with less. *Clin Lung Cancer.* 2017;18(6):e467-e472.
50. Oki M, Saka H, Ando M, et al. How many passes are needed for endobronchial ultrasound-guided transbronchial needle aspiration for sarcoidosis? A prospective multicenter study. *Respiration.* 2018;95(4):251-257.

51. Mfokazi A, Wright CA, Louw M, et al. Direct comparison of liquid-based and smear-based cytology with and without rapid on-site evaluation for fine needle aspirates of thoracic tumors. *Diagn Cytopathol.* 2016;44(5):363-368.
52. Coley SM, Crapanzano JP, Saqi A. FNA, core biopsy, or both for the diagnosis of lung carcinoma: obtaining sufficient tissue for a specific diagnosis and molecular testing. *Cancer Cytopathol.* 2015;123(5):318-326.
53. Chang YC, Yu CJ, Lee WJ, et al. Imprint cytology improves accuracy of computed tomography-guided percutaneous transthoracic needle biopsy. *Eur Respir J.* 2008;31(1):54-61.
54. Welborn SL, Ohori NP, Nason KS, et al. Percutaneous computed tomography-guided biopsy performed by thoracic surgeons in 955 patients: a paradigm shift in image-guided thoracic procedures. *J Thorac Cardiovasc Surg.* 2019;157(3):1239-1245.
55. Lalji UC, Wildberger JE, Zur Hausen A, et al. CT-guided percutaneous transthoracic needle biopsies using 10G large-core needles: initial experience. *Cardiovasc Intervent Radiol.* 2015;38(6):1603-1610.
56. DiBardino DM, Yarmus LB, Semaan RW. Transthoracic needle biopsy of the lung. *J Thorac Dis.* 2015;7(Suppl 4):S304-316.
57. Jaconi M, Pagni F, Vacirca F, et al. C-arm cone-beam CT-guided transthoracic lung core needle biopsy as a standard diagnostic tool: an observational study. *Medicine (Baltimore).* 2015;94(12):e698. doi: 10.1097/MD.0000000000000698
58. Chen HJ, Yang JJ, Fang LY, et al. Feasibility of computed tomography-guided core needle biopsy in producing state-of-the-art clinical management in Chinese lung cancer. *Thorac Cancer.* 2014;5(2):155-161.
59. Tam AL, Kim ES, Lee JJ, et al. Feasibility of image-guided transthoracic core-needle biopsy in the BATTLE lung trial. *J Thorac Oncol.* 2013;8(4):436-442.
60. Khan KA, Zaidi S, Swan N, et al. The use of computerised tomography guided percutaneous fine needle aspiration in the evaluation of solitary pulmonary nodules. *Ir Med J.* 2012;105(2):50-52.
61. D'Alessandro V, Parracino T, Stranieri A, et al. Computed-tomographic-guided biopsy of thoracic nodules: a revision of 583 lesions. *Clin Ter.* 2007;158(6):509-513.
62. Konofaos P, Tomos P, Malagari K, et al. The role of ThinPrep cytology in the investigation of lung tumors. *Surg Oncol.* 2006;15(3):173-178.
63. Uzun Ç, Akkaya Z, Atman ED, et al. Diagnostic accuracy and safety of CT-guided fine needle aspiration biopsy of pulmonary lesions with non-coaxial technique: a single center experience with 442 biopsies. *Diagn Interv Radiol.* 2017;23(2):137-143.
64. Konjengbam R, Singh NB, Gatphoh SG. Computed tomography guided percutaneous transthoracic fine needle aspiration cytology of pulmonary mass lesions: two years cross sectional study of 61 cases. *J Med Soc.* 2014;28(2):112-116.
65. Aktaş AR, Gözlek E, Yılmaz Ö, et al. CT-guided transthoracic biopsy: histopathologic results and complication rates. *Diagn Interv Radiol.* 2015;21(1):67-70.
66. Halloush RA, Khasawneh FA, Saleh HA, Soubani AO, Piskorowski TJ, Al-Abbadi MA. Fine needle aspiration cytology of lung lesions: a clinicopathological and cytopathological review of 150 cases with emphasis on the relation between the number of passes and the incidence of pneumothorax. *Cytopathology.* 2007;18(1):44-51.
67. Diacon AH, Theron J, Schubert P, et al. Ultrasound-assisted transthoracic biopsy: fine-needle aspiration or cutting-needle biopsy? *Eur Respir J.* 2007;29(2):357-362.
68. Dotson T, Bellinger C, Su J, et al. Feasibility of lung cancer RNA acquisition from a single transbronchial or transthoracic needle pass (FASTT trial). *Lung Cancer.* 2019;127:6-11.
69. Baum JE, Zhang P, Hoda RS, et al. Accuracy of next-generation sequencing for the identification of clinically relevant variants in cytology smears in lung adenocarcinoma. *Cancer.* 2017;125(6):398-406.
