



**IVD** dispositivo medico-diagnostico in vitro

Mycobact for cold staining of Mycobacteria technical information  
 Technical's card code 12-113  
 Product code 12-113  
 Pack 1kit. Number of tests 100 or on request  
 Stability of product properly conserved at 15-20°C 24 months

Produce in Italy by  
 DDKItalia S.r.l  
 Via Marche, 19 • 27029 Vigevano (I)  
[info@ddkitalia.com](mailto:info@ddkitalia.com)•[www.ddkitalia.com](http://www.ddkitalia.com)

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Koch's bacillus (*Mycobacterium tuberculosis*) belonging to the family of Mycobacteriacee is the bacillus responsible for tuberculosis in humans. Bacilli are property, non-sporing, aerobic required, the size of 1-10 x 0.2-0.6 µm, characterized by slow growth, from a wall rich in mycolic acids and a DNA with high content of guanine and cytosine (60 - 70%).

**Method**

Use clean slides.  
 Cover the slide with the Kinyoun solution for five ten minutes depending on thickness of the smear  
 Wash in tap water  
 Decolorize the excess with Degommier  
 Countre stain with Gabett solution for two minutes  
 Wash in tap water for two minutes  
 Air dry  
 Examine immersion (1000x)

**Results**

Acid fast bacilli are colored in red on a blue background.

Acid resistant bacilli appear stained in red on the blue background of the preparation. In every case, the bacteriologist's answer should always refer to the number of observed field and therefore be expressed as "absence of BAAR on 200 (or 100) microscopic fields" and not as "bacilloscopy was negative". The answer, "bacilloscopy was positive" is also a bad answer because it does not give any information on the sputum's relative richness in bacteria. It is essential that a quantitative result be issued.

**Comment.**

Observation of one bacterium over all the slides raised a doubt and should always cause the microscopic examination to be repeated on other samples. Increase the staining time for fuchsin, according to the smear thickness.

**Reagent**

Kinyoun solution	2x30 ml
Degommier decolorizing solutions	2x30 ml
Gabett solution	2x30 ml

**Staining according to HOK**

During all phases of the above methods, the preparations must be washed in water free of alcohol-acid resistant species.  
 Cover the plate with Kinyoun solution and leave in contact for 3 minutes.  
 Wash with water for 30 seconds.  
 Cover the plate with Gabett solution and leave in contact for 1 minute.  
 Wash with water and dry.  
 Observe the preparation under the microscope using the immersion lens.

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#### Hot Devulder method

Cover the plate with Kinyoun solution and heat for 10 minutes.  
Wash with water.  
Cover the plate with Gabett solution. Leave in contact for 5 minutes.  
Wash with water and dry.  
Observe the preparation under the microscope using the immersion lens.

#### Cold Devulder method

Immerse the plate in the Kinyoun solution for about 3 hours.  
Wash with water.  
Cover the plate with Gabett solution. Leave in contact for 5 minutes.  
Wash with water and dry.  
Observe the preparation under the microscope using the immersion

\* Technical's note: staining time vary according to age, types of solutions, thickness of sections, et. When Gill (code 09-178) modified solution is used, get the best result, staining time (maximum 1-5 minutes), for best change in color, wash quickly in tap water, and then in Scott acidulated solution, (code 00-136). For sections fixed in Bouin, we recommend the use of haematoxylin modified acid AB (code 09-183). Please note the alcoholic loses eosin stain with the use, of the days are stretched over time colouring. If you are using purified eosin, check the time, and possibly diluted in ethyl alcohol 96°C, if the cytoplasmic staining was too strong. Before use, filter the following solutions; alcoholic eosin, eosin phloxine; Harris haematoxylin, Gill's haematoxylin. The acidified aqueous solution of eosin is prepared by slowly adding glacial acetic acid. Follow normal precautions for laboratory reagents. Dispose of waste according to regulations at the local, regional or national level. Refer to Data Sheet Material Safety Data for updated information on risks, hazards and safety associated with the use of these products.

