Online Appendix B4 BTS Guideline for Pleural Disease

Section B Investigation of the undiagnosed pleural effusion

Question B4 Evidence Review and Protocol

B4 What is the diagnostic accuracy of pleural fluid tests when diagnosing adult patients with unilateral pleural effusion?

Contents

Question Evidence Review

B4 What is the diagnostic accuracy of pleural fluid tests when diagnosing adult patients with unilateral pleural effusion?

Background

Unilateral pleural effusion may result from a variety of diseases, including malignant, inflammatory, infectious and cardiovascular illnesses. Pleural fluid aspiration facilitates measurement of various disease biomarkers. If accurate, these tests may obviate the need for pleural biopsy or other investigations and facilitate early treatment initiation, including early intercostal drainage in patients with complex parapneumonic effusion or empyema.

To address this review, it was first necessary to define the disease states that are of clinical interest in adults presenting with unilateral effusion, and to define a relevant gold standard for each (Table B4a). The index tests reviewed vary with the target disease resulting in five sub-questions, each containing relevant index test-gold standard pairs.

Table B4a: Reviewed disease state subgroups and associated gold standards

Disease State	Gold Standard
Secondary pleural malignancy	Malignant fluid cytology or pleural biopsy, or malignant pleural nodules/thickening on imaging and confirmed extra-pleural primary cancer
Tuberculous pleural effusion (TPE)	Clinical composite, including definite TPE (AAFB in pleural tissue or fluid culture, or sputum AAFB plus effusion) and probable TB (granulomatous histology or lymphocytic fluid, effusion resolved after TB therapy and other causes excluded)
Heart failure	Clinical composite including reduced LVEF on echo +/- MRI
Complex parapneumonic effusion or empyema	Clinical composite including evidence of infection plus purulent fluid, positive culture or Gram's stain, fluid pH <7.2
Autoimmune pleuritis	Clinical compositive based on all available data

Outcomes

Diagnostic accuracy of pleural fluid tests (biomarkers) for diagnosing unilateral pleural effusion

Evidence Review

The initial literature review identified 261 studies, of which 45 were deemed relevant (please see individual sections for references). Biomarker studies that did not report or constitute an external validation cohort were excluded.

The evidence review has been split into five sections based on the five target diseases listed in Table B4a.

1. Secondary pleural malignancy

12 eligible studies reported on 12 biomarkers, but each study did not include all biomarkers.[1-11] Reasons for study exclusion included use of an inappropriate gold standard (e.g. negative cytology but visceral malignancy elsewhere) or early phase biomarkers. All mesothelioma studies were excluded. Meta-analyses were possible for cytology[1-7] (Figure B4a) and five protein biomarkers (CEA[4,6-12], CYFRA21-1[4,10,12], CA19-9[6,10,11], CA15-3[6,7,9-12] and CA72-4[6,7,11]) (Figure B4b, Figure B4c, Figure B4d, Figure B4e and Figure B4f respectively).

The pooled sensitivity of fluid cytology was low (0.46 [0.40, 0.52] [95% confidence intervals]) but highly specific, with positive results obviating the need for further diagnostic sampling (Table B4b). However, this analysis did not address the quality of predictive information provided (e.g. for response to targeted cancer therapies).

All protein biomarkers offered poor sensitivity (Table B4b). Although specificity was generally high (all pooled estimates ≥0.88, Table 6), positive results would not obviate the requirement for tissue sampling, further limiting clinical utility.

Table B4b: Summary of secondary pleural malignancy biomarker point estimate sensitivities and specificities

Biomarker	Contributing studies (n)	Sensitivity [95% CI]	Specificity [95% CI]
Cytology[1-7]	7	0.46 [0.40, 0.52]	1.00 [0.00, 1.00]
CEA[4,6-12]	8	0.54 [0.40, 0.68]	1.00 [0.96, 1.00]
CYFRA 21-1[4,10,12]	3	0.58 [0.48, 0.67]	0.88 [0.78, 0.94]
CA19-9[6,10,11]	3	0.22 [0.18 0.27]	1.00 [0.00,1.00]
CA15-3[6,7,9-12]	6	0.44 [0.39, 0.50]	0.99 [0.97, 1.00]
CA72-4[6,7,11]	3	0.38 [0.30, 0.46]	0.99 [0.97, 1.00]

CA 15-3 – cancer antigen 15-3; CA 19-9 – carbohydrate antigen 19-9; CA72-4 – cancer antigen 72-4; CEA – carcinoembryonic antigen; CI – confidence intervals; CYFRA 21-1 – fragment of cytokeratin 19

2. Tuberculous pleural effusion (TPE)

Twenty-four studies looked at tuberculous pleural effusion (TPE).[13-36] All evaluated adenosine deaminase (ADA) as a single marker, or in combination with other tests in a mixture of blood and pleural fluid. Reasons for study exclusion included recruitment of selected populations (e.g. lymphocytic effusion) and use of serum markers. Only data regarding pleural fluid measurements were extracted from included studies, allowing meta-analyses of two markers, ADA[13-36] (Figure B4g) and interferon gamma (IFN-gamma)[14,16,20-22,36] (Figure B4h).

The sensitivity and specificity of ADA and IFN-gamma were notably high and most studies were reasonably sized (Table B4c). However, the prevalence of TPE was >10% in all but two studies and very high in some (≥70% in three studies). Greater imprecision was notable in the performance estimates of the two lowest prevalence studies contributing to ADA.[15,32]

Table B4c: Summary of tuberculous pleural effusion biomarker point estimate sensitivities and specificities

Biomarker	Contributing studies (n)	Sensitivity [95% CI]	Specificity [95% CI]
ADA[13-36]	24	0.91 [0.87, 0.93]	0.88 [0.86, 0.93]
IFN-gamma[14,16,20-22,36]	6	0.95 [0.85, 0.98]	0.96 [0.90, 0.98]

ADA **–** adenosine deaminase; CI – confidence intervals; IFN-gamma – interferon gamma

3. Heart failure

Five studies evaluated N-terminal pro hormone BNP (NT-proBNP) for diagnosing pleural effusion in heart failure, but each used a different cut-point to define a positive test result. A summary of the cut-points is provided in Table B4d. Meta-analysis of these studies gave a pooled sensitivity of 0.93 [0.88, 0.96] and pooled specificity of 0.93 [0.86, 0.97] [95% confidence intervals] (Figure B4i).[37-41]

Table B4d: Heart failure N-terminal pro hormone BNP (NT-proBNP) cut-point per study

Study	Cut-point
Kolditz 2006[37]	4000 ng/L
Han 2008[38]	1714 pg/ml
Bayram 2009[39]	925 ng/L
Porcel 2009[40]	1300 pg/ml
Valdes 2011[41]	1409 pg/ml

4. Pleural infection (complex parapneumonic effusion (CPPE) or empyema)

There were no studies that directly investigated the diagnostic accuracy of pleural fluid tests for diagnosing pleural infection (complex parapneumonic effusion (CPPE) or empyema). This was primarily due to the use of inappropriate reference standards, failure to adequately describe reference standards used, discovery biomarker analyses without validation and the use of biomarkers for prognostic, not diagnostic, analyses.