70. Treece AL, Montgomery ND, Patel NM, et al. FNA smears as a potential source of DNA for targeted next-generation sequencing of lung adenocarcinomas. *Cancer Cytopathol.* 2016;124(6):406-414.

71. Son C, Kang EJ, Roh MS. Strategic management of transthoracic needle aspirates for histological subtyping and EGFR testing in patients with peripheral lung cancer: an institutional experience. *Diagn Cytopathol.* 2015;43(7):532-538.
72. Wang S, Yu B, Ng CC, et al. The suitability of small biopsy and cytology specimens for EGFR and other mutation testing in non-small cell lung cancer. *Transl Lung Cancer Res.* 2015;4(2):119-125.
73. Schneider F, Smith MA, Lane MC, Pantanowitz L, Dacic S, Ohori NP. Adequacy of core needle biopsy specimens and fine-needle aspirates for molecular testing of lung adenocarcinomas. *Am J Clin Pathol.* 2015;143(2):193-200; quiz 306.
74. Ugurluoglu C, Kurtipek E, Unlu Y, Esme H, Duzgun N. Importance of the cell block technique in diagnosing patients with non-small cell carcinoma accompanied by pleural effusion. *Asian Pac J Cancer Prev.* 2015;16(7):3057-3060.
75. Grunes D, Yankelevitz D, Beasley MB, et al. Fine-needle aspiration of small pulmonary nodules yields material for reliable molecular analysis of adenocarcinomas. *J Am Soc Cytopathol.* 2016;5(2):57-63.
76. Wang G, Yang X, Zhu J, et al. Evaluation of the efficacy of Myco/F lytic system, MGIT960 system and Lowenstein-Jensen medium for recovery of Mycobacterium tuberculosis from sterile body fluids. *Sci Rep.* 2016;6:37757. doi: 10.1038/srep37757
77. Wehrschuetz M, Wehrschuetz E, Portugaller HR. Number of biopsies in diagnosing pulmonary nodules. *Clin Med Circ Respirat Pulm Med.* 2010;4(1):9-14.
78. Sanchez-Font A, Giralt L, Vollmer I, Pijuan L, Gea J, Curull V. Endobronchial ultrasound for the diagnosis of peripheral pulmonary lesions. A controlled study with fluoroscopy. *Arch Bronconeumol.* 2014;50(5):166-171.
79. Balbo PE, Bodini BD, Patrucco F, et al. Electromagnetic navigation bronchoscopy and rapid on site evaluation added to fluoroscopy-guided assisted bronchoscopy and rapid on site evaluation: improved yield in pulmonary nodules. *Minerva Chir.* 2013;68(6):579-585.
80. Mondoni M, Sotgiu G, Bonifazi M, et al. Transbronchial needle aspiration in peripheral pulmonary lesions: a systematic review and meta-analysis. *Eur Respir J.* 2016;48(1):196-204.
81. Madan NK, Madan K, Jain D, et al. Utility of conventional transbronchial needle aspiration with rapid on-site evaluation (c-TBNA-ROSE) at a tertiary care center with endobronchial ultrasound (EBUS) facility. *J Cytol.* 2016;33(1):22-26.
82. Chen CH, Cheng WC, Wu BR, et al. Improved diagnostic yield of bronchoscopy in peripheral pulmonary lesions: combination of radial probe endobronchial ultrasound and rapid on-site evaluation. *J Thorac Dis.* 2015;7(Suppl 4):S418-425.
83. Loo FL, Halligan AM, Port JL, Hoda RS. The emerging technique of electromagnetic navigation bronchoscopy-guided fine-needle aspiration of peripheral lung lesions: promising results in 50 lesions. *Cancer Cytopathol.* 2014;122(3):191-199.
84. Sindhwani G, Rawat J, Chandra S, Kusum A, Rawat M. Transbronchial needle aspiration with rapid on-site evaluation: a prospective study on efficacy, feasibility and cost effectiveness. *Indian J Chest Dis Allied Sci.* 2013;55(3):141-144.
85. Diacon AH, Koegelenberg CF, Schubert P, et al. Rapid on-site evaluation of transbronchial aspirates: randomised comparison of two methods. *Eur Respir J.* 2010;35(6):1216-1220.
86. Conti V, Marchi M, Turrini C, et al. Transbronchial needle aspiration: ThinPrep versus the conventional smear technique in the diagnosis of lung cancer. *Minerva Pneumol.* 2016;55(4):85-93.
87. Swiderek J, Morcos S, Donthireddy V, et al. Prospective study to determine the volume of pleural fluid required to diagnose malignancy. *Chest.* 2010;137(1):68-73.
88. Abouzgheib W, Bartter T, Dagher H, Pratter M, Klump W. A prospective study of the volume of pleural fluid required for accurate diagnosis of malignant pleural effusion. *Chest.* 2009;135(4):999-1001.
