

Sputum Analysis



Sputum:
An Indicator
of Lung Disease



By

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What is sputum?

- Mucus, or sputum, is different than saliva and is composed of **dead cells** and **debris from the lower respiratory tract**.
- **So it is function:** It traps debris and organisms such as bacteria so that they can be cleared, or coughed up, from the lungs.

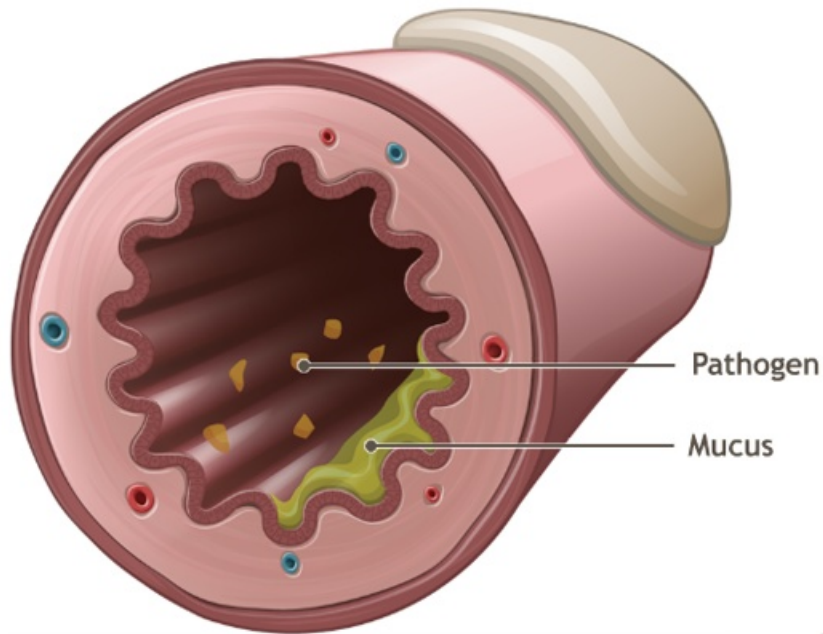
Sputum is different than saliva (spit):

- Sputum comes from deep inside your lungs and is usually thick. The respiratory tract produce about two liters of mucus a day from these glands.
- Saliva (spit) comes from your mouth and is thin and watery.

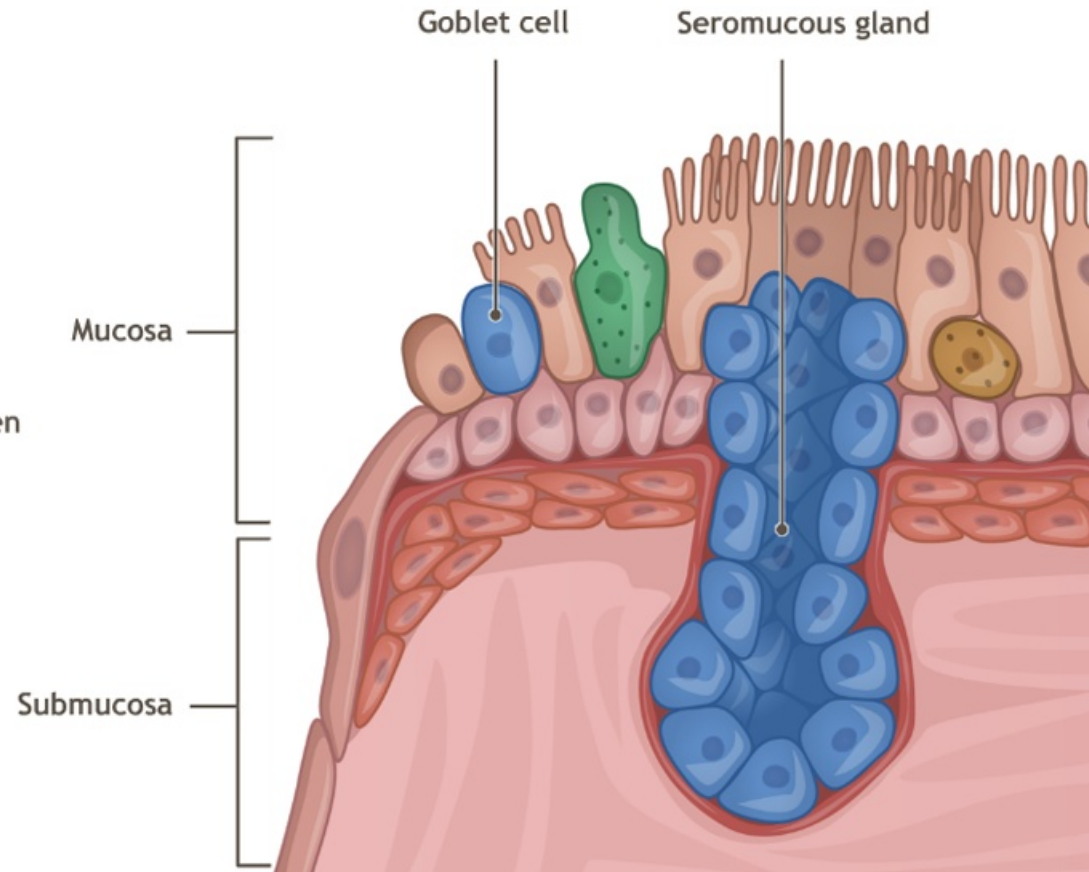
To get an accurate test result of TB, be sure to collect sputum, not saliva.

