

## 200 tests

IN VITRO DIAGNOSTIC KIT FOR THE SEMI-QUANTITATIVE  
DETERMINATION OF HUMAN SPERM VITALITY BY DYE EXCLUSION

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Nigrosin contains 0.05% Na-azide  
Reagent for Professional Use only.

### 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.

VitalScreen uses the eosin-nigrosin staining technique to establish the percentage of live spermatozoa.

The technique is based on the principle that dead cells will take up the eosin through their cell membrane, and as a result stain red. The nigrosin provides a dark background which makes it easier to assess the slides.

VitalScreen provides an accuracy check of the motility evaluation since the percentage dead spermatozoa should not exceed the percentage immotile spermatozoa.

The VitalScreen kit may help in assessing the diagnosis and the management of male infertility.

### MATERIAL INCLUDED IN THE KIT

- Reagent 1 - 20ml of 1% Eosin Y in saline
- Reagent 2 - 30ml of 5% Nigrosin in saline

A certificate of analysis and the MSDS can be downloaded from our website ([www.fertipro.com](http://www.fertipro.com)).

### MATERIAL NOT INCLUDED IN THE KIT

- Light microscope (400 - 600x magnification)
- Microscope slides
- Cover glasses
- Pipettes
- Test tubes (sterile)

### PREPARATIONS

Shake reagent 2 (Nigrosin stain) before use.

### METHOD

We recommend to view our demonstration video (download via the link on our website, or scan barcode):



1. Assess semen sample, preferably within 1 hour after ejaculation.
2. Mix 50 µL of semen with 2 drops of reagent 1 in a sterile test-tube. Avoid contact between the reagent bottle and the tube containing semen. Mix gently.
3. After 30 seconds, add 3 drops of reagent 2 and mix thoroughly. Avoid contact between the reagent bottle and the tube containing semen.

4. Within 30 seconds of adding reagent 2, place 20µl of the semen-stain mixture on a microscope slide and immediately place a coverslip on top.
5. Read immediately under the microscope.

Note 1: Do not wait until the drop has dried out, crystals of nigrosin will form which can interfere with the interpretation of the results.

Note 2: In case nigrosin precipitates in the semen sample (rarely occurs in viscous samples), it is recommended to only work with Eosin (Reagent 1) and not to use Nigrosin (Reagent 2) (after step 2, proceed to step 4).

### INTERPRETATION

- Colourless spermatozoa: live spermatozoa
- Spermatozoa stained red: dead spermatozoa

Scan the complete microscopic slide and count between 100 and 200 cells. 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 (WHO, 2010).

A semen sample is considered normal if 58% or more of the sperm cells are alive.

### LIMITATIONS OF THE METHOD

Spermatozoa stained with VitalScreen cannot be used for any further procedures.

### STORAGE AND STABILITY

Suitable for transport or short term storage at elevated temperatures (up to 5 days at 37°C). Store reagents between 2°C and 25°C. The reagents are stable for 24 months after date of production.

### 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.

### BIBLIOGRAPHY

WHO laboratory manual for the Examination and processing of human semen, WHO, 5th ed., 2010.



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