

SIMPLE POSITIVE STAIN WITH THREE EASY TO FIND DYES - GENTIAN VIOLET, METHYLENE BLUE, DENTAL PLAQUE REVEALER (ERYTHROSINE).

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INTRODUCTION:

Whether we like it or not, brightfield illumination is the main technique for optical microscopy, since it is the simplest way of illuminating a sample. It just requires us, even without a condenser, to turn on the light and the majority of microscopes intended for students, enthusiasts or professionals such as vets or physicians come with this kind of illumination system. If we want something more special, well there are other illumination techniques.

This article is related to brightfield microscopy because the samples are observed with this illumination. If the samples are less than a micron in size such as bacteria, they are difficult to see without enhancing the sample contrast relative to the object(s) surroundings. We will consider simple staining with dyes that can be very easy to find everywhere and which we can use to stain our samples.

DEVELOPMENT:

There are two terms to be defined from the title. I mean **simple** in the sense that we are just using one stain, and **positive** because the objects in the sample are going to acquire the color of the ink employed, in this case GENTIAN VIOLET, METHYLENE BLUE and DENTAL PLAQUE REVEALER.

There is obviously the procedure called negative staining where the object in the sample does not acquire the color of the ink and just the background is enhanced with the stain, but the explanation of this is for the moment out of the scope of this article.

Gentian violet is an antiseptic used to treat local mycotic infections. Since it has certain activity against bacteria, it is used to prevent infection of minor wounds and as its name indicates it produces a kind of purple when applied to any surface. It can be purchased over the counter and it is very cheap and found in any drug store.

The second one is methylene blue, a dye very common in pet shops it is used regularly as an antiseptic for fish when they are sick from parasitic infections and/or mycotic ones.

Finally the dental plaque revealer which can also be purchased over the counter. The purpose of this is to reveal bacterial dental plaque. It stains teeth positively revealing where this plaque is, I found the active substance is called erythrosine.

Today these stains are going to be used with microscopy samples to enhance contrast.



The common procedure for simple staining is as follows.

- ✓ First spread the sample upon a slide, then it is fixed it with heat from a flame,



Note: Since I do not have a burner, for the moment I use a candle - obviously it is necessary to clean the soot from below the slide.

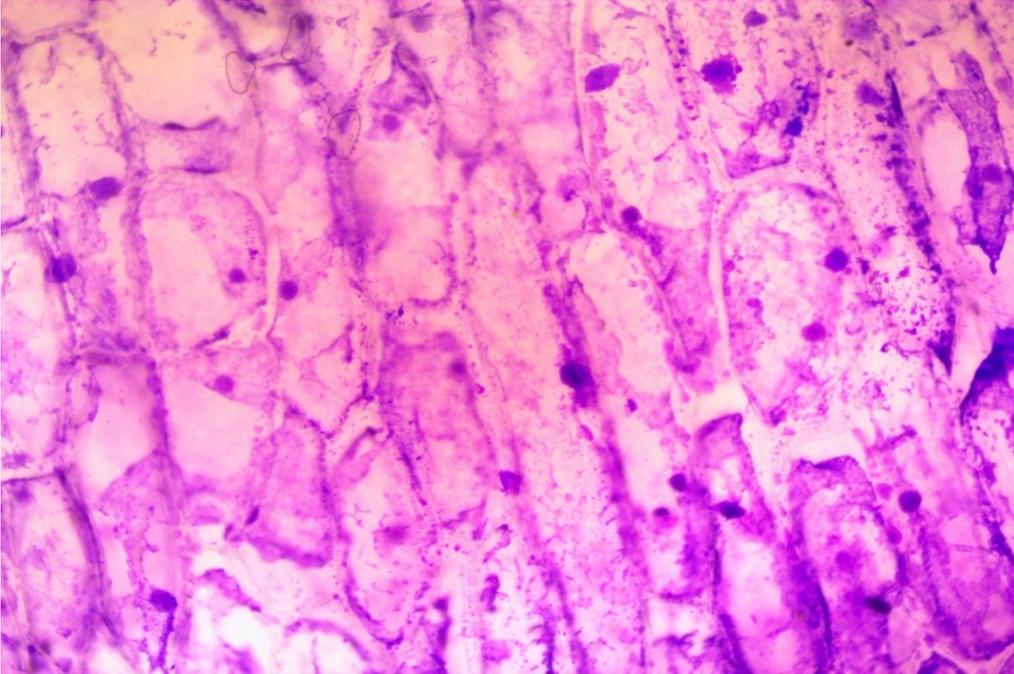
- ✓ Then the dye is applied, just enough drops to cover the whole sample, leave for a minute to allow it to penetrate the cells.



