SPERM STAIN READY TO USE

MICROPTIC
AUTOMATIC DIAGNOSTIC SYSTEMS

Principle

The **SPERM STAIN READY TO USE** constitutes a system to differentiate the morphological structures of the spermatozoa for a perfect functional assessment. It can also be used for the differential staining of blood cells in semen. The stains which make up **the SPERM STAIN READY TO USE** combine polychromy and the quality of classic cytology staining methods (May-Grünwald, Giemsa, Wright) with a very quick execution (time of just 15 seconds). The technique is performed by immersion in the staining solutions.

Product characteristics

The **SPERM STAIN READY TO USE** system is based on the original Romanowsky staining method for differential staining of several cellular structures.

Reagents

Kit 3 x 500 ml. (Ref. 98 30 95). Content:

1. **SPERM STAIN READY TO USE NR. 1**

1 x 500 Ml (Ref. 98 16 85)

Hexamethyl-p-roseniline methanolic solution. Handle with care.

2. SPERM STAIN READY TO USE NR. 2

1 x 500 mL (Ref. 99 42 43)

Xanthene buffered solution.

3. SPERM STAIN READY TO USE NR. 3

1 x 500 mL (Ref. 99 24 29)

Thiazine buffered solution

Working reagents

All the reagents are ready for use.

Storage and stability

The components of the kit, stored at room temperature (10-35º C) will remain stable until the expiration date stated on the label. Containers must always be kept tightly closed. Over time and temperatures below 10ºC could lead to the formation of light precipitate in some regents. This does not affect their functionality. The stain should be filtered before use. It is recommended to bring the reagent to room temperature (15-30ºC), agitate and filter before using.

Inuse stability should be determined by each user discretion.

Caution

Reagent nº 1, as a methanolic solution is flammable and toxic by contact, inhalation and ingestion. Handle with care. Waste products must be handled attending your local regulations.

Sample preparation

Sperm samples: prepare the smear with 15 µl of fresh sperm on a standard glass slide. Leave it for air drying at least 10 minutes. It is recommended to make a thin and homogeneous spreading of the sample for the best fixation of the dyes and avoiding blurring hypercolorations.

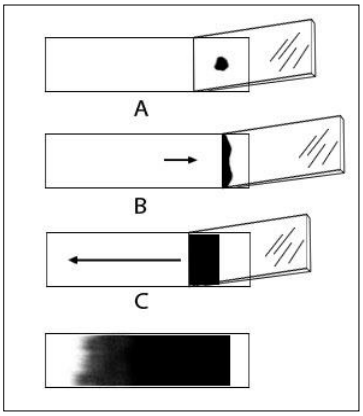
Staining procedure

1- Fix the smear by immersion in the Working Reagent Nr. 1 ; 5 times, 1 second each. Let drain.

2- Stain the smear by immersion in the Working Reagent Nr. 2 ; 5 times, 1 second each. Let drain again.

3- Stain the smear by immersion in the working Reagent Nr. 3 ; 5 times, 1 second each.

Rinse the smears gently with deionised water and leave them for air-drying.



In order to prevent deterioration of the smears by the immersion oil coversliping is recommend with an appropriate mounting media such as DPX, Eukitt ®. Use two drops and a standard glass coverslip (50 x 22 mm).

Remarks

Staining intensity can be modified by varying the number of the immersions in solutions 2 and 3, depending on what colour is preferred to be emphasized.
Curettes with stain shall always be stored capped, specially 1 in order to avoid undesirable evaporations that could promote colour deviations from the usual stainings.

Results

Sperm head: Dark violet

Sperm acrosome: Pale violet, clearer then the head colour

Midpiece and tail: dark violet

Background: Pale pink

Red blood cells: Pale or deep pink

Neutrophils: Dark blue nucleus. Pink cytoplasm with red-violet granulations

Eosinophils: Blue nucleus. Blue cytoplasm with red or red-orange granules

Basophils: Dark blue or purple nucleus. Purple, almost black, granules

Lymphocytes: Violet nucleus. Sky blue cytoplasm

Monocytes: Very pale violet nucleus. Sky blue cytoplasm

References

Gurr,E. (1965) "The rational use of dyes in Biology", p. 115. Leonard Hill, London.

Gurr,E. (1971) "Synthetic dyes in Biology, Medicine and Chemistry". Academic Press. London & New York

Maree, L.; du Plessis, S.S.; Menkvels, R. and van der Horst, G. Human Reproduction, 25 (6), 1369 – 1382 (2010).

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