5. Autoimmune pleuritis

Four studies evaluated pleural fluid antinuclear antibody (ANA) as a biomarker of lupus pleuritic and meta-analysis of the results gave a pooled sensitivity of 0.94 [0.72, 0.99] and a pooled specificity of 0.87 [0.77, 0.93] [95% confidence intervals] relative to cases with non-lupus effusion (Figure B4j).[42-45]

Evidence Statements

Pleural fluid biomarkers do not provide improved sensitivity, when compared with cytology, for diagnosing secondary pleural malignancy (**Low**)

Pleural fluid adenosine deaminase (ADA) and interferon gamma (IFN-gamma) provide high sensitivity and specificity for diagnosing tuberculous pleural effusion (**Very Low**)

Pleural fluid N-terminal pro hormone pro-brain natriuretic peptide (NT-proBNP) provides high sensitivity and specificity for diagnosing heart failure in unilateral pleural effusion patients (**Very Low**)

Pleural fluid antinuclear antibody (ANA) provides high sensitivity and specificity for diagnosing lupus pleural effusion (**Low**)

Recommendations

- Pleural fluid cytology should be used as an initial diagnostic test in patients with suspected secondary pleural malignancy, accepting that a negative cytology should lead to consideration of further investigation (**Conditional**)
- Pleural fluid biomarkers should not be used for diagnosing secondary pleural malignancy (**Conditional**)
- In high prevalence populations, pleural fluid adenosine deaminase (ADA) and/or interferon gamma (IFN-gamma) test(s) can be considered for diagnosing tuberculous pleural effusion (**Conditional**)
- In low prevalence populations, pleural fluid adenosine deaminase (ADA) can be considered as an exclusion test for tuberculous pleural effusion (**Conditional**)
- Tissue sampling for culture and sensitivity should be the preferred option for all patients with suspected tuberculous pleural effusion (**Strong** – by consensus)
- Pleural fluid antinuclear antibody (ANA) should be considered to support a diagnosis of lupus pleuritis (**Conditional**)

Good Practice Points

- ✓ The clinical utility of pleural fluid cytology varies by tumour sub-type, including diagnostic sensitivity and predictive value for response to subsequent cancer therapies. This should be taken into consideration when planning the most suitable diagnostic strategy (for example, direct biopsies in those with a likely low cytological yield can be considered)
- ✓ Pleural fluid N-terminal pro brain natriuretic peptide (NT-proBNP) is useful when considering heart failure as a cause in unilateral pleural effusions but not superior to serum NT-proBNP and therefore should not be ordered routinely

Research Recommendations

- Further research is needed to determine the clinical utility of pleural fluid cytology, as defined by combined diagnostic and predictive value, in individual tumour sub-types
- Further research is needed into determining the diagnostic value of pleural fluid markers for tuberculous pleural effusion, heart failure and auto-immune pleuritis, to improve the strength of the clinical recommendations

Meta-analyses

Diagnostic accuracy table contents and summary receiver operating characteristic (SROC) curve legend

Table contents

Pooled sensitivity [95% confidence intervals]

Pooled specificity [95% confidence intervals]

Likelihood ratio of a positive test result (LR+) [95% confidence intervals]

Likelihood ratio of a negative test result (LR-) [95% confidence intervals]

Diagnostic odds ratio (DOR, an indicator of the likelihood of a positive test result) [95% confidence intervals]

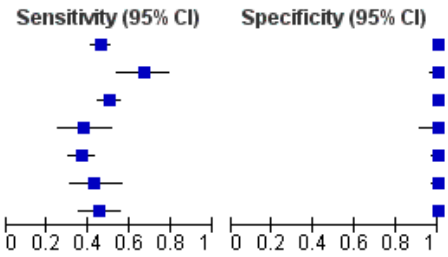
Summary receiver operating characteristic (SROC) curve legend

——	SROC
○	Study estimate
●	Summary point
·····	95% confidence region
- - -	95% prediction region

1. Secondary pleural malignancy

Figure B4a Cytology

Study	TP	FP	FN	TN	Sensitivity (95% CI)	Specificity (95% CI)
Arnold 2018	237	0	278	406	0.46 [0.42, 0.50]	1.00 [0.99, 1.00]
Loveland 2018	41	0	20	92	0.67 [0.54, 0.79]	1.00 [0.96, 1.00]
Mercer 2020	162	0	163	673	0.50 [0.44, 0.55]	1.00 [0.99, 1.00]
Sun 2014	22	0	36	40	0.38 [0.26, 0.52]	1.00 [0.91, 1.00]
Tsim 2019	87	0	151	125	0.37 [0.30, 0.43]	1.00 [0.97, 1.00]
Villena 1996	28	0	37	142	0.43 [0.31, 0.56]	1.00 [0.97, 1.00]
Villena 2003	46	0	55	151	0.46 [0.36, 0.56]	1.00 [0.98, 1.00]

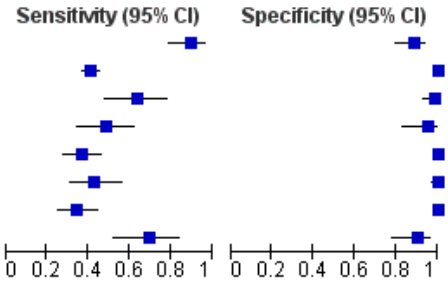


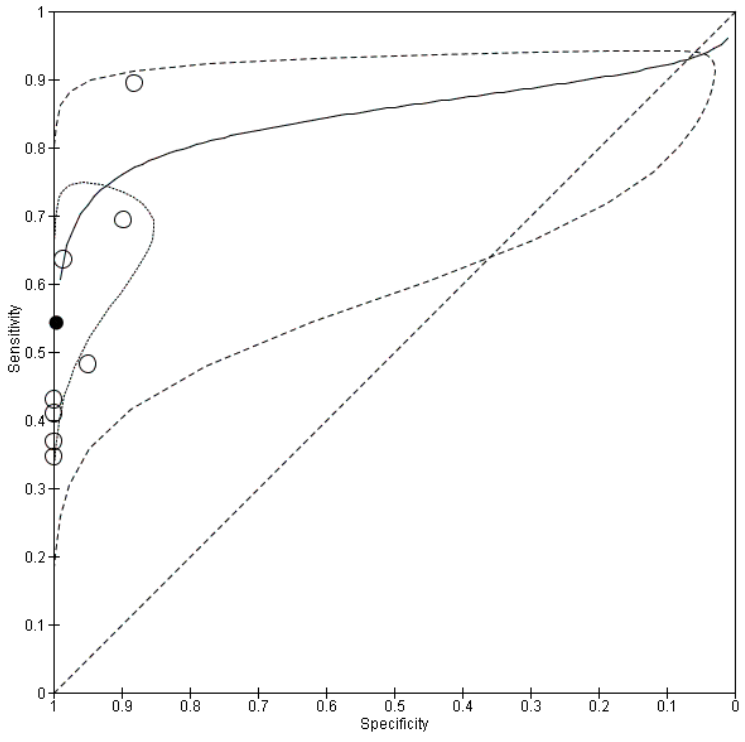
Pooled Sensitivity	0.461	[0.401, 0.522]
Pooled Specificity	1.000	[0.000, 1.000]
LR+	∞	[NaN, NaN]
LR-	0.539	[NaN, NaN]
DOR	∞	[NaN, NaN]

NaN – not a number

Figure B4b CEA (carcinoembryonic antigen)

Study	TP	FP	FN	TN	Sensitivity (95% CI)	Specificity (95% CI)
Ji 2018	51	11	6	82	0.89 [0.78, 0.96]	0.88 [0.80, 0.94]
Porcel 2017	225	0	324	1026	0.41 [0.37, 0.45]	1.00 [1.00, 1.00]
Shitrit 2005	28	1	16	71	0.64 [0.48, 0.78]	0.99 [0.93, 1.00]
Sun 2014	28	2	30	38	0.48 [0.35, 0.62]	0.95 [0.83, 0.99]
Trape 2017	45	0	77	280	0.37 [0.28, 0.46]	1.00 [0.99, 1.00]
Villena 1996	28	0	37	142	0.43 [0.31, 0.56]	1.00 [0.97, 1.00]
Villena 2003	35	0	66	151	0.35 [0.25, 0.45]	1.00 [0.98, 1.00]
Wagner 2007	25	5	11	44	0.69 [0.52, 0.84]	0.90 [0.78, 0.97]

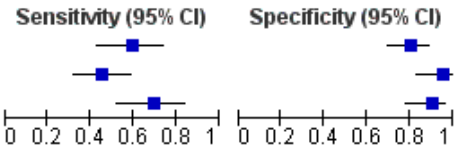




Pooled Sensitivity	0.543	[0.404, 0.675]
Pooled Specificity	0.996	[0.961, 1.000]
LR+	129.616	[-143.686, 402.918]
LR-	0.459	[0.322, 0.596]
DOR	282.412	[-271.203, 836.028]

Figure B4c CYFRA 21-1 (fragment of cytokeratin 19)

