

Protocol n° 106

MCDh* Staining

*Micro Chromatic Detection for haematology

Standard Procedure

Principle:

The MCDh panoptic staining allows to perform medullary and blood cell counting, realised by using successively four reagents: MCDh 1, MCDh 2, MCDh 3 and MCDh 4.

MCDh 1, formulated with ethylic alcohol, is a mixture of neutral stains. It allows a smear fixation and prepares the staining, especially the one of hydrosoluble elements such as basophilic granules.

Those stains are inactive in alcoholic medium, and only react selectively when released in MCDh 2 solution. This releasing generates the precipitation of neutral stains, leading to erythrocytes, cytoplasm of neutrophilic granulocytes as well as eosinophilic granules staining. MCDh 3 is a blue aqueous solution which stains cytoplasm of monocytes and lymphocytes. MCDh 3 also eases the metachromasia process as it colours azurophilic granules red. Eventually, MCDh 4 removes excess of stain and participates to differentiation of cellular elements thanks to action of specially selected rinsing agents.

The successive action of MCDh 1, MCDh 2, MCDh 3 and MCDh 4 brings the violet colour (typical Romanowsky-Giemsa effect), particularly visible in chromatin, platelets and neutrophilic granules.

Necessary staining products:

MCDh 1 Ref. 313590-	1000 mL, 2500 mL
MCDh 2 Ref. 313570-	1000 mL, 2500 mL
MCDh 3 Ref. 313560-	1000 mL, 2500 mL
MCDh 4 Ref. 313600-	1000 mL, 2500 mL

Processing Time: 9 minutes and 50 seconds (blood smears)
13 minutes and 40 seconds (medullary smears)

Specimen preparation:

Specimen must be processed in accordance with procedures available in the laboratory and promulgated by national Authorities.

Staining procedure:

Please read all this information carefully before using this device.

Blood smears

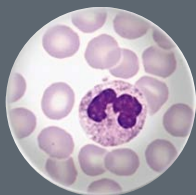
Processing Time: 9 minutes and 50 seconds

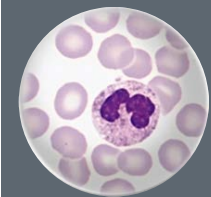
- Place in a bath of MCDh 1 6 minutes
- Drain surplus solution onto absorbent paper
- Place in a bath of MCDh 2 1 minute
- At the time to remove the slide, agitate slowly in the bath
- Drain surplus solution onto absorbent paper
- Place in a bath of MCDh 2 2 minutes
- At the time to remove the slide, agitate slowly in the bath
- Drain surplus solution onto absorbent paper
- Place in a bath of MCDh 3 40 seconds
- At the time to remove the slide, agitate slowly in the bath
- Drain surplus solution onto absorbent paper
- Place in a bath of MCDh 4 10 seconds with agitation
- Air dry

Medullary smears

Processing Time: 13 minutes and 40 seconds

- Place in a bath of MCDh 1 8 minutes
- Drain surplus solution onto absorbent paper
- Place in a bath of MCDh 2 1 minute
- At the time to remove the slide, agitate slowly in the bath
- Drain surplus solution onto absorbent paper
- Place in a bath of MCDh 2 2 minutes
- At the time to remove the slide, agitate slowly in the bath
- Drain surplus solution onto absorbent paper
- Place in a bath of MCDh 3 2 minutes and 30 seconds
- At the time to remove the slide, agitate slowly in the bath
- Drain surplus solution onto absorbent paper
- Place in a bath of MCDh 4 10 seconds with agitation
- Air dry



**Results:**

Nuclei / Chromatin: ± Dense purple

Granulocytes

Cytoplasm without RNA: Light purplish-pink

Eosinophilic granules: Orangey

Basophilic granules: Dark blue

Neutrophilic granules: ± Deep purple

Lymphocytes

Cytoplasm with RNA: Pure blue

Cytoplasm without RNA: Light blue

Azurophilic granules: Red

Monocytes

Cytoplasm: Cloudy blue

Erythrocytes: Pinkish-Beige

Platelets

Chromomere: Purplish-red

Hyalomere: Bluish

Blood Parasites / Plasmodium

Nucleus: Red

Cytoplasm: Blue

Recommendations and/or Note of use:

For professional use only.

In vitro use only.

The collection and processing of chemical biological waste must be conducted by specialized and registered companies.

Storage: 15 - 25 °C.

According to the thickness of the smear, especially in case of medullary smears, it may be necessary to increase the first bath staining time (MCDh 1).

Bibliography:

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