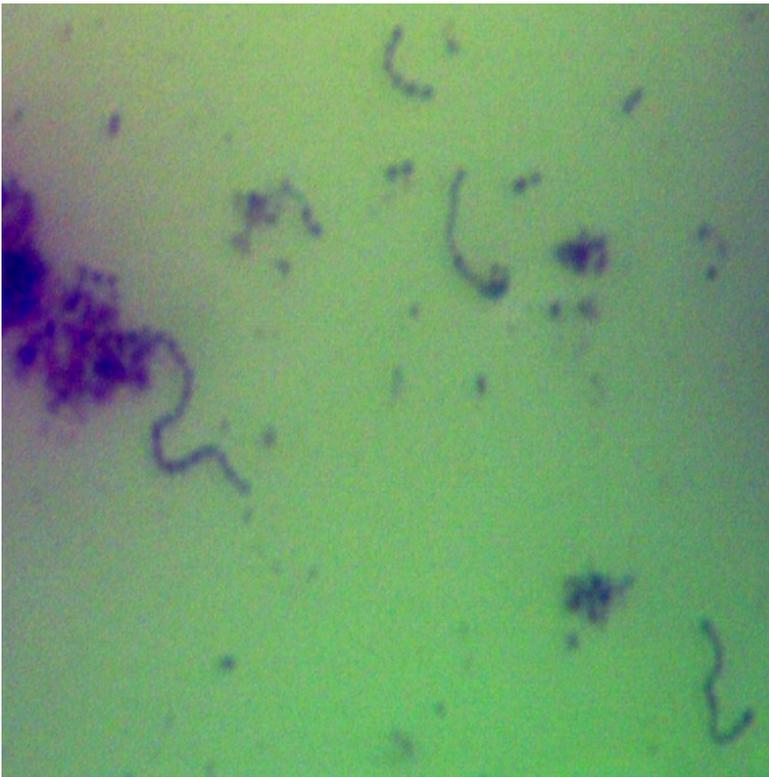
- ✓ After that pour off the remainder of the dye and wash it with water or alcohol.
- ✓ Finally leave the sample to dry to the air and it is ready to be observed.



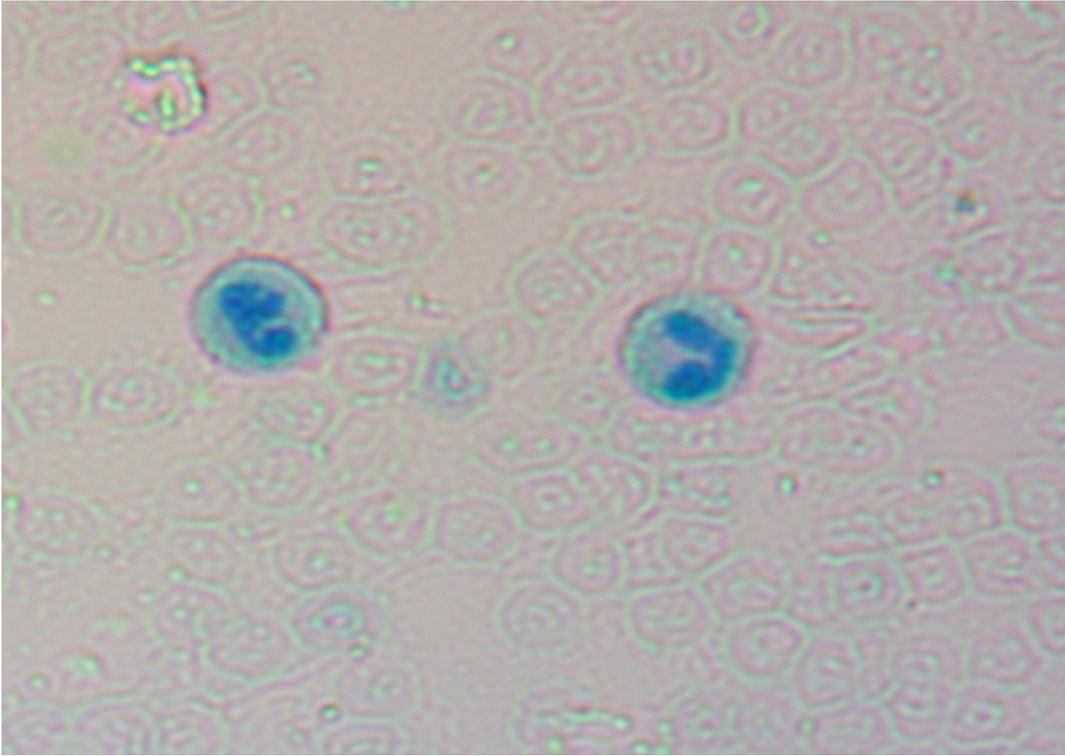
Results:



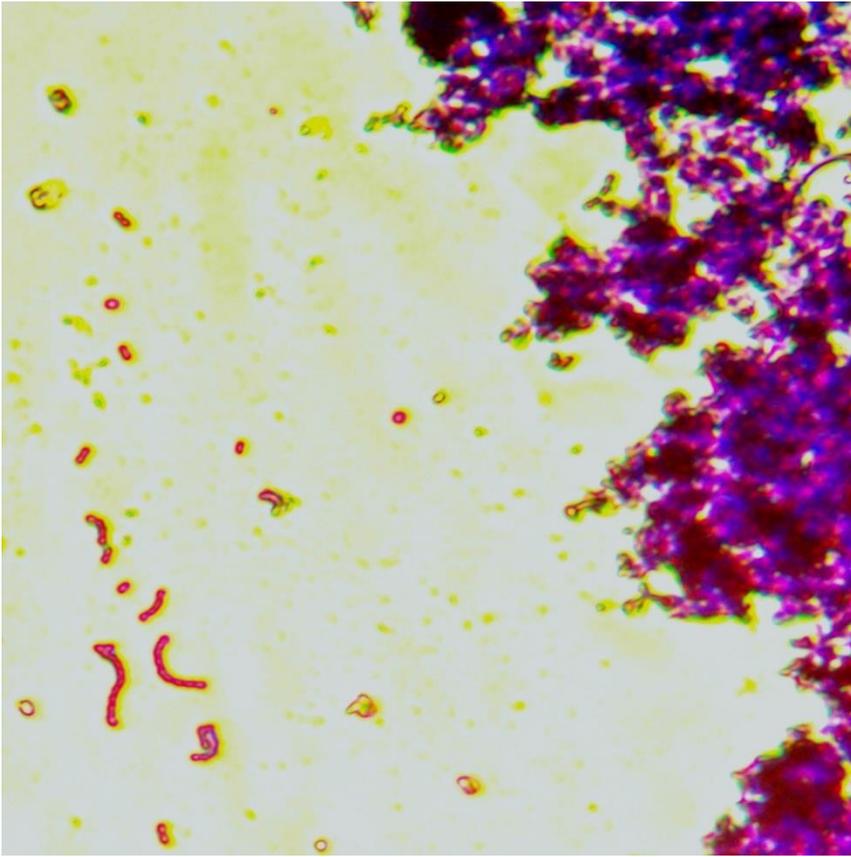
Onion gentian violet 10x



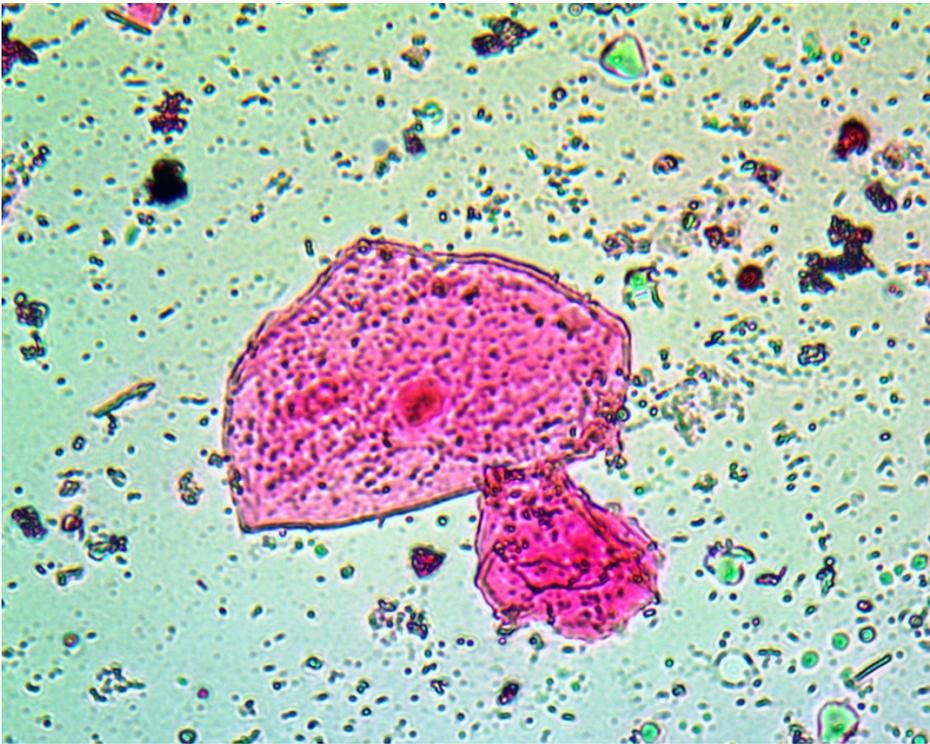
Streptococcus 40x found in a drop of pond water.