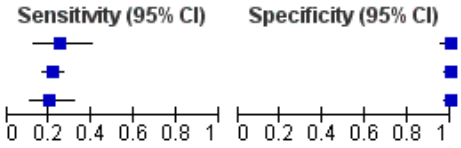
Study	TP	FP	FN	TN	Sensitivity (95% CI)	Specificity (95% CI)
Shitrit 2005	26	14	18	58	0.59 [0.43, 0.74]	0.81 [0.70, 0.89]
Sun 2014	26	2	32	38	0.45 [0.32, 0.58]	0.95 [0.83, 0.99]
Wagner 2007	25	5	11	44	0.69 [0.52, 0.84]	0.90 [0.78, 0.97]



Pooled Sensitivity	0.578	[0.477, 0.674]
Pooled Specificity	0.885	[0.782, 0.943]
LR+	5.042	[1.831, 8.252]
LR-	0.476	[0.371, 0.582]
DOR	10.588	[2.863, 18.314]

Figure B4d CA19-9 (carbohydrate antigen 19-9)

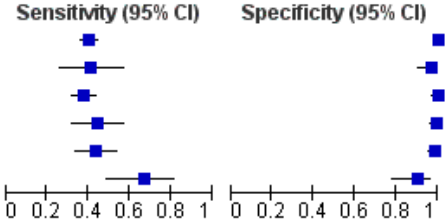
Study	TP	FP	FN	TN	Sensitivity (95% CI)	Specificity (95% CI)
Shitrit 2005	11	0	33	72	0.25 [0.13, 0.40]	1.00 [0.95, 1.00]
Trape 2017	62	0	218	122	0.22 [0.17, 0.27]	1.00 [0.97, 1.00]
Villena 1996	13	0	52	142	0.20 [0.11, 0.32]	1.00 [0.97, 1.00]

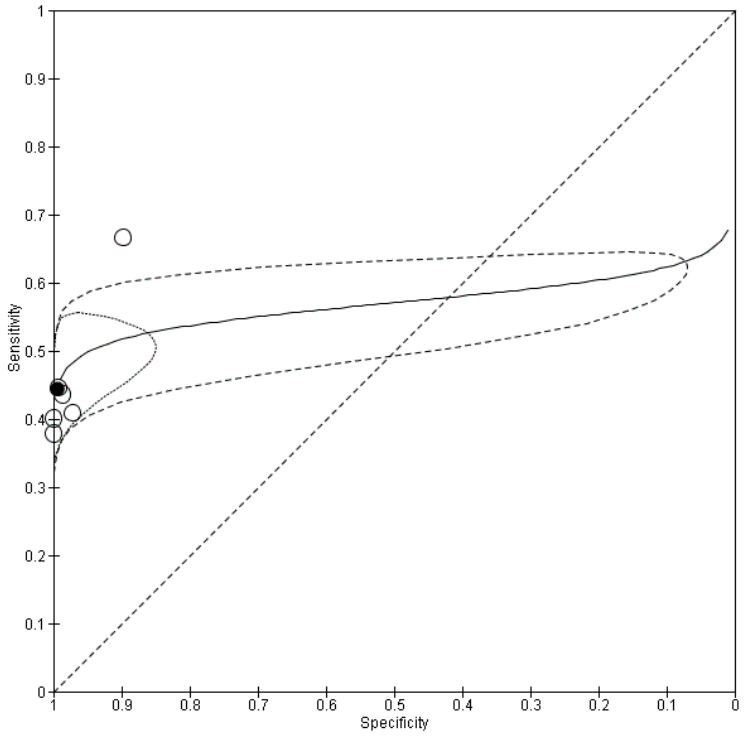


Pooled Sensitivity	0.221	[0.183, 0.265]
Pooled Specificity	1.000	[0.000, 1.000]
LR+	∞	[-∞, ∞]
LR-	0.779	[0.738, 0.820]
DOR	∞	[-∞, ∞]

Figure B4e CA15-3 (cancer antigen 15-3)

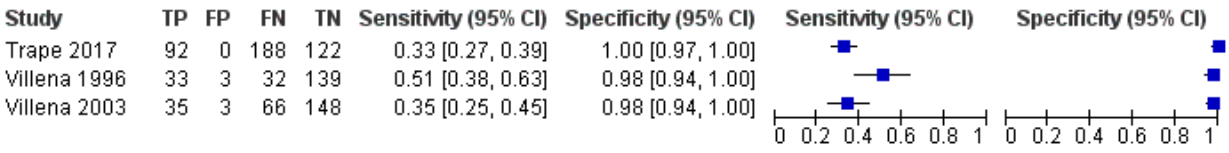
Study	TP	FP	FN	TN	Sensitivity (95% CI)	Specificity (95% CI)
Porcel 2017	220	0	329	1026	0.40 [0.36, 0.44]	1.00 [1.00, 1.00]
Shitrit 2005	18	2	26	70	0.41 [0.26, 0.57]	0.97 [0.90, 1.00]
Trape 2017	106	0	174	122	0.38 [0.32, 0.44]	1.00 [0.97, 1.00]
Villena 1996	29	1	36	141	0.45 [0.32, 0.57]	0.99 [0.96, 1.00]
Villena 2003	44	2	57	149	0.44 [0.34, 0.54]	0.99 [0.95, 1.00]
Wagner 2007	24	5	12	44	0.67 [0.49, 0.81]	0.90 [0.78, 0.97]





Pooled Sensitivity	0.444	[0.387, 0.503]
Pooled Specificity	0.994	[0.966, 0.999]
LR+	76.255	[-54.570, 207.080]
LR-	0.559	[0.503, 0.615]
DOR	136.410	[-92.434, 365.255]

Figure B4f CA72-4 (cancer antigen 72-4)

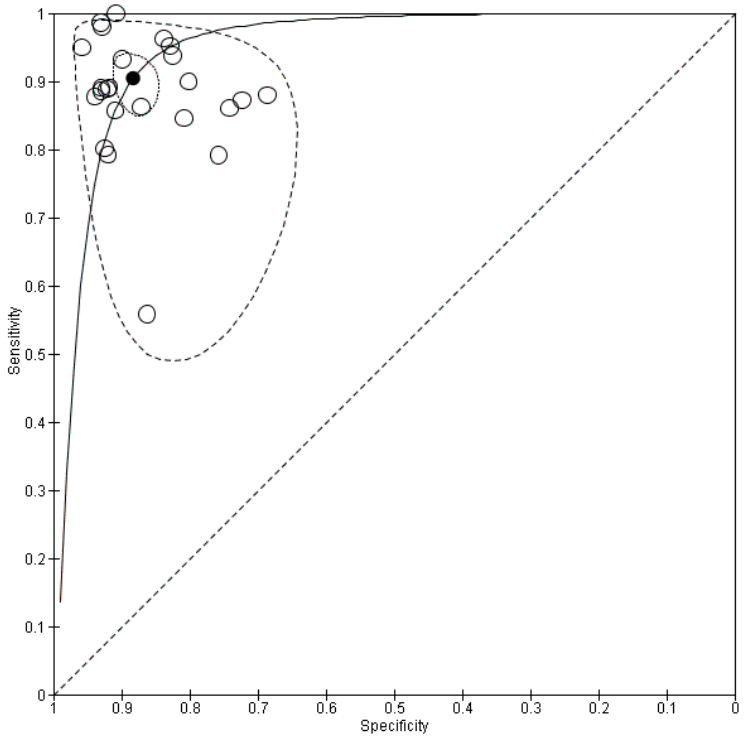


Pooled Sensitivity	0.380	[0.303, 0.464]
Pooled Specificity	0.987	[0.965, 0.995]
LR+	28.955	[1.324, 56.586]
LR-	0.628	[0.548, 0.708]
DOR	46.124	[1.747, 90.501]

