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Art.-No ZR1060201



(Seminal Hypo-Osmotic Test)

Distribution:

GYNEMED

Application

IVD

This HOS test is used to test the vitality of sperm cells. The hypo-osmotic swelling is based on the semipermeability of the intact cell membrane and their ability of active water transport, in order not to burst. In sperms with intact membranes the flagellum swells up within 5 min. This change remains stable up to 30 min.

Principle

In this hypo-osmotic swelling test swelling of cells only occurs in vital cells with an intact membrane by using hypotonic solution.

Storage

2-8°C. Sterile sampling. Contains no antibiotic.

Stability

24 months from date of manufacture. After opening use within 7 days.

Content

SemenHOS solution 20 ml

Necessary utensils

- Coverslips (18 x 18 mm)
- Gloves
- Contrasting phase microscope
- Native ejaculate or washed sperm (105-110 μl)
- Slides
- Paper towels
- Pipettes and tips (10-100 µl)
- Water bath or heating cabinet (37°C)

Preparation of SemenHOS solution

Preheat the SemenHOS solution at 37°C

Procedure

- 1. Occasionally native ejaculate without additions includes hypo-osmotic sperm forms. Transfer 5-10 µl liquefied semen without air bubbles to a slide and cover it with cover slips. Microscope at 400x magnification. This is the zero-value.
- 2. Examine the percentage of sperms with swollen flagellum by observing 100 sperms, calculated in duplicate. Note this value note* (a₀ %).
- 3. Add 100 µl ejaculate without air bubbles to 900 µl preheated SemenHOS solution and mix
- 4. Incubate this mixture for 10 min at 37°C.
- 5. Transfer 10 μI of the mixture to a slide and place and cover it with a cover slip.
- 6. Microscope at 200x or 400x magnification.
- 7. Repeat twice step 2 to 6.



Fig.1: Schematic representation of typical morphological changes of human spermatozoa after exposure with hypo-osmotic solution (extract WHO 2010).

No change (a). Different tail changes (b-g). The swelling in the tail region is indicated in gray.

Evaluation:

Percentage of vitality of spermatozoa

Calculate the difference between the percentage of sperms with swollen flagellum before and after incubation with the SemenHOS solution.

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Example:

Before incubation:

1. Count 2/100

2. Count 3/100

Mean value: 2.5 /100= 2.5%

After incubation:

- 1. Count 88/100
- 2. Count 97/100
- Mean value: 92.5/100= 92.5%

Result: 92.5% - 2.5%= 90%

90 % of the sperms are vital

The HOS test is regarded as normal, if after incubation more than 60% of the sperms show a swollen flagellum. The sample is not normal when the result is less than 50% (WHO 2010).

Safety information / Precautions

- All semen samples should be considered potentially infectious.
- Handle with all samples like HIV or hepatitis infected material.
- When working with samples and reagents wear always protective clothing (gloves, gowns, eye / face protection).
- All ingredients of reagents are classified as nontoxic

References

Drevius L, Eriksson H, (1966) Osmotic swelling of mammalian spermatozoa, Experimental *Cell Research*, 42: 136-56

Jeyendran RS, et al, (1984) Development of an assay to assess the functional integrity of the human sperm membrane and its relationship to the other sperm characteristics, *Journal of Reproduction and Fertility*, 70: 219-28

WHO Press, (2010) Laboratory manual for the examination and processing of human semen, $5^{\rm Th}$ edition

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