

# FERTILITY SOLUTIONS

THE SEMEN ANALYSIS EXPERTS®

Sperm Wizard SPERMOCYTOMETER®

Fertility Solutions Inc.

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Manufactured by Leja Products B.V Netherlands

See our website for the complete line of semen analysis quality control and training products.

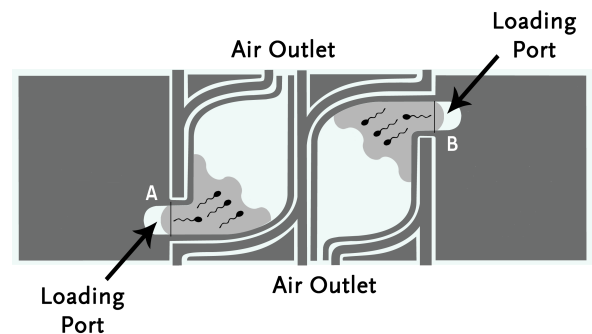
## Requirements:

**Microscope** (phase contrast with 20X objective recommended) fitted with KR406B 10x10 eyepiece reticle (reticles.com)

**Scaling Factor** for objective and microscope (FSI Technical Bulletin "Scaling Factor Determination" available on our website)

**Micropipetor** with 6 $\mu$ L capacity and tips

**Semen sample or QC**

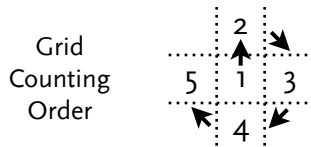


## Loading the SPERMOCYTOMETER®:

1. Mix semen well using vortex mixer, 2-3 pulses of 3-5 seconds each.
2. Slowly dispense ~6 $\mu$ L of sample into a chamber loading port (A or B) and observe filling. The chamber is filled when the sample reaches the air outlet. A small amount of seminal fluid volume may remain in loading area.
3. Position the SPERMOCYTOMETER® on the microscope stage so chamber is under the objective. Wait 1-2 minutes for the sample to settle until no drifting is observed.
4. Scan the chamber to verify the absence of clumping and air bubbles.

## Counting Sperm:

1. Position the center of the chamber under the microscope objective.
2. Count the sperm in **all** 100 squares of the reticle grid using Sperm Counting Rules. Do not count partial grids.
3. Record the number of sperm in the grid.
4. Repeat steps 2 & 3 in four adjacent fields.



## Calculate Sperm Concentration:

$$\text{Grid 1} + \text{Grid 2} + \text{Grid 3} + \text{Grid 4} + \text{Grid 5} = \text{Total Sperm}$$

$$\frac{\text{Total Sperm}}{500 \text{ (squares counted)}} = \text{Average \# of sperm per square}$$

$$\text{Average \# of sperm per square} \times \text{Scaling Factor} = \text{Sample Concentration in million/mL}$$

## Additional Notes:

- For calculation examples, visit our website.
- Round cells and aggregation/agglutination also can be evaluated using this chamber.
- For semen analysis procedure including motility, call Technical Service: (800) 959-7656
- Mark each chamber after use with permanent marker to indicate it was used.

## Sperm Counting Rules

- Analyze each square of the grid individually.
- Analyze all 100 squares in the grid and a minimum of 5 grids.
- Count only intact sperm with head and tail.
- Count sperm whose heads lie completely within the square and any heads that lie on the upper and left lines. Do not count tail-less heads, head-less tails or sperm whose heads lie on the lower and right lines.
- If the total number of sperm in 5 grids is less than 200, consider counting another aliquot to confirm.
- If a high sperm concentration makes counting difficult, dilute the semen and reload a new chamber.
- For post-vasectomy samples, examine entire chamber using your laboratory's procedure.

## Troubleshooting:

### Incomplete Chamber Load

*Sample volume may be insufficient*  
Solution: check pipettor settings & calibration

*Semen may have high viscosity or incomplete liquefaction*  
Solution: add chymotrypsin to semen, incubate and load new chamber

### Clumping

*Can result from incomplete mixing*  
Solution: mix thoroughly before loading

*Sperm aggregation or agglutination may indicate an abnormal pathology.*

### Field suddenly shifts

*Air may have been introduced in loading*  
Solution: load new chamber