

# Product Name : Ziehl-Neelsen Color Kit

Complete kit for coloring acid-alcohol resistant bacteria.

## CONTENT OF THE PACKAGES

Fuchsin Solution .....	250 mL
Acid-Alcohol Solution .....	250 mL
Methylene Blue Solution .....	250 mL
1 Instruction sheet	

## DESCRIPTION

Ziehl-Neelsen Color Kit is a kit for coloring acid-alcohol resistant bacteria present in pathological samples or in culture media. This coloring is particularly useful is performing a microscope examination for the presence of mycobacteria.

## PRINCIPLE OF THE METHOD

Ziehl-Neelsen coloring is based on the fact that phenol allows basic Fuchsin to penetrate the interior of the bacterial cell. After they have been stained with basic Fuchsin, some bacteria remain red if treated with the Acid-Alcohol Solution (acid-alcohol resistant bacteria). Others lose the red color and take up methylene blue, becoming blue in color (bacteria sensitive to decoloring with acid-alcohol).

## USE

### 1. Preparation and fixing

- On clean slides make a smear of the pathological material or of the culture. Leave to dry in the air and fix by passing rapidly over the flame, or by using other fixing methods. Do not overheat the sample when fixing.

### 2. Coloring

In all phases of the coloring, the preparation must be rinsed with water free of acid-alcohol resistant species.

- Cover the slide with the Fuchsin Solution. Heat the slide slowly until steam forms. Continue heating the slide for 3-5 minutes. Leave to cool and then rinse with water.
- Decolor with the Acid-Alcohol Solution for as long as the preparation releases color (about 1-2 minutes). Then rinse with water.
- Cover the slide with Methylene Blue Solution. Leave in contact for 30-60 seconds. Then rinse with water and leave to dry.
- Examine the preparation under the microscope with the objective for immersion.

## INTERPRETATION OF THE RESULTS

The mycobacteria and acid-alcohol resistant bacteria appear colored red, whereas the background material and other non acid-alcohol resistant material are colored blue.

## QUALITY CONTROL

Each batch of Ziehl-Neelsen Color Kit is submitted to the quality control using the following microorganisms :

Microorganism		Color	
<i>Mycobacterium smegmatis</i>	ATCC 14468	Red	(acid-alcohol resistant)
<i>Staphylococcus aureus</i>	ATCC 25923	Blue	(non acid-alcohol resistant)

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## LIMITS

- Ziehl-Neelsen coloring provides presumptive evidence of the presence of acid-alcohol resistant bacteria in the sample, evidence that must be confirmed using other methods. A negative result for the coloring does not necessarily prove that the sample is negative for the presence of acid-alcohol resistant bacteria.
- Fast growing mycobacteria may retain the coloring to different degrees.
- When decoloring with the Acid-Alcohol Solution, take care to avoid insufficient decoloration.

## PRECAUTIONS

The Ziehl-Neelsen Color Kit contains substances classified as hazardous by current legislation. It is recommended that the Safety Data Sheets be consulted on their use. Ziehl-Neelsen Color Kit is a kit for bacteria coloring, only for diagnostic use *in vitro*. It is intended for use in a professional environment and must be used in a laboratory by adequately trained personnel using approved asepsis and safety methods for dealing with pathogenic agents.

## STORAGE

Store the Ziehl-Neelsen Color Kit at room temperature in its original package. Keep away from sources of heat and avoid excessive changes of temperature. In such conditions the Ziehl-Neelsen Color Kit will remain valid until the expiry date indicated on the label. Do not use beyond that date. Eliminate without using if there are signs of deterioration (change in the color of the solutions or presence of substantial precipitates).

## DISPOSAL OF USED MATERIAL

After use, the slides colored with the Ziehl-Neelsen Color Kit and any material that has come into contact with the sample must be decontaminated and disposed of in accordance with the techniques used in the laboratory for decontamination and disposal of potentially infected material.

## REFERENCES

- B.G. Metchock, F.S. Nolte & R.J.Wallace, 1999. *Mycobacterium*, p.399-437 In P.R.Murray, E.J Baron, M.A. Pfaller, F.C. Tenover and R.H. Tenover (eds.). *Manual of Clinical Microbiology*, 7th ed. American Society for Microbiology, Washington, D.C.
- Murray, P.R. (ed.) 1999. *Manual of Clinical Microbiology*, 7th ed. American Society of Microbiology, Washington, D.C.

## PACKAGING

Cat. No : MB-80276 Ziehl-Neelsen Color Kit	3 X 250 mL
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