2. Tuberculous pleural effusion (TPE)

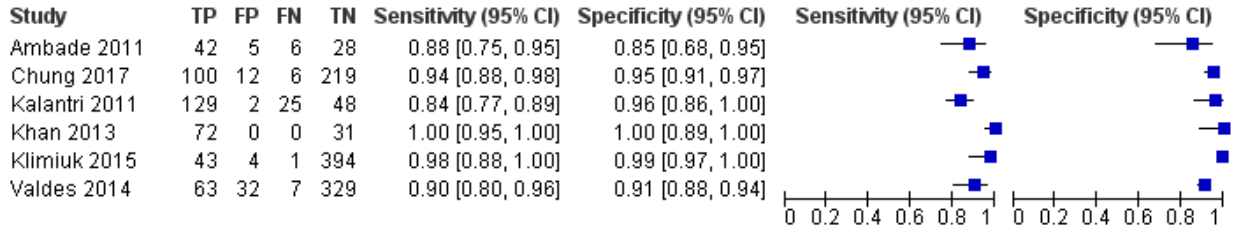
Figure B4g ADA (adenosine deaminase)

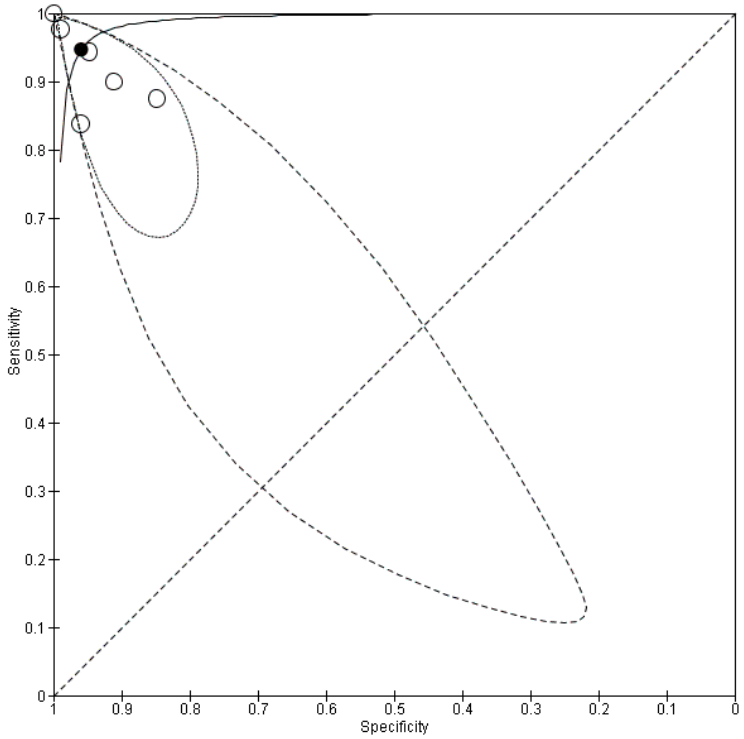
Study	TP	FP	FN	TN	Sensitivity (95% CI)	Specificity (95% CI)
Abrao 2014	155	11	19	124	0.89 [0.83, 0.93]	0.92 [0.86, 0.96]
Ambade 2011	38	8	10	25	0.79 [0.65, 0.90]	0.76 [0.58, 0.89]
Arnold 2015	6	30	1	301	0.86 [0.42, 1.00]	0.91 [0.87, 0.94]
Chung 2017	93	14	13	216	0.88 [0.80, 0.93]	0.94 [0.90, 0.97]
Demirer 2012	137	26	20	68	0.87 [0.81, 0.92]	0.72 [0.62, 0.81]
Garcia-Zamalloa 2012	65	28	8	371	0.89 [0.80, 0.95]	0.93 [0.90, 0.95]
He 2015	109	6	27	74	0.80 [0.72, 0.86]	0.93 [0.84, 0.97]
Kalantri 2011	122	4	32	46	0.79 [0.72, 0.85]	0.92 [0.81, 0.98]
Khan 2013	62	8	10	23	0.86 [0.76, 0.93]	0.74 [0.55, 0.88]
Klimiuk 2015	39	28	5	370	0.89 [0.75, 0.96]	0.93 [0.90, 0.95]
Kosar 2015	44	16	6	35	0.88 [0.76, 0.95]	0.69 [0.54, 0.81]
Liao 2014	157	7	124	44	0.56 [0.50, 0.62]	0.86 [0.74, 0.94]
Neves 2007	99	19	5	92	0.95 [0.89, 0.98]	0.83 [0.75, 0.89]
Ogata 2011	107	40	17	271	0.86 [0.79, 0.92]	0.87 [0.83, 0.91]
Perez-Rodriguez 1999	24	6	3	70	0.89 [0.71, 0.98]	0.92 [0.84, 0.97]
Porcel 2010	55	50	4	446	0.93 [0.84, 0.98]	0.90 [0.87, 0.92]
Riantawan 1999	95	4	5	93	0.95 [0.89, 0.98]	0.96 [0.90, 0.99]
Sahn 2013	537	30	11	393	0.98 [0.96, 0.99]	0.93 [0.90, 0.95]
Sivakumar 2017	26	17	1	88	0.96 [0.81, 1.00]	0.84 [0.75, 0.90]
Skouras 2015	9	22	1	89	0.90 [0.55, 1.00]	0.80 [0.72, 0.87]
Tay 2013	75	14	5	66	0.94 [0.86, 0.98]	0.82 [0.72, 0.90]
Tural Onur 2015	44	13	8	55	0.85 [0.72, 0.93]	0.81 [0.70, 0.89]
Valdes 1996	76	25	0	249	1.00 [0.95, 1.00]	0.91 [0.87, 0.94]
Valdes 2014	69	25	1	336	0.99 [0.92, 1.00]	0.93 [0.90, 0.95]



Pooled Sensitivity	0.905	[0.865, 0.933]
Pooled Specificity	0.883	[0.856, 0.906]
LR+	7.744	[5.980, 9.508]
LR-	0.108	[0.069, 0.147]
DOR	71.693	[35.089, 108.298]

Figure B4h IFN-gamma (interferon gamma)



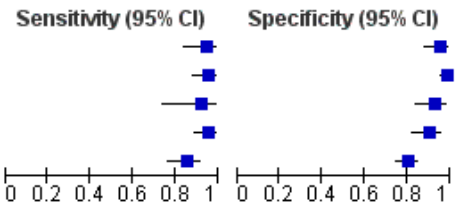


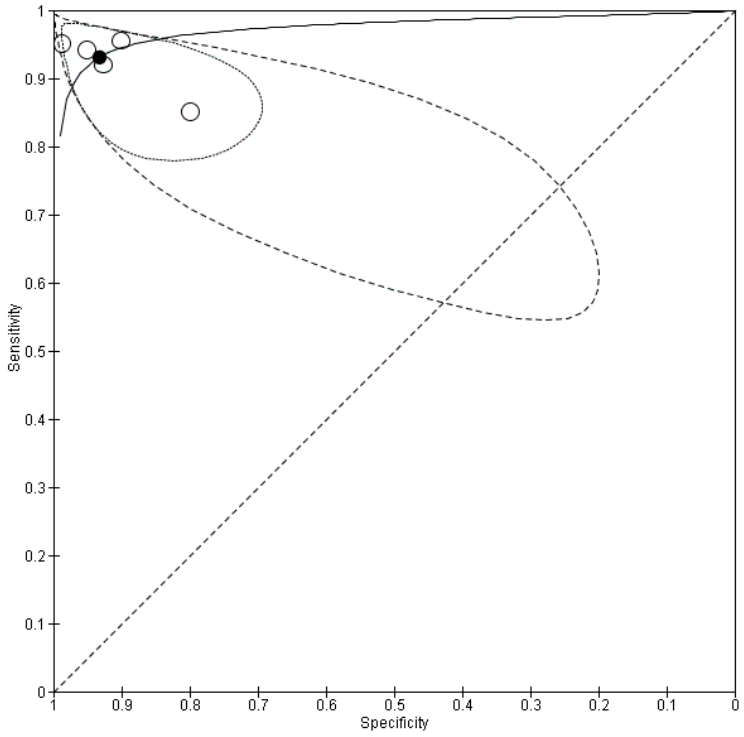
Pooled Sensitivity	0.947	[0.851, 0.982]
Pooled Specificity	0.959	[0.899, 0.984]
LR+	23.363	[0.339, 46.388]
LR-	0.055	[-0.006, 0.117]
DOR	421.957	[-422.547, 1266.461]

3. Heart failure

Figure B4i NT-proBNP (N-terminal pro hormone BNP)

Study	TP	FP	FN	TN	Sensitivity (95% CI)	Specificity (95% CI)
Bayram 2009	48	4	3	78	0.94 [0.84, 0.99]	0.95 [0.88, 0.99]
Han 2008	78	2	4	156	0.95 [0.88, 0.99]	0.99 [0.96, 1.00]
Kolditz 2006	23	5	2	63	0.92 [0.74, 0.99]	0.93 [0.84, 0.98]
Porcel 2009	86	9	4	82	0.96 [0.89, 0.99]	0.90 [0.82, 0.95]
Valdes 2011	80	61	14	243	0.85 [0.76, 0.92]	0.80 [0.75, 0.84]



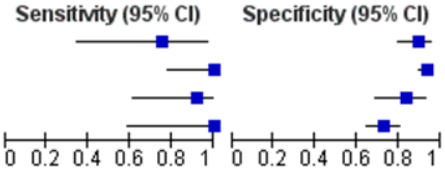


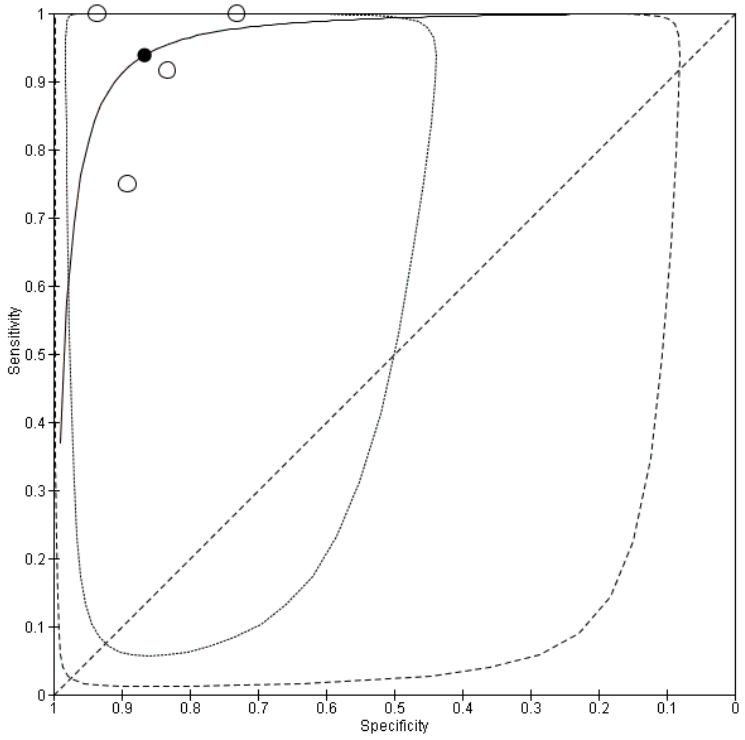
Pooled Sensitivity	0.931	[0.880, 0.961]
Pooled Specificity	0.932	[0.860, 0.969]
LR+	13.783	[2.971, 24.595]
LR-	0.074	[0.030, 0.118]
DOR	185.717	[-49.979, 421.413]

5. Autoimmune pleuritis

Figure B4j ANA (antinuclear antibody)

Study	TP	FP	FN	TN	Sensitivity (95% CI)	Specificity (95% CI)
Khare 1994	6	8	2	66	0.75 [0.35, 0.97]	0.89 [0.80, 0.95]
Porcel 2007	15	16	0	235	1.00 [0.78, 1.00]	0.94 [0.90, 0.96]
Toworakul 2011	11	7	1	35	0.92 [0.62, 1.00]	0.83 [0.69, 0.93]
Wang 2000	7	32	0	87	1.00 [0.59, 1.00]	0.73 [0.64, 0.81]



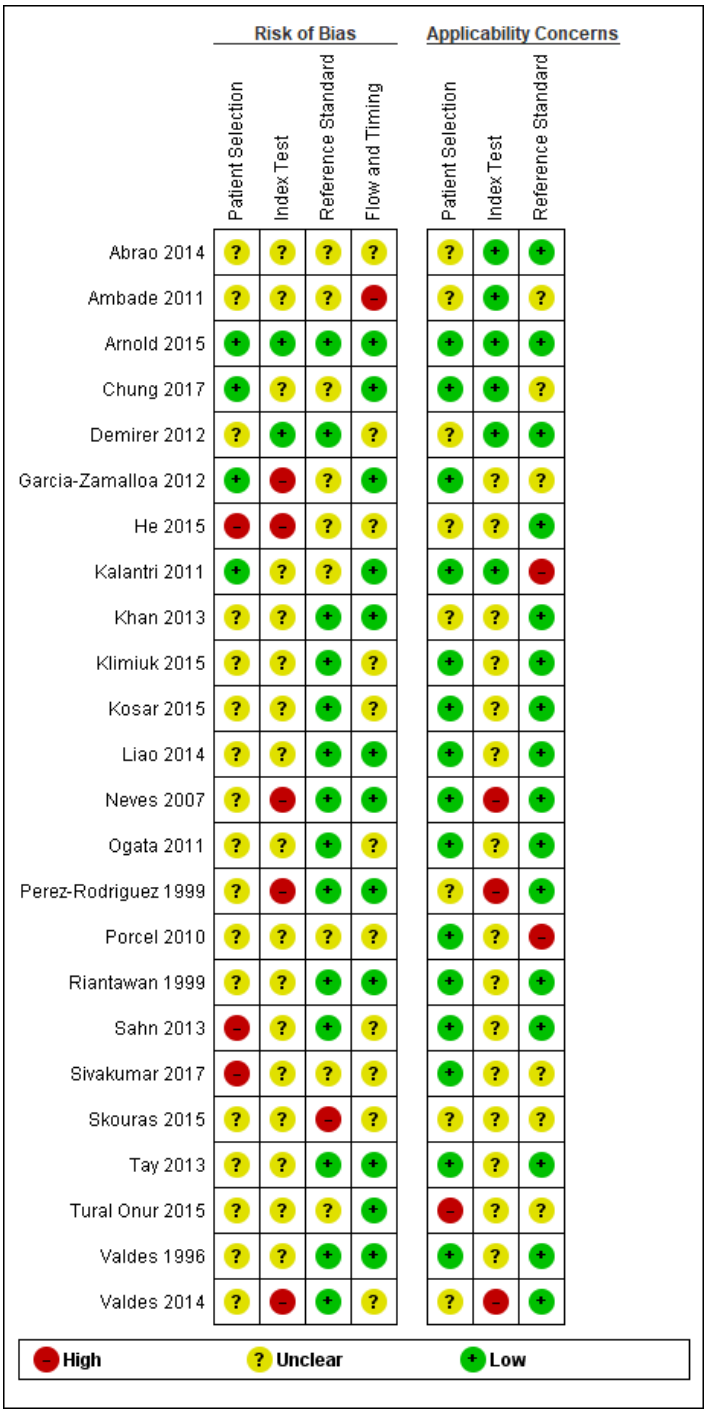


Pooled Sensitivity	0.938	[0.724, 0.989]
Pooled Specificity	0.867	[0.768, 0.927]
LR+	7.030	[2.832, 11.228]
LR-	0.071	[-0.047, 0.189]
DOR	98.995	[-89.290, 287.281]

