# Sperm Vitality Kit

Quantitative eosin staining kit for the percentage measurement of live spermatozoa in semen samples

# MenidiMedica **Biotechnology** Applications



#### GENERAL INFORMATION

Sperm vitality is reflected in the proportion of spermatozoa that are "alive". Sperm vitality should be determined in semen samples with less than about 40% progressive motile spermatozoa. Sperm Vitality Test uses the eosin staining technique to establishes the percentage of live spermatozoa. The technique is based on the principle that dead cells will take up the eosin, and as a result stain red. Sperm Vitality Test provides an accuracy check of the motility evaluation since the percentage dead spermatozoa should not exceed the percentage immotile spermatozoa. Sperm Vitality Test may help in assessing the diagnosis and the management of male infertility.

MATERIAL INCLUDED IN THE KIT Reagent A - 1 ml of red stain

# MATERIAL NOT INCLUDED IN THE KIT

Light microscope (400 - 600x magnification)

- Microscope glasses
- Cover glasses
- Pipettes
- Test tubes (sterile)
- Microscope slides Laboratory counter

#### METHOD

1. Mix the semen sample well

2. Remove an aliquot of 5  $\mu$ L of semen and combine with 5  $\mu$ L of reagent A on a microscope slide. Mix with a pipette tip, swirling the sample on the slide.

- 3. Cover with a 22 mm x 22 mm coverslip and leave for 30 seconds.
- 4. Remix the semen sample, remove a replicate aliquot, mix with reagent A and treat as in steps 2 and 3 above.
- 5. Examine each slide, preferably with negative-phase-contrast optics at x200 or x400 magnification.
- 6. Tally the number of stained (dead) and unstained (vital) cells with the aid of a laboratory counter.
- 7. Evaluate 200 spermatozoa in each replicate, in order to achieve an acceptably low sampling error.

8. Calculate the average and difference of the two percentages of vital cells from the replicate preparations.

9. Determine the acceptability of the difference.

10. If the difference between the percentages is acceptable, report the average percentage vitality. If the difference is too high, make 2 new preparations from 2 new aliquots of semen and repeat the assessment.

11. Report the average percentage of vital spermatozoa to the nearest whole number

# INTERPRETATION

- Colourless spermatozoa: live spermatozoa
- Red stained spermatozoa: dead spermatozoa Count between 100 and 200 cells and differentiate the living from the dead spermatozoa.

Read results immediately, waiting too long will yield lower vitality percentages. It is clinically important to know whether immotile spermatozoa are alive or dead. Vitality results should be assessed in conjunction with motility results from the same semen sample. The presence of a large proportion of vital but immotile cells may be indicative of structural defects in the flagellum; a high percentage of immotile and non-viable cells (necrozoospermia) may indicate epididymal pathology. A semen sample is considered normal if 58% or more of the sperm cells are alive

#### LIMITATIONS OF THE METHOD

Spermatozoa stained with Renata Sperm Vitality Test cannot be used for any further procedures.

#### STORAGE

Suitable for transport or short term storage at elevated temperatures (up to 5 days at 37°C). Store reagent between 2°C and 25°C.

### WARNINGS AND PRECAUTIONS

All human, organic material should be considered potentially infectious. Handle all specimens as if capable of transmitting HIV or hepatitis. Always wear protective clothing when handling specimens.

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