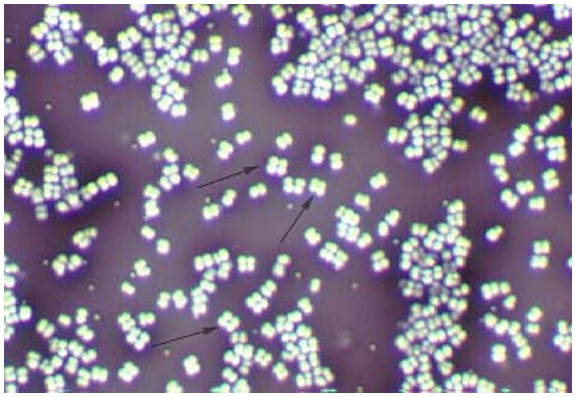
An Introduction to Biological Staining

This brief introduction is not to explain how to do a particular stain. Rather, it is to illustrate what various stains look like once they have been accomplished successfully. You will have the opportunity to examine actual slides of these stains under the light microscope during your lab time.

Background Stain

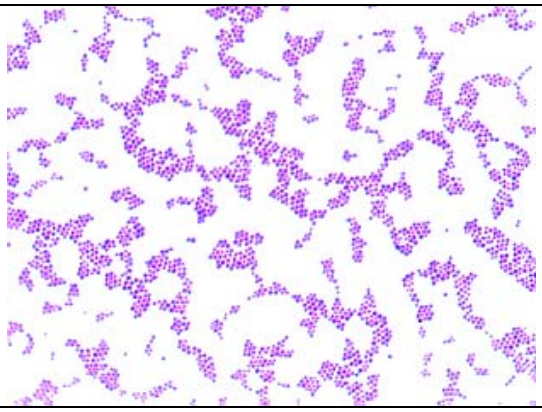
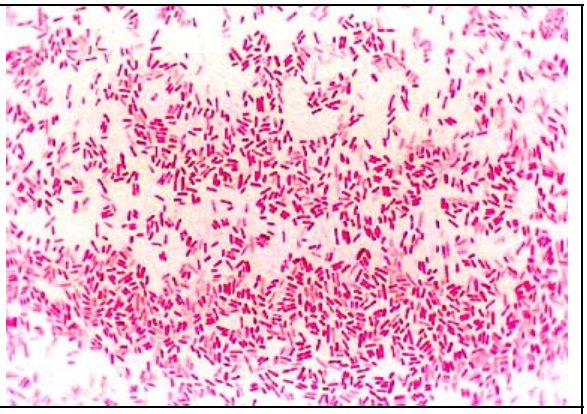
This stain is so called because it is the background that is stained. Either India ink or Black Nigrosin is used to stain the background black and the bacteria are opaque:



This is a background stain of cocci – spherical bacteria. Some are in pairs, some are in tetrads (arrows) and, periodically, there is a single coccus. The background is black – the bacteria appear opaque.

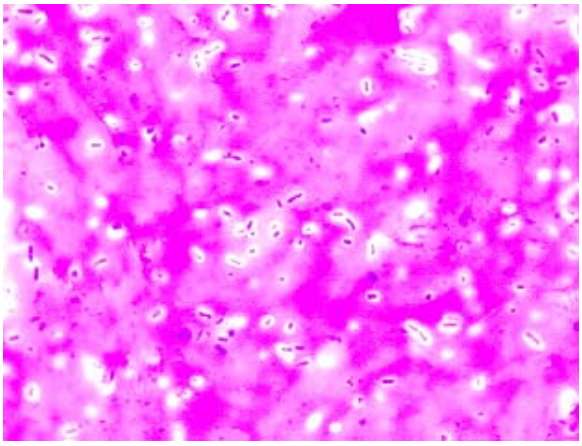
Gram Stain

This is one of the most commonly used stains in clinical practice. Bacteria which appear purple by this method – and this method ONLY – are called Gram positive; bacteria which appear pinkish orange by the method – and this method ONLY – are called Gram negative. Below are two graphics of each, respectively:

	
Gram positive cocci in clusters.	Gram negative rods.