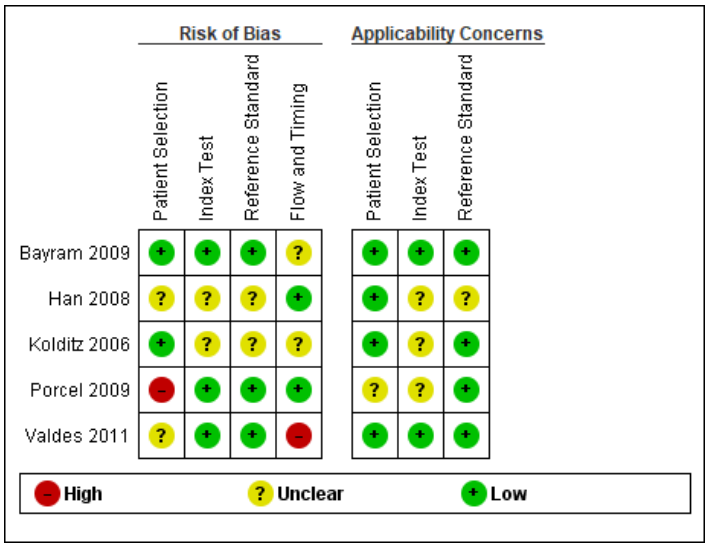
Risk of bias summaries

1. Secondary pleural malignancy

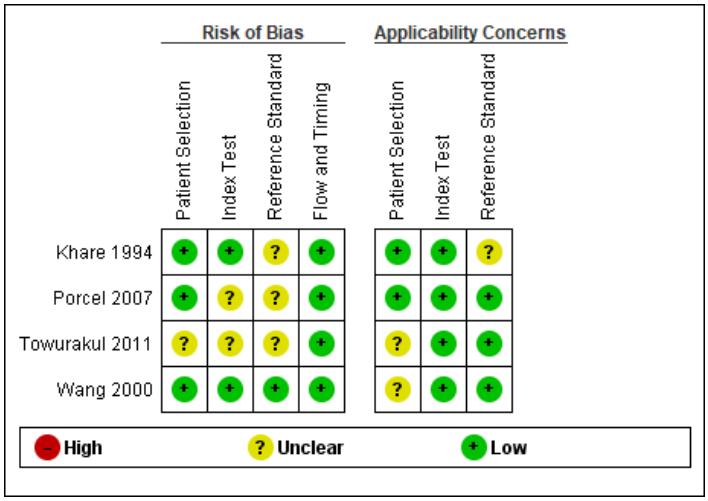
2. Tuberculous pleural effusion (TPE)



3. Heart failure



5. Autoimmune pleuritis



GRADE analyses

1. Secondary pleural malignancy

Cytology

What is the diagnostic accuracy of pleural fluid tests when diagnosing adult patients with unilateral pleural effusion?

Patient or population: Adults aged 18+ with unilateral pleural effusion

New test: Cytology

Pooled sensitivity: 0.46 (95% CI: 0.40 to 0.52) | **Pooled specificity**: 1.00 (95% CI: 0.00 to 1.00)

Test result	Number of results per 1,000 patients tested (95% CI) **Prevalence** 45% Typically seen in	Number of participants (studies)	Certainty of the Evidence (GRADE)
True positives	**207** (180 to 235)	1363 (7)	⨁⨁◯◯ **LOW** [a,b]
False negatives	**243** (215 to 270)		
True negatives	**550** (1 to 550)	1629 (7)	⨁⨁◯◯ **LOW** [a,b]
False positives	**0** (0 to 549)		

CI: Confidence interval

Explanations

a. Some risk of bias across studies
b. Some indirectness across studies

CEA (carcinoembryonic antigen)

What is the diagnostic accuracy of pleural fluid tests when diagnosing adult patients with unilateral pleural effusion?

Patient or population: Adults aged 18+ with unilateral pleural effusion

New test: CEA (carcinoembryonic antigen)

Pooled sensitivity: 0.54 (95% CI: 0.40 to 0.68) | **Pooled specificity**: 1.00 (95% CI: 0.96 to 1.00)

Test result	Number of results per 1,000 patients tested (95% CI) **Prevalence** 45%* Typically seen in	Number of participants (studies)	Certainty of the Evidence (GRADE)
True positives	**244** (182 to 304)	1032 (8)	⨁◯◯◯ **VERY LOW** [a,b,c]
False negatives	**206** (146 to 268)		
True negatives	**548** (529 to 550)	1853 (8)	⨁◯◯◯ **VERY LOW** [a,b,c]
False positives	**2** (0 to 21)		

CI: Confidence interval

Explanations

a. Serious risk of bias across studies
b. Some indirectness across studies
c. Some inconsistency across sensitivities

* Mean prevalence across 8 studies

CYFRA 21-1 (fragment of cytokeratin 19)

What is the diagnostic accuracy of pleural fluid tests when diagnosing adult patients with unilateral pleural effusion?

Patient or population: Adults aged 18+ with unilateral pleural effusion

New test: CYFRA 21-1 (fragment of cytokeratin 19)

Pooled sensitivity: 0.58 (95% CI: 0.48 to 0.67) | **Pooled specificity**: 0.88 (95% CI: 0.78 to 0.94)

Test result	Number of results per 1,000 patients tested (95% CI) **Prevalence** 46%* Typically seen in	Number of participants (studies)	Certainty of the Evidence (GRADE)
True positives	**266** (219 to 310)	138 (3)	⨁◯◯◯ **VERY LOW** [a,b,c]
False negatives	**194** (150 to 241)		
True negatives	**478** (422 to 509)	161 (3)	⨁◯◯◯ **VERY LOW** [a,b,c]
False positives	**62** (31 to 118)		

CI: Confidence interval

Explanations

a. Serious risk of bias across studies
b. Some indirectness across studies
c. Some inconsistency across sensitivities

* Mean prevalence across 3 studies

CA19-9 (carbohydrate antigen 19-9)

What is the diagnostic accuracy of pleural fluid tests when diagnosing adult patients with unilateral pleural effusion?

Patient or population: Adults aged 18+ with unilateral pleural effusion

New test: CA19-9 (carbohydrate antigen 19-9)

Pooled sensitivity: 0.22 (95% CI: 0.18 to 0.27) | **Pooled specificity**: 1.00 (95% CI: 0.00 to 1.00)

Test result	Number of results per 1,000 patients tested (95% CI) **Prevalence** 44%* Typically seen in	Number of participants (studies)	Certainty of the Evidence (GRADE)
True positives	**97** (81 to 117)	389 (3)	⨁⨁⨁◯ **MODERATE** [a]
False negatives	**343** (323 to 359)		
True negatives	**560** (6 to 560)	336 (3)	⨁⨁⨁◯ **MODERATE** [a]
False positives	**0** (0 to 554)		

CI: Confidence interval

Explanations

a. Some risk of bias across studies

* Mean prevalence across 3 studies

CA15-3 (cancer antigen 15-3)

What is the diagnostic accuracy of pleural fluid tests when diagnosing adult patients with unilateral pleural effusion?

Patient or population: Adults aged 18+ with unilateral pleural effusion

New test: CA15-3 (cancer antigen 15-3)

Pooled sensitivity: 0.44 (95% CI: 0.39 to 0.50) | **Pooled specificity**: 0.99 (95% CI: 0.97 to 1.00)

Test result	Number of results per 1,000 patients tested (95% CI) **Prevalence** 43% Typically seen in	Number of participants (studies)	Certainty of the Evidence (GRADE)
True positives	**191** (166 to 216)	1075 (6)	⨁◯◯◯ **VERY LOW** [a,b]
False negatives	**239** (214 to 264)		
True negatives	**567** (551 to 569)	1562 (6)	⨁◯◯◯ **VERY LOW** [a,b]
False positives	**3** (1 to 19)		

CI: Confidence interval

Explanations

a. Serious risk of bias across studies
b. Some indirectness across studies

* Mean prevalence across 8 studies

CA72-4 (cancer antigen 72-4)

What is the diagnostic accuracy of pleural fluid tests when diagnosing adult patients with unilateral pleural effusion?

Patient or population: Adults aged 18+ with unilateral pleural effusion

New test: CA72-4 (cancer antigen 72-4)

Pooled sensitivity: 0.38 (95% CI: 0.30 to 0.46) | **Pooled specificity**: 0.99 (95% CI: 0.97 to 1.00)

Test result	Number of results per 1,000 patients tested (95% CI)	Number of participants (studies)	Certainty of the Evidence (GRADE)
	Prevalence 47%* Typically seen in		
True positives	**179** (142 to 218)	446 (3)	⨁⨁◯◯ **LOW** [a,b]
False negatives	**291** (252 to 328)		
True negatives	**523** (511 to 527)	415 (3)	⨁⨁◯◯ **LOW** [a,b]
False positives	**7** (3 to 19)		
	Prevalence 31%* Typically seen in		
True positives	**118** (94 to 144)	446 (3)	⨁⨁◯◯ **LOW** [a,b]
False negatives	**192** (166 to 216)		
True negatives	**681** (666 to 687)	415 (3)	⨁⨁◯◯ **LOW** [a,b]
False positives	**9** (3 to 24)		
	Prevalence 70%* Typically seen in		
True positives	**266** (212 to 325)	446 (3)	⨁⨁◯◯ **LOW** [a,b]
False negatives	**434** (375 to 488)		
True negatives	**296** (290 to 299)	415 (3)	⨁⨁◯◯ **LOW** [a,b]
False positives	**4** (1 to 10)		

CI: Confidence interval

Explanations

a. Some risk of bias across studies
b. Some inconsistency across sensitivities

* Mean (47%), lowest (31%) and highest (70%) prevalence across 3 studies

2. Tuberculous pleural effusion (TPE)

ADA (adenosine deaminase)

What is the diagnostic accuracy of pleural fluid tests when diagnosing adult patients with unilateral pleural effusion?