- **Mucus** is secreted from two distinct areas within the lung tissue:
 1. **In the surface epithelium**, which is part of the tissue lining of the airways, there are mucus-producing cells called **goblet cells**.
 2. **The connective tissue layer** beneath the mucosal (submucosa) epithelium contains **seromucous glands** which also produce mucus.

Airway mucus is a viscous gel that coats the inner surface of the airways and serves to trap foreign debris and **pathogens**, allowing them to be removed from the airway.



The mucus is produced and secreted by specialized cells known as goblet cells in the epithelium and glands in the submucosa (ie, the layer beneath the mucosa).



Select the Slideshow icon for information about the steps of mucus secretion.

- Mucus is composed of **water, carbohydrates, proteins and lipids**. Mucus contains glycoproteins (or mucins) as well as proteins derived from plasma, and products of cell death such as DNA.
- Mucus is **sticky** and this helps to trap dust particles, bacteria and other inhaled debris.
- Mucus also contains **natural antibiotics**, which help to destroy bacteria.
- Mucus also contains lysozyme, which is an antibacterial enzyme.

Why do we collect sputum?

- The sputum is examined grossly (macroscopically) and microscopically to aid medical diagnosis.
- The sputum analysis involves an analytical approach to investigate the cellular and non-cellular components expelled from the patient's upper respiratory tract. This procedure is essential in the evaluation and management of lower respiratory infections or other longstanding health conditions.
- Sputum cytology helps detect both lung cancer cells and non-cancer cellular and acellular material useful for the diagnosis of conditions such as pneumonia, tuberculosis, interstitial lung diseases, or pneumoconiosis (e.g., asbestosis).

Indications of testing sputum

1. Clinical diagnostic sputum tests aim to detect the causes of lower respiratory tract infections. Bacteria or fungi causing the airways or lung infection.
2. Sputum culture is the most common test needed to be performed when the patient has pneumonia.
3. Sputum can be collected to test it for tuberculosis (Tuberculosis) germs. *Mycobacterium tuberculosis*.
4. It also provides an efficacious tool for monitoring the effectiveness of clinical treatment.

Causes of Pneumonia

- Viruses, bacteria, and fungi can all cause pneumonia.

1. Viruses: Influenza viruses, SARS-CoV-2 (the virus that causes COVID-19)

2. Bacteria: Common causes of bacterial pneumonia are *Streptococcus pneumoniae* (pneumococcus) and, especially in kids, *Mycoplasma pneumoniae*.

