TEST

Pleural fluid gram stain

A pleural fluid Gram stain checks fluid around a lung for bacteria visible under a microscope.

WHAT IT MEASURES
Bacteria visible under a microscope after special staining

SAMPLE
Fluid collected from around a lung

FASTING NEEDED?
Usually no; sedation or another procedure may require fasting

HOW LONG IT TAKES
Usually 30 to 60 minutes including fluid collection

RESULTS IN
Often same day; timing varies by laboratory

USED FOR
Diagnosing a possible infection around the lung

MAIN LIMITATION
A negative stain does not rule out pleural infection

Overview

A pleural fluid Gram stain looks for bacteria in fluid from the pleural space. This is the thin space between the lung and the inside of the chest wall.

Normally, the pleural space contains only a small amount of fluid. More fluid may collect because of pneumonia, heart failure, cancer, inflammation, or other conditions. This collection is called a pleural effusion .

The fluid is usually collected through a procedure called thoracentesis , in which a needle or small tube passes through the chest wall. In the laboratory, the sample is stained with special dyes and examined under a microscope.

Bacteria described as Gram-positive usually appear purple. Gram-negative bacteria usually appear pink or red. Their shape and arrangement may offer an early clue, but the stain usually cannot identify the exact species or show which antibiotics like amoxicillin or doxycycline will work.

Why the test is ordered

The test may be ordered when an infection in the pleural space is possible. This can happen alongside pneumonia or after chest surgery, injury, or another infection.

An infected pleural effusion may be called a complicated parapneumonic effusion . A collection of pus in the pleural space is called an empyema .

A Gram stain can give an early result while a pleural fluid culture is still being processed. It is commonly ordered with other tests, including:

- Bacterial culture
- Pleural fluid pH, glucose, and protein
- Cell count
- Lactate dehydrogenase, or LDH
- Tests for tuberculosis or fungi when relevant
- Cytology, which looks for abnormal cells

Who should have the test

This is not a routine screening test. It is used after imaging shows pleural fluid and your healthcare team decides that taking a sample may help explain why it is there.

A Gram stain is particularly relevant when infection is suspected. It may also be performed on fluid removed through an existing chest drain.

Not every pleural effusion needs to be sampled. The decision depends on its size, appearance on imaging, likely cause, your symptoms, and whether the result would change your care.

How to prepare

Fasting is not usually needed for thoracentesis done with local anaesthetic. If sedation or another procedure is planned at the same time, you may receive different instructions. Follow the instructions from the facility performing the procedure.

Tell your healthcare team about:

Do not stop a prescribed blood thinner like warfarin or apixaban unless the team responsible for the procedure tells you to. The risks of continuing or pausing these medicines must be considered for you individually.

An ultrasound is often used before and during fluid collection to find a safe pocket of fluid.

- Blood-thinning or antiplatelet medicines
- Bleeding disorders or previous heavy bleeding
- Medicine or dressing allergies
- Pregnancy or possible pregnancy
- Recent chest procedures or surgery

What happens during the test

You will usually sit upright and lean slightly forward, although another position may be used. The skin is cleaned, and local anaesthetic is injected to numb the area. This can sting briefly.

Using ultrasound for guidance, a clinician passes a needle or small tube between the ribs and into the pleural space. Fluid is drawn into a syringe or drainage container. You may feel pressure or the need to cough.

The needle or tube is removed when enough fluid has been collected, unless a drain needs to remain in place. A dressing is placed over the site.

In the laboratory, a small amount of fluid is spread onto a glass slide, stained, and examined under a microscope. Other portions may be sent for culture and chemical testing.

Understanding your results

A Gram stain is usually reported as organisms seen or no organisms seen . The report may describe bacteria as Gram-positive or Gram-negative and note their shape, such as round bacteria called cocci or rod-shaped bacteria called bacilli.

Seeing bacteria in pleural fluid strongly supports infection, but the stain usually does not provide a final bacterial identification. Culture or molecular testing may be needed to identify the organism and guide antibiotic choice.

A result of "no organisms seen" does not rule out infection. There may be too few bacteria in the sample to see, or treatment may already have reduced their number.

This test has no universal numerical reference range. Reporting and interpretation vary by laboratory and clinical context. Age, sex, pregnancy status, medicines, imaging findings, other pleural fluid results, and your overall health can affect how the result is used, even though most do not directly change how bacteria take up the stain.