Patient or population: Adults aged 18+ with unilateral pleural effusion

New test: ADA (adenosine deaminase)

Pooled sensitivity: 0.91 (95% CI: 0.87 to 0.93) | **Pooled specificity**: 0.88 (95% CI: 0.86 to 0.91)

Test result	Number of results per 1,000 patients tested (95% CI)	Number of participants (studies)	Certainty of the Evidence (GRADE)
	Prevalence 40%* Typically seen in		
True positives	**362** (346 to 373)	2579 (24)	⨁◯◯◯ **VERY LOW** [a,b]
False negatives	**38** (27 to 54)		
True negatives	**530** (514 to 544)	4396 (24)	⨁◯◯◯ **VERY LOW** [a,b]
False positives	**70** (56 to 86)		
	Prevalence 2%* Typically seen in		
True positives	**18** (17 to 19)	2579 (24)	⨁◯◯◯ **VERY LOW** [a,b]
False negatives	**2** (1 to 3)		
True negatives	**865** (839 to 888)	4396 (24)	⨁◯◯◯ **VERY LOW** [a,b]
False positives	**115** (92 to 141)		
	Prevalence 8%* Typically seen in		
True positives	**72** (69 to 75)	2579 (24)	⨁◯◯◯ **VERY LOW** [a,b]
False negatives	**8** (5 to 11)		
True negatives	**812** (788 to 834)	4396 (24)	⨁◯◯◯ **VERY LOW** [a,b]
False positives	**108** (86 to 132)		

CI: Confidence interval

Explanations

a. Serious risk of bias across studies
b. Serious indirectness across

* Typical prevalence in 22/24 studies (40%); 1/24 studies (2%); 1/24 studies (8%)

IFN-gamma (interferon gamma)

What is the diagnostic accuracy of pleural fluid tests when diagnosing adult patients with unilateral pleural effusion?

Patient or population: Adults aged 18+ with unilateral pleural effusion

New test: IFN-gamma (interferon gamma)

Pooled sensitivity: 0.95 (95% CI: 0.85 to 0.98) | **Pooled specificity**: 0.96 (95% CI: 0.90 to 0.98)

Test result	Number of results per 1,000 patients tested (95% CI)	Number of participants (studies)	Certainty of the Evidence (GRADE)
	Prevalence 43%* Typically seen in		
True positives	**407** (366 to 422)	494 (6)	⨁◯◯◯ **VERY LOW** [a,b]
False negatives	**23** (8 to 64)		
True negatives	**547** (512 to 561)	1104 (6)	⨁◯◯◯ **VERY LOW** [a,b]
False positives	**23** (9 to 58)		
	Prevalence 70%* Typically seen in		
True positives	**663** (596 to 687)	494 (6)	⨁◯◯◯ **VERY LOW** [a,b]
False negatives	**37** (13 to 104)		
True negatives	**288** (270 to 295)	1104 (6)	⨁◯◯◯ **VERY LOW** [a,b]
False positives	**12** (5 to 30)		
	Prevalence 13%* Typically seen in		
True positives	**123** (111 to 128)	494 (6)	⨁◯◯◯ **VERY LOW** [a,b]
False negatives	**7** (2 to 19)		
True negatives	**834** (782 to 856)	1104 (6)	⨁◯◯◯ **VERY LOW** [a,b]
False positives	**36** (14 to 88)		

CI: Confidence interval

Explanations

a. Serious risk of bias across studies
b. Some indirectness across

* Mean prevalence across 6 studies (43%); typical prevalence in 2/6 studies (70%); 2/6 studies (13%)

3. Heart failure

NT-proBNP (N-terminal pro hormone BNP)

What is the diagnostic accuracy of pleural fluid tests when diagnosing adult patients with unilateral pleural effusion?

Patient or population: Adults aged 18+ with unilateral pleural effusion

New test: NT-proBNP (N-terminal pro hormone BNP)

Pooled sensitivity: 0.93 (95% CI: 0.88 to 0.96) | **Pooled specificity**: 0.93 (95% CI: 0.86 to 0.97)

Test result	Number of results per 1,000 patients tested (95% CI)	Number of participants (studies)	Certainty of the Evidence (GRADE)
	Prevalence 32%* Typically seen in		
True positives	**298** (282 to 308)	342 (5)	⨁◯◯◯ **VERY LOW** [a,b]
False negatives	**22** (12 to 38)		
True negatives	**634** (585 to 659)	703 (5)	⨁◯◯◯ **VERY LOW** [a,b]
False positives	**46** (21 to 95)		
	Prevalence 20%* Typically seen in		
True positives	**186** (176 to 192)	342 (5)	⨁◯◯◯ **VERY LOW** [a,b]
False negatives	**14** (8 to 24)		
True negatives	**746** (688 to 775)	703 (5)	⨁◯◯◯ **VERY LOW** [a,b]
False positives	**54** (25 to 112)		
	Prevalence 50%* Typically seen in		
True positives	**466** (440 to 481)	342 (5)	⨁◯◯◯ **VERY LOW** [a,b]
False negatives	**34** (19 to 60)		
True negatives	**466** (430 to 485)	703 (5)	⨁◯◯◯ **VERY LOW** [a,b]
False positives	**34** (15 to 70)		

CI: Confidence interval

Explanations

a. Serious risk of bias across studies
b. Some indirectness across studies

* Mean (32%), lowest (20%) and highest (50%) prevalence across 5 studies

5. Autoimmune pleuritis

ANA (antinuclear antibody)

What is the diagnostic accuracy of pleural fluid tests when diagnosing adult patients with unilateral pleural effusion?

Patient or population: Adults aged 18+ with unilateral pleural effusion

New test: ANA (antinuclear antibody)

Pooled sensitivity: 0.94 (95% CI: 0.72 to 0.99) | **Pooled specificity**: 0.87 (95% CI: 0.77 to 0.93)

Test result	Number of results per 1,000 patients tested (95% CI)	Number of participants (studies)	Certainty of the Evidence (GRADE)
	Prevalence 11%* Typically seen in		
True positives	**103** (80 to 109)	42 (4)	⨁⨁◯◯ **LOW** [a,b]
False negatives	**7** (1 to 30)		
True negatives	**772** (684 to 825)	486 (2)	⨁⨁◯◯ **LOW** [a,b]
False positives	**118** (65 to 206)		
	Prevalence 6%* Typically seen in		
True positives	**56** (43 to 59)	42 (4)	⨁⨁◯◯ **LOW** [a,b]
False negatives	**4** (1 to 17)		
True negatives	**815** (722 to 871)	486 (2)	⨁⨁◯◯ **LOW** [a,b]
False positives	**125** (69 to 218)		
	Prevalence 22%* Typically seen in		
True positives	**206** (159 to 218)	42 (4)	⨁⨁◯◯ **LOW** [a,b]
False negatives	**14 (2 to 61)**		
True negatives	**676** (599 to 723)	486 (2)	⨁⨁◯◯ **LOW** [a,b]
False positives	**104** (57 to 181)		

CI: Confidence interval

Explanations

a. Some risk of bias across studies
b. Some indirectness across studies

* Mean (11%), lowest (6%) and highest (22%) prevalence across 4 studies

Recommendation Tables

Question Details

POPULATION:	Adults (18+) with unilateral pleural effusion
INDEX TESTS:	Pleural fluid tests
GOLD STANDARD:	Clinico-pathology
OUTCOME:	Diagnostic accuracy of pleural fluid tests for diagnosing unilateral pleural effusion

