

HT CASA Counting chambers Protocol

DISPOSABLE COUNTING CHAMBER TO ANALYSE MOTILITY and CONCENTRATION OF SEMEN (10 micron – 2 chambers)

Principle:

These slides are suitable for the analysis of sperm motility and concentration with the HT CASA systems. However, can also be used for manual assessments in case a calibrated eye-piece reticule is mounted in the microscope ocular.

Sperm cells distribute homogenously over the chamber by capillary action. As disposable chambers, no washing is required after use. This fact minimizes the risk of contamination resulting from the re-use of chambers.

Product characteristics:

1 Box content: 25 slides/ 50 tests

Material required:

Warming stage (In case of assessing motility at 37°C)

Pipette and tips

A piece of paper tissue

Storage and stability:

HT CASA Counting chambers should be stored in a closed box, protected from sunlight, and at room temperature. The shelf life of the slides is two years, but haze can form inside the chamber. This can be

washed away after filling without affecting the functioning of the chamber.

Loading instructions:

Step 1: Aspirate ~ 2,5 µl of fully liquefied and homogenized semen.

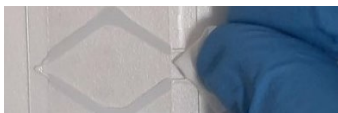
Carefully place the tip of the pipette on the slide within the boundaries of the loading area. Be careful not to touch the coverslip to prevent premature filling of the chamber.



In just one step, push the piston down till the first resistance to load the loading area. The chamber will be filled by capillarity. If chamber is loaded in more than one step air-bubbles could be inserted inside the chamber. Thus, affecting the reliability of results.

Depending on the viscosity of the sample filling will take 3 - 20 seconds or even more.

Step 2: Carefully, take away the excess of sample with a paper towel or similar to stop the drifting of cells inside the chambers.



For performing **automated semen assessment** with the HT CASA systems start the analysis **immediately after Step 2** to prevent evaporation of the sample.

Manual assessments should be performed according to WHO standards and with the help of a calibrated eye-piece reticule on the microscope ocular, considering the magnification factor used on each case.

NOTES:

If the analysis of sample concentration cannot be performed directly after filling, and if no motility analysis is needed, the inlet and air outlet of the chamber can be sealed with nail polish to prevent evaporation.

It's recommended to prewarm slides and pipette tips at 37°C to prevent temperature shock during sample handling.

Sperm motility features are variable according to the type of sample analysed (fresh semen, capacitated sperms, etc.). Hence, manufacturer recommends using chambers of 10 microns depth for evaluating fresh or diluted semen samples and 20 microns depth chambers for capacitated/hyperactive sperms and/or very viscous semen samples.



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