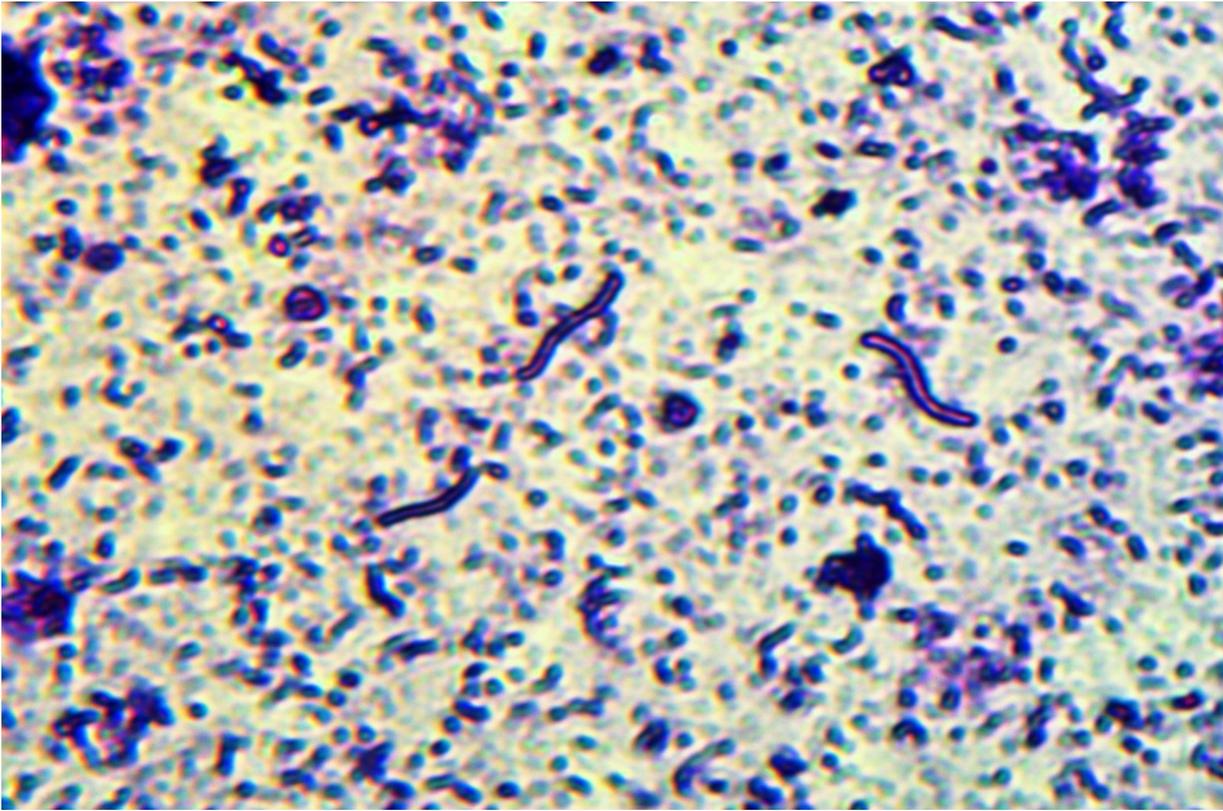
White blood cells methylene blue 40x