1. Secondary pleural malignancy

SUMMARY OF JUDGEMENTS

	JUDGEMENT						
PROBLEM	No	Probably no	**Probably yes**	Yes		Varies	Don't know
TEST ACCURACY	Very inaccurate	Inaccurate	**Accurate**	Very accurate		Varies	Don't know
DESIRABLE EFFECTS	Trivial	Small	**Moderate**	Large		Varies	Don't know
UNDESIRABLE EFFECTS	Large	Moderate	**Small**	Trivial		Varies	Don't know
CERTAINTY OF EVIDENCE	Very low	**Low**	Moderate	High			No included studies
BALANCE OF EFFECTS	Favours the comparison	**Probably favours the comparison**	Does not favour the intervention or the comparison	Probably favours the intervention	Favours the intervention	Varies	Don't know

TYPE OF RECOMMENDATION

Strong recommendation against the intervention	**Conditional recommendation against the intervention**	Conditional recommendation for either the intervention or the comparison	Conditional recommendation for the intervention	Strong recommendation for the intervention
☐	☒	☐	☐	☐

CONCLUSIONS

Recommendation

Pleural fluid cytology should be used as an initial diagnostic test in patients with suspected secondary pleural malignancy, accepting that a negative cytology should lead to consideration of further investigation

Pleural fluid biomarkers should not be used for diagnosing secondary pleural malignancy

Justification

Pleural fluid biomarkers do not provide improved sensitivity, when compared with cytology, for diagnosing secondary pleural malignancy (Low)

Subgroup considerations

All data were considered as subgroups

Research priorities

Further research is needed to determine the clinical utility of pleural fluid cytology, as defined by combined diagnostic and predictive value, in individual tumour sub-types

2. Tuberculous pleural effusion (TPE)

SUMMARY OF JUDGEMENTS

	JUDGEMENT						
PROBLEM	No	Probably no	**Probably yes**	Yes		Varies	Don't know
TEST ACCURACY	Very inaccurate	Inaccurate	**Accurate**	Very accurate		Varies	Don't know
DESIRABLE EFFECTS	Trivial	Small	**Moderate**	Large		Varies	Don't know
UNDESIRABLE EFFECTS	Large	Moderate	**Small**	Trivial		Varies	Don't know
CERTAINTY OF EVIDENCE	**Very low**	Low	Moderate	High			No included studies
BALANCE OF EFFECTS	Favours the comparison	Probably favours the comparison	Does not favour the intervention or the comparison	**Probably favours the intervention**	Favours the intervention	Varies	Don't know

TYPE OF RECOMMENDATION

Strong recommendation against the intervention	Conditional recommendation against the intervention	Conditional recommendation for either the intervention or the comparison	**Conditional recommendation for the intervention**	Strong recommendation for the intervention
☐	☐	☐	☒	☐

CONCLUSIONS

Recommendation

In high prevalence populations, pleural fluid adenosine deaminase (ADA) and/or interferon gamma (IFN-gamma) test(s) can be considered for diagnosing tuberculous pleural effusion

In low prevalence populations, pleural fluid adenosine deaminase (ADA) can be considered as an exclusion test for tuberculous pleural effusion

Justification

Pleural fluid adenosine deaminase (ADA) and interferon gamma (IFN-gamma) provide high sensitivity and specificity for diagnosing tuberculous pleural effusion (**Very Low**)

Subgroup considerations

All data were considered as subgroups

Research priorities

Further research is needed into determining the diagnostic value of pleural fluid markers for tuberculous pleural effusion to improve the strength of the clinical recommendations

5. Autoimmune pleuritis

SUMMARY OF JUDGEMENTS

	JUDGEMENT						
PROBLEM	No	Probably no	**Probably yes**	Yes		Varies	Don't know
TEST ACCURACY	Very inaccurate	Inaccurate	**Accurate**	Very accurate		Varies	Don't know
DESIRABLE EFFECTS	Trivial	Small	**Moderate**	Large		Varies	Don't know
UNDESIRABLE EFFECTS	Large	Moderate	**Small**	Trivial		Varies	Don't know
CERTAINTY OF EVIDENCE	Very low	**Low**	Moderate	High			No included studies
BALANCE OF EFFECTS	Favours the comparison	Probably favours the comparison	Does not favour the intervention or the comparison	**Probably favours the intervention**	Favours the intervention	Varies	Don't know

TYPE OF RECOMMENDATION

Strong recommendation against the intervention	Conditional recommendation against the intervention	Conditional recommendation for either the intervention or the comparison	**Conditional recommendation for the intervention**	Strong recommendation for the intervention
☐	☐	☐	☒	☐

CONCLUSIONS

Recommendation
Pleural fluid antinuclear antibody (ANA) should be considered to support a diagnosis of lupus pleuritis
Justification
Pleural fluid antinuclear antibody (ANA) provides high sensitivity and specificity for diagnosing lupus pleural effusion (**Low**)
Subgroup considerations
All data were considered as subgroups
Research priorities
Further research is needed into determining the diagnostic value of pleural fluid markers for auto-immune pleuritis to improve the strength of the clinical recommendations

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Question Protocol

Field	Content
Review Question	What is the diagnostic accuracy of pleural fluid tests (biomarkers) when diagnosing unilateral pleural effusion in adults?
Type of review question	Diagnostic accuracy
Objective of the review	The use of a range of pleural fluid biomarkers in specific diagnoses
Eligibility criteria – population / disease / condition / issue / domain	Adults with unilateral pleural effusion 18+
Eligibility criteria – index test(s)	Pleural fluid tests (Basic biochemistry (protein / glucose / LDH) Cytology Microbiology Cell count Lipid profile (chylomicrons / cholesterol) NT pro-BNP ADA pH Tumour markers)
Eligibility criteria – gold standard	Clinico-pathology
Outcomes and prioritisation	Diagnostic accuracy
Eligibility criteria – study design	RCTs Prospective comparative studies Case series of >100 patients
Other inclusion /exclusion criteria	Non-English language excluded unless full English translation Conference abstracts, Cochrane reviews, systematic reviews, reviews Cochrane reviews and systematic reviews can be referenced in the text, but **DO NOT** use in a meta-analysis

Proposed sensitivity / subgroup analysis, or meta-regression	Basic biochemistry (protein / glucose / LDH) Cytology Microbiology Cell count Lipid profile (chylomicrons / cholesterol) NT pro-BNP ADA pH Tumour markers
Selection process – duplicate screening / selection / analysis	Agreement should be reached between Guideline members who are working on the question. If no agreement can be reached, a decision should be made by the Guideline co-chairs. If there is still no decision, the matter should be brought to the Guideline group and a decision will be made by consensus
Data management (software)	RevMan5 — Meta-analysis data input. Evidence review/considered judgement. Storing Guideline text, tables, figures, etc. MetaDTA — Data meta-analyses Gradepro — Quality of evidence assessment / Recommendations
Information sources – databases and dates	MEDLINE, Embase, PubMED, Central Register of Controlled Trials and Cochrane Database of Systematic Reviews 1966 - present
Methods for assessing bias at outcome / study level	RevMan5 diagnostic accuracy full review template (based on QUADAS2) (follow instructions in '*BTS Guideline Process Handbook - Diagnostic Accuracy*')
Methods for quantitative analysis – combining studies and exploring (in)consistency	If 3 or more relevant studies: RevMan5 for forest plots, summary ROC plot MetaDTA to combine studies (pooled specificity, sensitivity, likelihood ratios, diagnostic odds ratio and confidence intervals) and calculate RevMan parameters for summary ROC plot (follow instructions in '*BTS Guideline Process Handbook - Diagnostic Accuracy*')
Meta-bias assessment – publication bias, selective reporting bias	GRADEpro — Diagnostic accuracy quality of evidence assessment for each index test (follow instructions in '*BTS Guideline Process Handbook - Diagnostic Accuracy*')

Rationale / context – what is known	Specific tests are known to have diagnostic meaning in pleural disease – pleural pH for infection, cytology positivity varies according to underlying malignancy