What affects the results

Several factors can make the result less representative of what is happening in the pleural space without there being a laboratory error.

Antibiotics started before the sample is collected can reduce the number of visible bacteria. This can also make cultures less likely to grow an organism.

Bacteria may be unevenly distributed, especially if the fluid has formed separate pockets. A sample from one pocket may not reflect another. A small sample or fluid containing very few organisms may also produce a negative stain.

Delays in sending the sample, unsuitable storage, or exposure to air can affect fragile organisms, particularly bacteria that grow without oxygen. Collecting fluid directly into appropriate culture bottles may improve culture yield, but this does not replace a properly prepared Gram-stain sample.

Skin contamination during collection can occasionally introduce bacteria that were not present in the pleural space. Stain particles and cell debris may also resemble bacteria.

Tuberculosis bacteria do not stain reliably with the standard Gram method. They require acid-fast staining, culture, or molecular tests. Some fungi and other unusual organisms also need different stains or tests.

Coffee, exercise, meals, kidney function, pregnancy, age, and a recent blood transfusion do not usually change the Gram-stain result directly. Blood-thinning medicines mainly affect the safety planning for fluid collection rather than the microscopic result.

Accuracy and limitations

A positive result can provide useful early evidence of bacterial infection. However, even when pleural infection is present, the stain may show no organisms. Published results vary because bacterial numbers, previous antibiotics, collection methods, and laboratory processing differ. There is no single miss rate that applies to every setting.

A false-positive result is less common but can occur through contamination, stain debris, or mistaken interpretation. Findings are more convincing when the appearance matches the culture and the other pleural fluid results.

The test cannot reliably identify the exact bacterial species, determine antibiotic susceptibility, or explain non-bacterial causes of an effusion. Culture is better for identifying living bacteria and testing antibiotics, although culture can also be negative. Molecular tests may help in selected cases.

If tuberculosis, cancer, a fungal infection, or another non-bacterial cause is suspected, different tests are needed.

Risks and complications

The Gram stain itself does not add risk once the fluid has been collected. The risks come from thoracentesis.

Temporary discomfort, bruising, coughing, or feeling faint can occur. Less common complications include bleeding, infection, or a pneumothorax , which means air has entered the space around the lung and caused part of it to collapse. Ultrasound guidance helps reduce procedure-related risk.

Seek urgent medical advice after thoracentesis if you develop increasing shortness of breath, worsening chest pain, coughing up blood, fever, or persistent bleeding from the site. Severe difficulty breathing, blue or grey lips, collapse, or confusion requires emergency care.

What happens next

Your healthcare team will interpret the Gram stain alongside your symptoms, examination, scans, culture, and other pleural fluid tests.

A positive stain may support prompt treatment for bacterial infection. Treatment may include antibiotics and, when infected fluid cannot drain adequately on its own, a chest drain or another procedure.

If the stain is negative but infection remains possible, treatment or further testing may still be appropriate. Culture commonly takes longer and may provide more specific information.

For routine follow-up, ask when the culture and other fluid results are expected and how you will receive them. New or worsening breathing problems should not wait for a routine appointment.

Common questions

Can I drink coffee before the test?

Usually, yes. Thoracentesis generally does not require fasting. Do not drink coffee if you were specifically told to fast because sedation or another procedure is planned.

Does the time of day matter?

No. The Gram-stain result is not known to change with the time of day. Scheduling is based mainly on procedure and laboratory availability.

Can I exercise beforehand?

Ordinary activity does not directly alter the stain. If you are short of breath, have chest pain, or feel unwell, avoid strenuous exercise and tell the healthcare team.

Will antibiotics affect the result?

They can reduce the number of bacteria visible on the stain and make culture less likely to grow an organism. Do not delay or stop prescribed antibiotics unless instructed by the clinician managing your care.

Is thoracentesis painful?

The numbing injection may sting, and you may feel pressure during fluid removal. Sharp or severe pain should be reported immediately during the procedure.

Does a negative result mean there is no infection?

No. Gram staining can miss an infection when bacterial numbers are low, antibiotics were started earlier, or the sampled area did not contain many organisms. Other fluid tests, culture, imaging, and your clinical condition remain important.

Sources

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- Merck Manual Professional Edition: How To Do Thoracentesis
- American Family Physician: Pleural Effusion-Diagnostic Approach in Adults, 2023