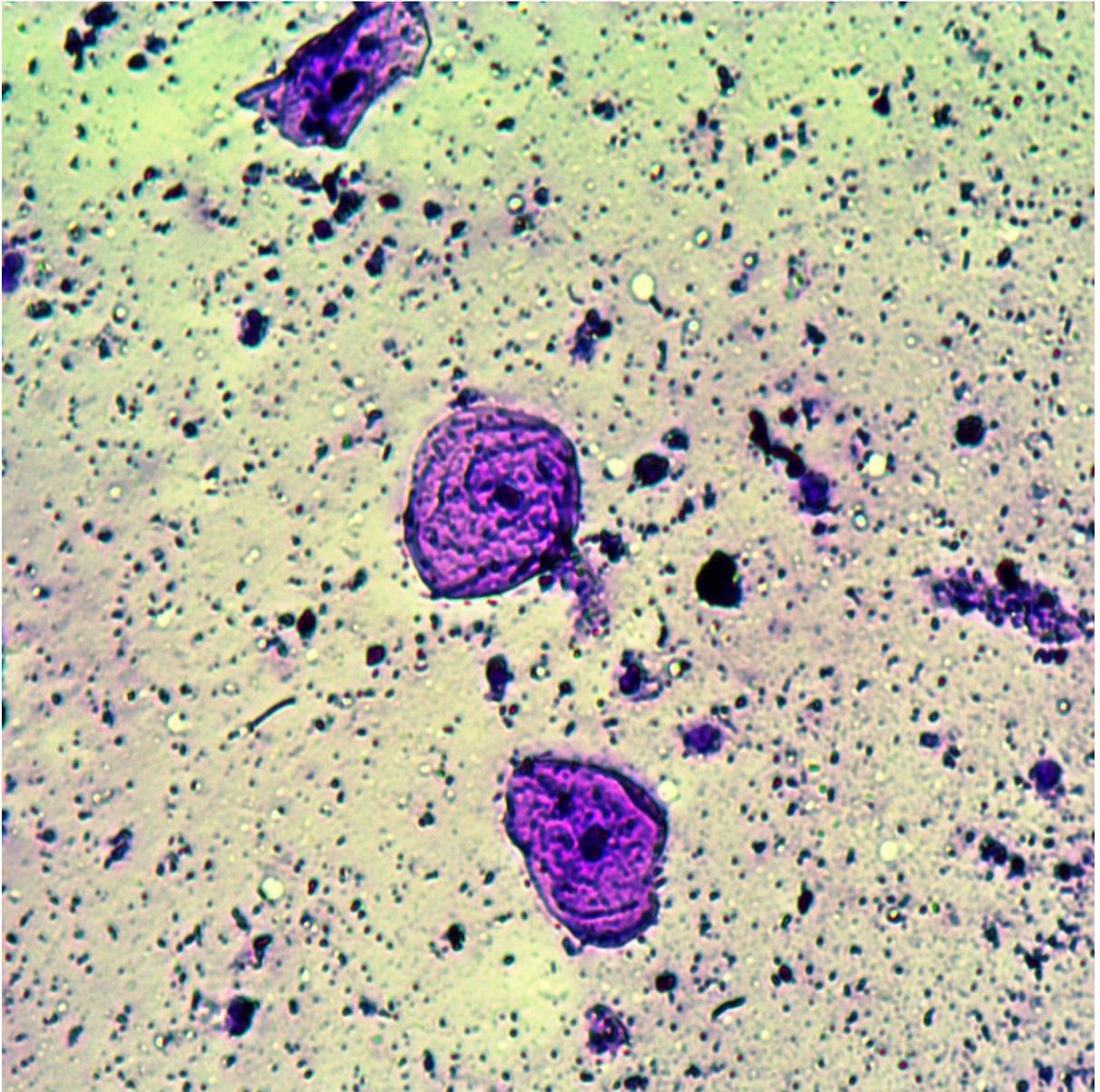
Streptococcus of yogurt 40x gentian violet