Capsule Stain

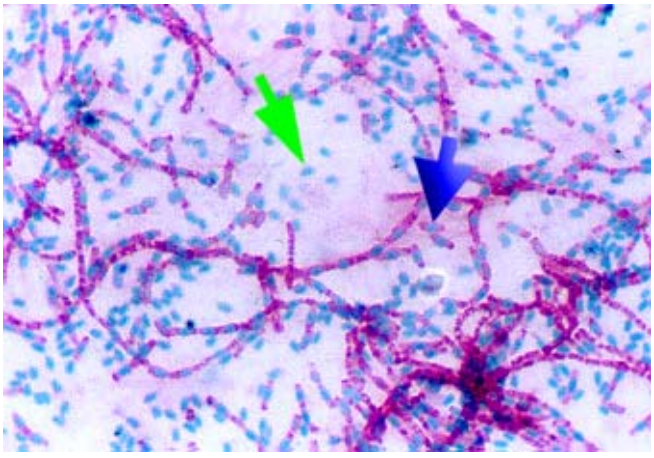
The capsule stain is used to help determine what general type of bacterium a patient might be infected with, e.g., *Streptococcus pneumoniae* or *Klebsiella pneumoniae*. In this stain, the bacterium is stained purple, the background is stained fuchsia and the clear space between the two is the capsule:



In this case, these are encapsulated rods – those that appear to be cocci, are actually rods "on end".

Spore Stain

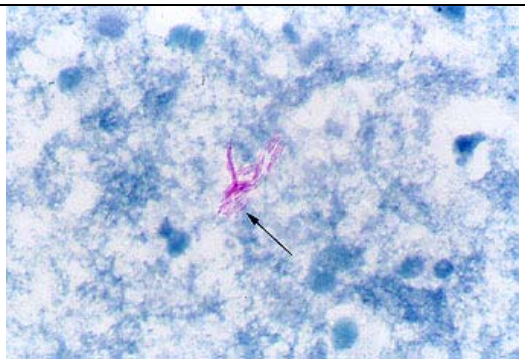
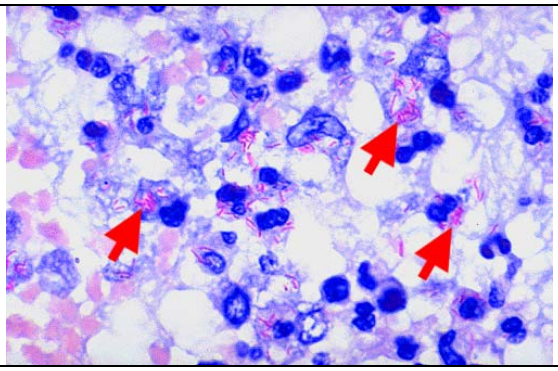
The spore stain stains free spores (spores free of a "cell body") and endospores (spores contained within a "cell body") green. The remaining "cell body" is called a sporangium and stains fuchsia:



The blue arrow is pointing to an endospore; the lime green arrow is pointing to a free spore. The fuchsia rods are the sporangia (if they contain endospores) or rods (bacilli) if they do not contain endospores.

Acid Fast Stain

This stain is commonly used to determine if someone has a tuberculosis-like disease. The acid fast bacteria (AFB) stain fuchsia – non-AFB stain blue. Below are two examples of the acid fast stain:

	
AFB – source suspected to be sputum.	AFB – arrows pointing out example clusters – source suspected to be from bronchial washings.

Silver Stain for Treponemes

The last stain is that used for treponemes when dark field microscopy is not available. The most significant treponeme for our purposes is *Treponema pallidum*, the causative agent of syphilis. Treponemes are spirochaetes. This means that they are spiral and because of their unique morphology, actually "screw themselves into" the tissues they infect, much like a corkscrew into a bottle cork. By this stain, the treponemes are blue and the remaining cells and debris are yellowish-brown to brown: