TEST

Gram stain of tissue biopsy

A Gram stain examines biopsy tissue for bacteria and shows their broad type

WHAT IT MEASURES
Bacteria and their Gram reaction within tissue

SAMPLE
Tissue

FASTING NEEDED?
Sometimes, based on biopsy site and sedation

HOW LONG IT TAKES
Biopsy often 15-60 minutes, depending on method

RESULTS IN
Hours to two days; laboratory turnaround differs

USED FOR
Diagnosing suspected bacterial infection

MAIN LIMITATION
Low bacterial numbers or antibiotics can cause negative results

Overview

A Gram stain of a tissue biopsy is a laboratory test used to look for bacteria in a small piece of tissue. The tissue is collected during a biopsy and examined under a microscope after special dyes are applied.

The test groups many bacteria by how they react to the dyes. Gram-positive bacteria usually appear purple, while Gram-negative bacteria usually appear pink or red. Their shape and arrangement may also provide early clues about the type of bacteria present.

A Gram stain can give information sooner than a culture, which involves trying to grow organisms from the sample. However, it usually cannot identify the exact bacterial species or show which antibiotics like amoxicillin or doxycycline are likely to work.

The laboratory may examine fresh tissue directly in the microbiology department. A pathologist may also use related bacterial stains on preserved tissue sections. These approaches are not interchangeable, so tissue may need to be divided between microbiology and pathology.

Why the test is ordered

This test may be ordered when an infection is suspected in tissue beneath the skin or inside the body. It is often used when a surface swab would not represent what is happening deeper in the tissue.

Examples include suspected infection involving:

The result may help distinguish bacterial infection from other causes of inflammation. It can also provide an early clue while culture, molecular testing or pathology results are still pending.

A Gram stain is not generally used as a screening test in someone without a suspected infection.

- Bone, joints or an implanted medical device
- A deep wound, abscess or surgical site
- An organ or mass seen on imaging
- Tissue that appears inflamed, damaged or unusually swollen

Who should have the test

A Gram stain may be appropriate if you already need a tissue biopsy and your healthcare team thinks infection could explain the findings. The decision is based on your symptoms, examination, imaging, previous procedures and other test results.

The biopsy itself is the main procedure. A Gram stain is one of several tests that may be performed on the tissue. Other portions may be sent for routine pathology, bacterial culture, fungal tests, acid-fast testing for mycobacteria, or molecular tests that look for genetic material.

Not every biopsy needs a Gram stain. It may add little if infection is not suspected or if the available sample has been placed in a preservative that is unsuitable for microbiology culture.

How to prepare

Preparation is determined by the biopsy site and how the tissue will be collected, not by the Gram stain.

You may need to avoid food and drink if sedation or a general anaesthetic is planned. Some biopsies performed using only local anaesthetic do not require fasting. Follow the instructions

from the facility doing the procedure.

Tell the healthcare team about prescription medicines, over-the-counter products and supplements. This is especially important for medicines that affect bleeding or blood clotting. Do not stop a prescribed medicine unless the team responsible for your procedure tells you to.

Also tell them if you are pregnant or could be pregnant, have allergies, have a bleeding disorder, or have previously reacted to anaesthetic medicines or contrast dye.

If possible and medically safe, tissue for microbiology is often collected before antibiotics are started. Never delay or stop antibiotics on your own.

What happens during the test

The biopsy method depends on where the tissue is located. A needle biopsy may be guided by ultrasound, CT or another imaging method. Some tissue is collected during endoscopy or surgery.

You may receive local anaesthetic to numb the area. Sedation or general anaesthesia may be needed for deeper or more complex biopsies. The clinician removes one or more small tissue samples, and a dressing or stitches may be applied afterward.

For microbiology testing, tissue is placed in a sterile container and sent promptly to the laboratory. It is generally not placed in formalin, because formalin kills organisms and prevents routine culture. A separate sample may be preserved in formalin for examination by pathology.

In the laboratory, the tissue is processed and placed on a glass slide. Several dyes are applied in sequence. A trained laboratory professional then examines the slide under a microscope for bacteria, inflammatory cells and other material.

Understanding your results

Gram stain results are usually descriptive rather than numerical. Your report may say that no organisms were seen or may describe Gram-positive or Gram-negative bacteria, along with their shape.

Common descriptions include cocci, meaning round bacteria, and bacilli, meaning rod-shaped bacteria. The report may also describe whether bacteria occur in clusters, chains, pairs or another arrangement. These clues can narrow the possibilities but rarely confirm an exact species.

A positive result means structures consistent with bacteria were seen in the sampled tissue.

Its importance depends on where the tissue came from, whether inflammation was present, and whether culture or other testing supports the finding.

"No organisms seen" does not rule out infection. The sample may contain too few bacteria to be visible, or the affected area may not have been included in the biopsy.

There is no universal normal range for this test. Interpretation varies with the laboratory method, biopsy site, age, sex, pregnancy status, medicines and the wider clinical context.