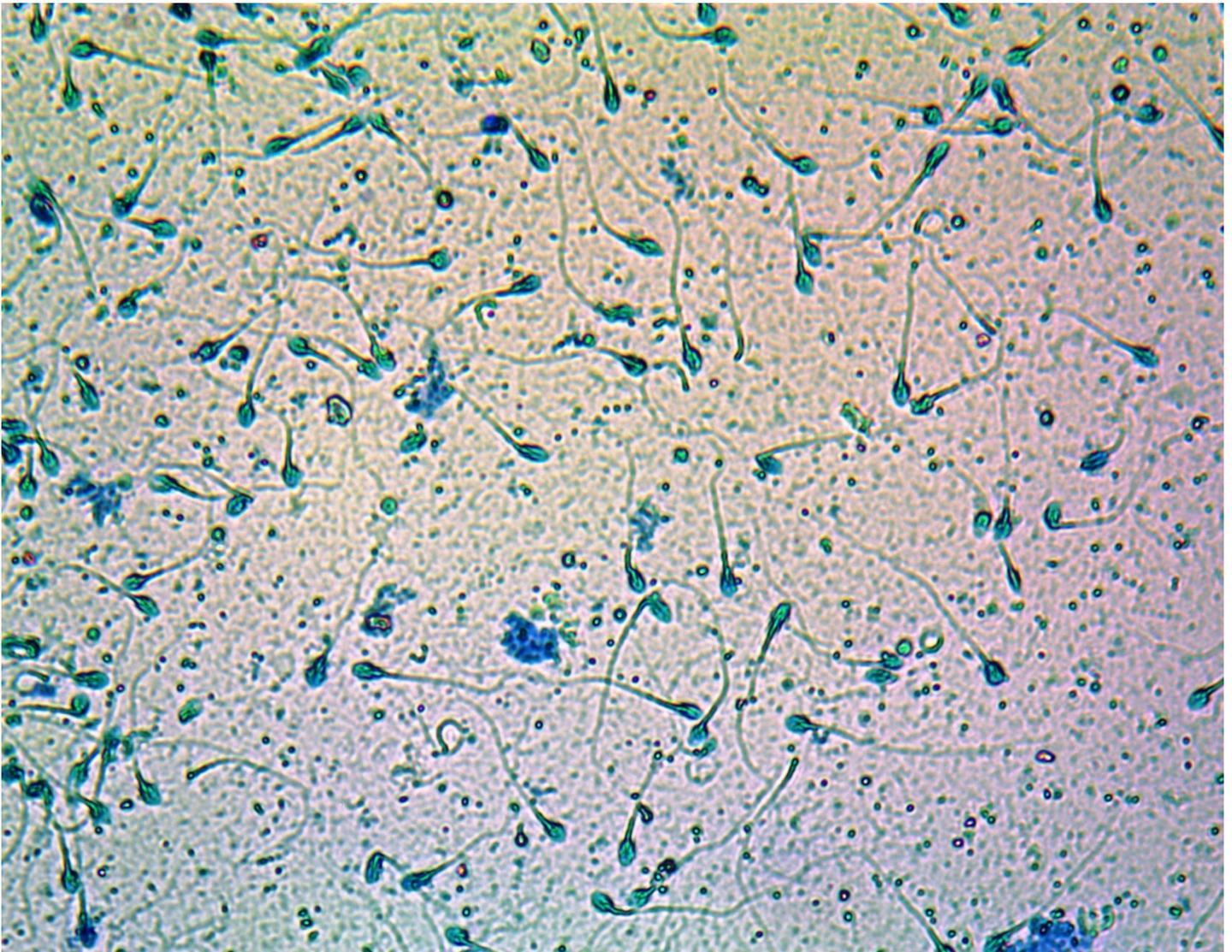
Mouth epithelial cells stained with dental plaque revealer 40x



Spirillum 40x gentian violet



Mouth epithelial cells and mouth bacteria 10x



Sperm with methylene blue 40x

CONCLUSION:

What to stain with simple positive staining? Whatever is tried, not all are going to get the stained look as seen for the bacteria above in the sample with the dental plaque revealer of mouth epithelial cells. Samples are poorly stained if they look as if they were stained in green, obviously this is a negative staining for these bacteria. But on a trial and error basis we can get beautiful results and is a lot of fun. No matter if we do not possess all the instruments of a lab - for example I lack a burner, nevertheless that is not an obstacle for staining, I use a candle.

I can say that I LOVE TO STAIN.

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(Above in anti-spam format. Copy string to email software, remove spaces and manually insert the capitalised characters.)

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