

Jun 10, 2024

Staining Spores for Conidia Counting

DOI

<https://dx.doi.org/10.17504/protocols.io.6qpvr8462lmk/v1>

IAC¹, Sara de Oliveira Olimpio¹, Rebecca Ogioni¹

¹Instituto Federal do Espírito Santos - Campus Alegre



Sara de Oliveira Olimpio

Instituto Federal do Espírito Santo - Campus Alegre

Create & collaborate more with a free account

Edit and publish protocols, collaborate in communities, share insights through comments, and track progress with run records.

Create free account

OPEN  ACCESS



DOI: <https://dx.doi.org/10.17504/protocols.io.6qpvr8462lmk/v1>

Protocol Citation: IAC , Sara de Oliveira Olimpio, Rebecca Ogioni 2024. Staining Spores for Conidia Counting. **protocols.io**
<https://dx.doi.org/10.17504/protocols.io.6qpvr8462lmk/v1>

License: This is an open access protocol distributed under the terms of the **Creative Commons Attribution License**, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited

Protocol status: Working

We use this protocol and it's working

Created: May 14, 2024



Last Modified: June 10, 2024

Protocol Integer ID: 99758

Keywords: spores for conidia counting methylene blue, conidia counting methylene blue, conidia with the specific dye, staining spore, morphological identification of the microorganism, identification of microorganism, microorganism, conidia, specific dye, dye, morphological identification, bacteria, visualization of dead tissue

Abstract

Methylene blue has different functions depending on its concentration. Its use allows the visualization of dead tissues and bacteria, for example. As a solution, this dye allows the identification of microorganisms in general. For practical use in this matter, the morphological identification of the microorganism is obtained from the visualization of its conidia with the specific dye.



Procedure

- 1 Add sample in 1,5 mL tube;
- 2 Add 100 uL of methylene blue dye;
- 3 Homogenize by vortexing for 10 seconds and wait 2 minutes;
- 4 Centrifuge for 1 minute at 16.000g;
- 5 Pipette 50 uL into the Neubauer chamber or microscope slide and cover with a cover slip;
- 6 Evaluate under an optical microscope;
- 7 Store at 4°C overnight before to use.

Materials

- 8 Tween® 80;
- 9 Methylene blue;
- 10 Distilled water.

Solution Preparation (100 mL)

- 11 In a becker add 80 mL of distilled water;



- 12 Weigh 0.7g of methylene blue;
- 13 Pipette out 20 uL of Tween® 80;
- 14 Fill volume to 100 mL with distilled water.

References

- 15 Pham, Tuan Anh et al. "Production of Blastospore of Entomopathogenic Beauveria bassiana in a Submerged Batch Culture." *Mycobiology* vol. 37,3 (2009): 218-24. doi:10.4489/MYCO.2009.37.3.218
- 16 Oktari, A., et al. "The bacterial endospore stain on Schaeffer Fulton using variation of methylene blue solution." *Journal of Physics: Conference Series*. Vol. 812. No. 1. IOP Publishing, 2017.