What affects the results

Several factors can produce a negative or misleading result even when the laboratory performed the stain correctly.

The main influences are:

Contamination can also mislead. Bacteria from skin or the environment may enter the sample during collection, especially when the needle passes through skin. Results are more convincing when bacteria are seen within inflamed tissue and the same organism grows from properly collected cultures.

- Sampling location: Infection can be patchy, so a small biopsy may miss it
- Amount of tissue: Very small samples contain fewer organisms to examine
- Recent antibiotics: Treatment may reduce or damage bacteria before collection
- Specimen handling: Delay, drying or an unsuitable container can reduce usefulness
- Formalin exposure: Preserved tissue may be examined histologically but cannot undergo routine culture
- Low organism numbers: Bacteria must usually be present above a visible level
- Blood, damaged tissue or debris: These can make organisms harder to distinguish
- Type of organism: Some bacteria do not stain reliably with a standard Gram stain

Accuracy and limitations

A Gram stain is fast, but it is less likely than culture or some molecular tests to find small numbers of bacteria. How often it misses an infection differs greatly by tissue type, bacterial amount, previous antibiotic use and collection method. A negative result therefore cannot safely exclude infection by itself.

A positive result can provide useful early evidence, but it is not always a true infection.

Contamination, non-living bacteria after treatment, stain deposits or tissue structures that resemble bacteria can occasionally produce a misleading appearance.

The test cannot usually identify the exact species. It also does not determine antibiotic susceptibility, meaning whether a bacterium is likely to respond to a particular antibiotic.

Other tests may be better in specific situations. Culture can identify living bacteria and test antibiotic susceptibility. Molecular tests may detect organisms after antibiotics or when bacteria are difficult to grow. Acid-fast stains are more suitable for mycobacteria, and fungal stains are better for many fungi. Routine pathology helps show whether organisms are actually located within damaged or inflamed tissue.

Risks and complications

The Gram stain itself does not create additional physical risk because it is performed after the tissue has been removed. Risks come from the biopsy.

Temporary soreness, bruising or minor bleeding can occur. Less common complications depend on the biopsy site and may include infection, heavier bleeding, injury to a nearby structure or a reaction to sedation. A lung biopsy can sometimes allow air to collect around the lung.

Follow the specific aftercare instructions you receive. Mild soreness or a small bruise can usually be discussed at routine follow-up.

Seek urgent medical advice for increasing redness, swelling, drainage, fever, worsening pain or bleeding that does not settle. Call emergency services for severe breathing difficulty, collapse, chest pain after a chest or lung biopsy, or uncontrolled bleeding.

What happens next

A preliminary Gram stain result may be available before the culture or full pathology report. Your healthcare team will interpret it alongside your symptoms, imaging and other laboratory findings.

If bacteria are seen, treatment may be started or adjusted while identification and antibiotic susceptibility results are pending. A negative stain may still be followed by culture, molecular testing or special stains if infection remains a concern.

Cultures can take several days, and some slow-growing organisms take longer. Routine follow-up is often used to review the combined results and discuss whether any further testing

or treatment is needed.

Common questions

Can I drink coffee before the test?

The stain does not require fasting. However, you may need to avoid coffee, food and other drinks before the biopsy if sedation or anaesthesia is planned. Follow the instructions provided for your procedure.

Does the time of day matter?

Usually not. The timing is based on the biopsy appointment, specimen transport and laboratory availability rather than a daily change in bacteria.

Can I exercise beforehand?

Exercise does not directly alter a Gram stain. Your biopsy team may ask you to avoid strenuous activity before or after the procedure, particularly if the biopsy involves a muscle, bone or internal organ.

Should I stop antibiotics before the biopsy?

Do not stop them unless the healthcare team managing the procedure tells you to. Antibiotics can reduce the chance of seeing or growing bacteria, but delaying treatment may not be safe.

How quickly will I get the result?

A Gram stain may be reported within hours or by the next day, although laboratory schedules differ. Culture and pathology results usually take longer.

Will it show which antibiotic I need?

Not by itself. The appearance may offer an early clue, but culture and antibiotic susceptibility testing are usually needed to guide a more specific choice.

Sources

- World Health Organization. Basic Laboratory Procedures in Clinical Bacteriology . 2nd edition. WHO; 2003.
- Leber AL, editor. Clinical Microbiology Procedures Handbook . 4th edition. ASM Press; 2016.

- Miller JM, Binnicker MJ, Campbell S, and colleagues. Guide to Utilization of the Microbiology Laboratory for Diagnosis of Infectious Diseases: 2024 Update by the Infectious Diseases Society of America and the American Society for Microbiology . Clinical Infectious Diseases; 2024.
- Suvarna SK, Layton C, Bancroft JD, editors. Bancroft's Theory and Practice of Histological Techniques . 8th edition. Elsevier; 2019.
- U.S. National Library of Medicine. Gram Stain . MedlinePlus Medical Test.
- National Health Service. Biopsy . NHS health information.