\* Risk and Safety Statements outside the EU.

The eosin solution in alcohol is flammable and harmful. Harmful by inhalation, in contact with skin or if swallowed. Harmful: possible risk of irreversible effects through inhalation, in contact with the skin or by ingestion. Irritating to eyes, respiratory system and skin. Keep away from sources of ignition - No smoking. Wear suitable protective clothing and gloves. In case of accident or if you feel unwell, seek medical attention immediately (show the label where possible). Target organs: eyes and nerves. Eosin in aqueous solution. Caution: substance not yet fully tested. Avoid contact and inhalation of the solution of Harris haematoxylin. Organs: heart and nerves. Solutions based hemallum are harmful. Harmful if swallowed. Irritating to eyes, respiratory system and skin. In case of contact with eyes, rinse immediately with plenty of water and seek medical attention. Wear suitable protective clothing. Organs affected: liver and kidneys.

In case of accident or if you feel unwell, seek medical attention immediately (show the label where possible).

\* Risk and Safety Statements (U.E.)

The eosin solution in alcohol is highly flammable and harmful. Highly flammable. Harmful by inhalation, in contact with skin or if swallowed. Harmful: possible risk of irreversible effects through inhalation, in contact with the skin or by ingestion. Keep away from sources of ignition - No smoking. Wear suitable protective clothing and gloves. In case of accident or if you feel unwell, seek medical attention immediately (show the label where possible).

Eosin in aqueous solution. Caution: Substance not yet fully tested. Solution of hemallum. Do not breathe vapors. Avoid contact with skin and eyes. Gill haematoxylin Solutions are harmful. Harmful if swallowed. Irritating to eyes, respiratory system and skin. In case of contact with eyes, rinse immediately with plenty of water and seek medical attention. Wear suitable protective clothing.

\* Sample preparation

All samples must be treated according to the technology. All samples must be marked so as to be easily identified. Tools should be used for sampling and sample preparation, which must be observed strictly to manufacturer's instructions about the application and instructions. Diagnostics. The diagnosis should be performed only by authorized and trained persons. Valid nomenclatures must be used. Further tests must be selected and implemented according to recognized methods.

\* Conservation. The staining solution should be stored at a temperature between +15°C to 20°C, the dye at +5°C to 30° C. Store at 4-6 °C all kit containing silver solutions and Schiff reagents. The solution and dyes must be used before the expiration date. Stability. After first opening the bottle, the dye solution and the dyes are stable until the expiration date when stored at the temperature requested. Always keep the bottles tightly closed.

\* Instructions for use

To avoid errors, the staining process must be performed by qualified personnel. For professional use only. Must observe the National guidelines for work safety and quality assurance. Microscopes are used according to the standard. Protection against infection. Must be taken with laboratory guidelines for the protection against infection. Instructions for disposal. The solutions used and those have expired must be disposed of as special waste according to local regulations regarding disposal of waste.

Endnotes

- 1 The timing suggested in the leaflet are approximate and may vary according to your specific needs. If they are used intensively, for staining solutions may lose their dyes, so it is necessary to extend the time of staining solutions, or replace with new products.
2. Include positive control slides in each session.
3. Some hydraulic systems deliver acidic water, unsuitable for use for the part of the procedure for the blue coloration. If tap water is acidic, instead using a dilute alkaline solution, for example, water buffered by Scott.
4. The presence of purple or red-brown nuclei a blue color indicates unsatisfactory.
5. If you over-eosin staining, nuclear staining may be masked. If done correctly, with eosin staining shows a three-tone effect. To increase the differentiation of eosin, extend the time of immersion in alcohol, or use a first alcohol with a higher water content. You can adjust the times of immersion in alcohol to obtain an adequate eosin staining.
6. We do not recommend the addition of stock solution in the working solutions of haematoxylin and eosin.
7. Avoid excessive drag (carryover) of water solutions in alcoholic eosin.
8. The data generated by this procedure are to be used only to support the diagnosis and should be evaluated in conjunction with other tests and diagnostic data

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