Defining Types of Pneumonia

1. Community-acquired pneumonia: is when someone develops pneumonia in the community (not in a hospital).

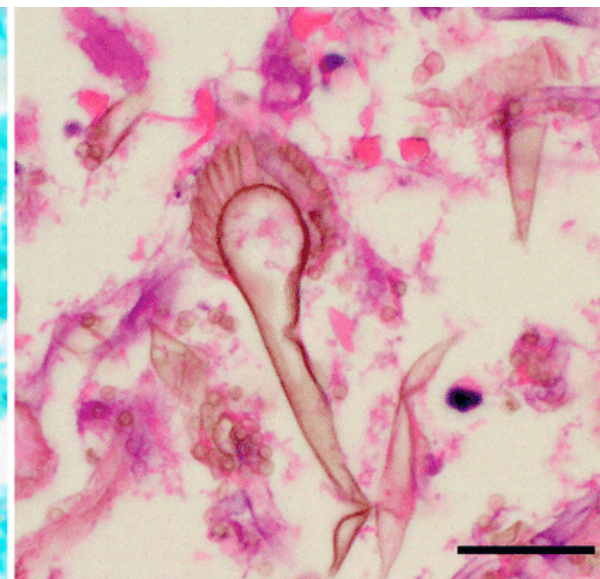
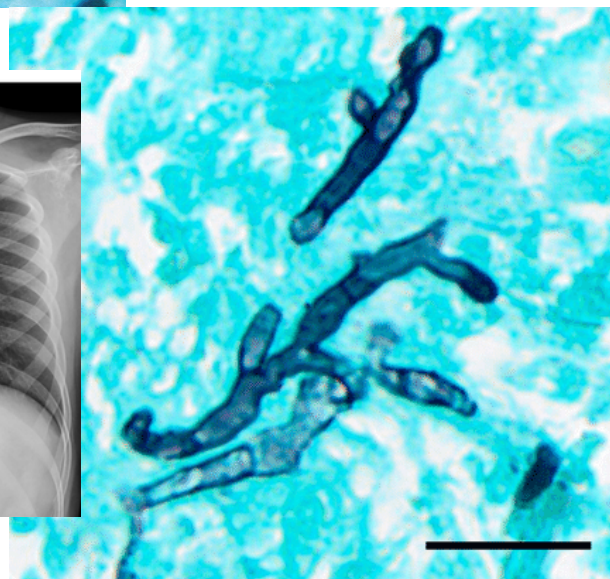
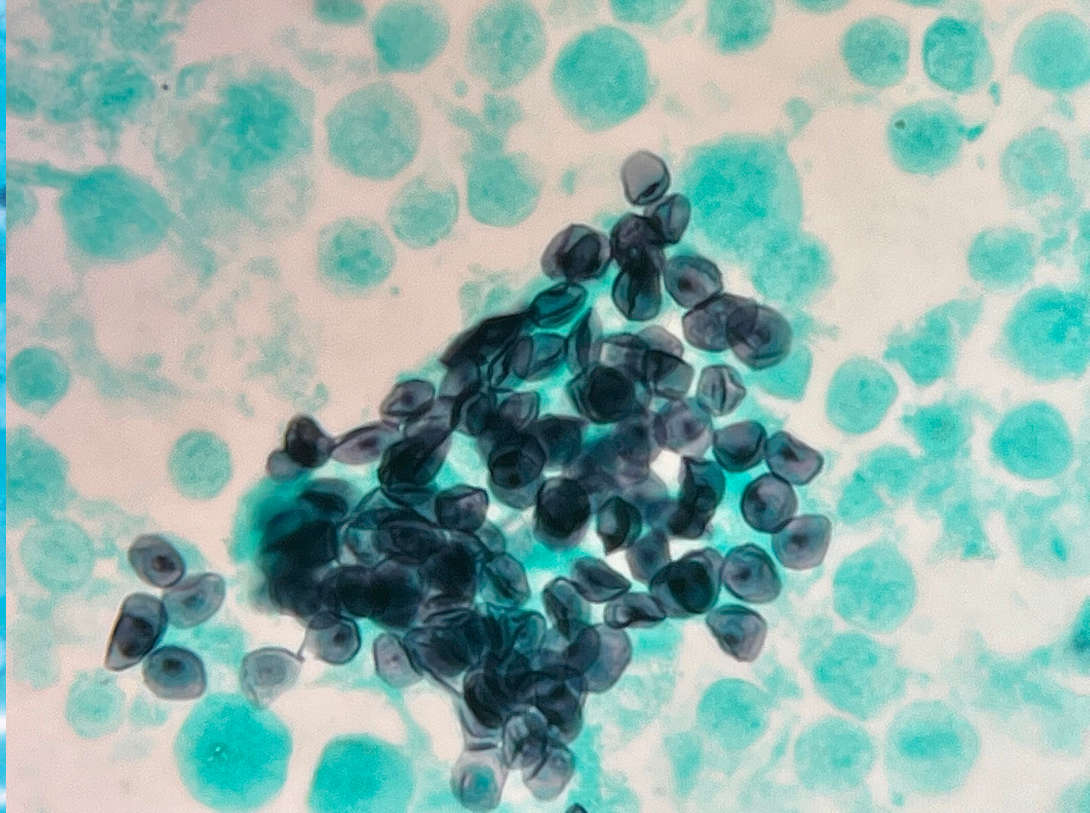
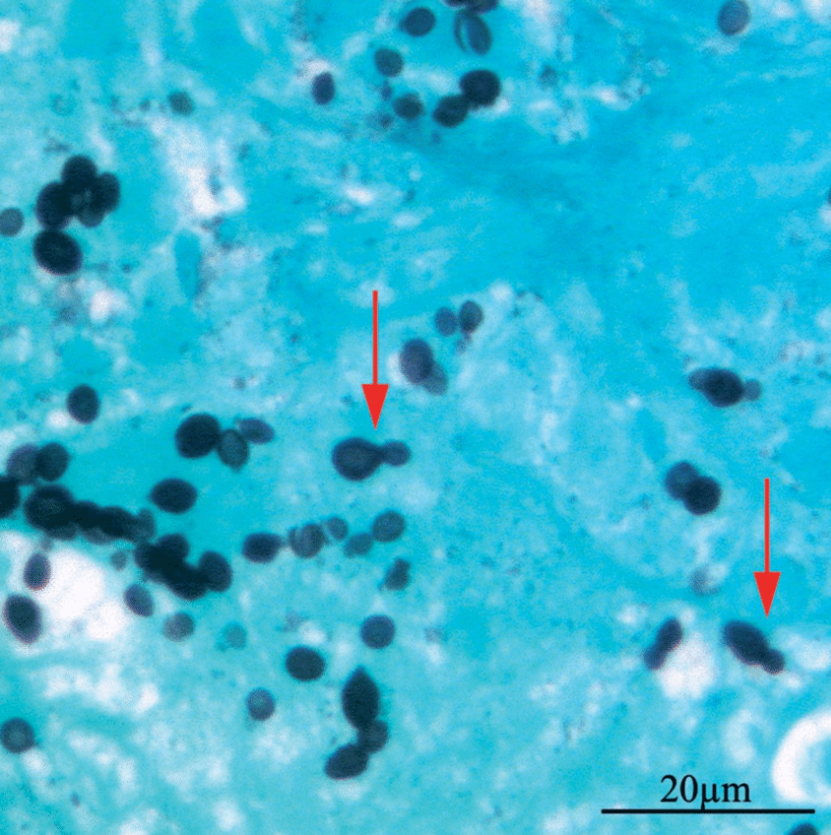
2. Healthcare-associated pneumonia is when someone develops pneumonia during or following a stay in a healthcare setting. Healthcare settings include **hospitals, long-term care facilities, and dialysis centers.**