89. Buckley O, Benfayed W, Geoghegan T, et al. Thoracocentesis for potential malignancy: does volume matter? *Hong Kong J Radiol.* 2008;11(2):72-75.

90. Wu H, Khosla R, Rohatgi PK, Chauhan SS, Paal E, Chen W. The minimum volume of pleural fluid required to diagnose malignant pleural effusion: a retrospective study. *Lung India.* 2017;34(1):34-37.
91. Rooper LM, Ali SZ, Olson MT. A minimum fluid volume of 75 mL is needed to ensure adequacy in a pleural effusion: a retrospective analysis of 2540 cases. *Cancer Cytopathol.* 2014;122(9):657-665.
92. Thomas SC, Davidson LR, McKean ME. An investigation of adequate volume for the diagnosis of malignancy in pleural fluids. *Cytopathology.* 2011;22(3):179-183.
93. DeMaio A, Clarke JM, Dash R, et al. Yield of malignant pleural effusion for detection of oncogenic driver mutations in lung adenocarcinoma. *J Bronchology Interv Pulmonol.* 2019;26(2):96-101.
94. Florentine BD, Helton RJ, Mitchell MM, Schmidt KE, Kozlov DB. Accuracy and adequacy of computed tomography-guided lung biopsies: experience from a community hospital. *J Am Osteopath Assoc.* 2015;115(10):592-603.
95. Lee K, Um SW, Jeong BH, et al. Triple gene analysis using samples obtained by endobronchial ultrasound-guided transbronchial needle aspiration. *Intern Med.* 2016;55(21):3105-3111.
96. Minca EC, Lanigan CP, Reynolds JP, et al. ALK status testing in non-small-cell lung carcinoma by FISH on ThinPrep slides with cytology material. *J Thorac Oncol.* 2014;9(4):464-468.
97. Ozluk Y, Firat P, Yegen G, Hocaoglu J, Tas S, Yilmazbayhan D. EGFR mutation testing using archival-stained smears in non-small cell lung carcinoma. *Cytopathology.* 2017;28(1):35-45.
98. Wu HH, Eaton JP, Jones KJ, et al. Utilization of cell-transferred cytologic smears in detection of EGFR and KRAS mutation on adenocarcinoma of lung. *Mod Pathol.* 2014;27(7):930-935.
99. Zhang C, Randolph ML, Jones KJ, Cramer HM, Cheng L, Wu HH. Anaplastic lymphoma kinase immunocytochemistry on cell-transferred cytologic smears of lung adenocarcinoma. *Acta Cytol.* 2015;59(2):213-218.
100. Hsiao SH, Chung CL, Lee CM, et al. Suitability of computed tomography-guided biopsy specimens for subtyping and genotyping of non-small-cell lung cancer. *Clin Lung Cancer.* 2013;14(6):719-725.
101. Tian P, Wang Y, Li L, Zhou Y, Luo W, Li W. CT-guided transthoracic core needle biopsy for small pulmonary lesions: diagnostic performance and adequacy for molecular testing. *J Thorac Dis.* 2017;9(2):333-343.
102. Wu HH, Jovonovich SM, Randolph M, et al. Utilization of cell-transfer technique for molecular testing on hematoxylin-eosin-stained sections: a viable option for small biopsies that lack tumor tissues in paraffin block. *Arch Pathol Lab Med.* 2016;140(12):1383-1389.
103. Lin SM, Chung FT, Huang CD, et al. Diagnostic value of endobronchial ultrasonography for pulmonary tuberculosis. *J Thorac Cardiovasc Surg.* 2009;138(1):179-184.
104. Eom JS, Mok JH, Lee MK, et al. Efficacy of TB-PCR using EBUS-TBNA samples in patients with intrathoracic granulomatous lymphadenopathy. *BMC Pulm Med.* 2015;15:166. doi: 10.1186/s12890-015-0162-4
105. Kiral N, Caglayan B, Salepci B, et al. Endobronchial ultrasound-guided transbronchial needle aspiration in diagnosing intrathoracic tuberculous lymphadenitis. *Med Ultrason.* 2015;17(3):333-338.
106. Thangakunam B, Isaac BTJ, Christopher DJ. Endobronchial ultrasound experience in a high tuberculosis prevalence setting. *Indian J Tuberc.* 2017;64(3):196-200.
107. Balshem H, Helfand M, Schunemann HJ, et al. GRADE guidelines: 3. Rating the quality of evidence. *J Clin Epidemiol.* 2011;64(4):401-406.
108. Andrews J, Guyatt G, Oxman AD, et al. GRADE guidelines: 14. Going from evidence to recommendations: the significance and presentation of recommendations. *J Clin Epidemiol.* 2013;66(7):719-725.

109. Moher D, Liberati A, Tetzlaff J, Altman DG. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. *PLoS Med.* 2009;6(7):e1000097. doi: 10.1371/journal.pmed.1000097