3. Ventilator-associated pneumonia is when someone gets pneumonia after being on a ventilator.

Diagnostic Tests for sputum:

A. Sputum smear microscopy is the initial step taken in laboratory sputum analysis.

- 1. Gram staining** is used to differentiate bacteria into two broad groups (gram-positive and gram-negative bacteria).
- 2.** When the physician suspects that the patient may have TB, acid-fast bacilli (AFB). **Tuberculosis (TB)** is a lung infection disease caused by *Mycobacterium tuberculosis*. Mycobacteria are a group of rod-shaped acid-fast bacilli. They can be distinguished under the microscope after an AFB staining procedure where the bacilli retain the red stain (Carbol fuchsin stain) color after an acid-fast wash.
- 3. The Grocott-Gomori's Methenamine Silver stain (GMS)** is a standard staining method (a popular staining method in histology) used to detect fungal microorganisms. GMS staining is critical in identifying *Pneumocystis jirovecii*. (initially called *Pneumocystis carinii*)



Mycoplasma pneumoniae

B. Real time PCR:

Respiratory viruses have been tested in sputum specimens from patients with **cystic fibrosis**, **asthma**, and **chronic obstructive pulmonary disease (COPD)**. Viral pathogens such as:

1. Severe acute respiratory syndrome (SARS) coronavirus,
2. H1N1 influenza
3. Middle Eastern respiratory syndrome coronavirus (MERS-CoV).
4. SARS coronavirus 2 (SARS-CoV-2), the causative agent of Coronavirus Virus disease 2019 (COVID-19), may be absent in upper airway secretions.

The sputum samples are frequently used for viral diagnosis using the **real-time polymerase chain reaction (RT-PCR)** method or the newly developed **next-generation sequencing (NGS)** method.