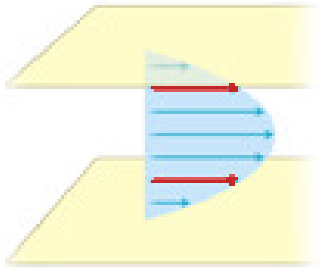
INSTRUCTIONS for Segre Silberberg correction factor

Leja® disposable counting chambers

What is the Segre Silberberg effect?

Capillary flow into a 20 µm counting chamber follows a classical Poiseuille flow. The flow of the fluid is dominated by its viscosity. A maximum velocity is reached at exactly half the depth of the chamber (see diagram) while the velocity at the chamber walls equals 0 µm sec-1.



One can image that the sperm-cells in the middle of the chamber-height move faster than the ones near the wall. It has been shown that all sperm-cells move to two planes at equidistance from each chamber wall (depicted by the red arrows in the diagram).
The distance of these planes from the wall (β) is depending on a few parameters:

- development of full Poiseuille flow
- chamber height
- surface properties of the counting chamber
- surface tension
- fluid viscosity
- flow velocity
- diameter of sperm-cell head

How to correct for the Segre Silberberg effect?

Because the sperm-cells in the two Segre Silberberg planes move faster than the average fluid velocity, there is an accumulation of sperm-cells at the filling front. When measuring the sperm concentration in the center of the Leja® slide, an underestimation of the concentration takes place.
Luckily, this is a constant underestimation that can be corrected for.

Since all variables are kept constant the only variable that affects the dimension of the Segre Silberberg effect is the viscosity of the sample. Filling time and viscosity are closely associated. By measuring the filling time of the Leja chamber with a capillary length of 21 mm, the Segre Silberberg compensation factor *Sx* for that specific sample can be read from the conversion table.

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The correction factor is only a constant when using a linear flow slide like the Leja® new design 2 chamber and the Leja® 4 chamber slides. Working with these slides will yield in an exact count that matches hemocytometer counts.
Due to the chamber height of 100 microns, the Segre Silberberg effect is negligible.

How to measure the filling time?

Load the sample into the chamber using a positive displacement pipette. Hold the pipette at an approximate angle of 45° and slowly deposit 1.5 times the indicated volume in the entry port.
Start measuring as soon as the capillary filling starts and stop when the liquid has filled the counting chamber completely.

Correction Factor conversion table

Filling time (sec.)	*Sx*	*Viscosity 2 chamber slide (cP)*	*Viscosity 4 chamber slide (cP)*
2.0	*1.32*		
2.1	*1.31*		
2.2	*1.30*		
2.3	*1.29*		
2.4	*1.28*		
2.5	*1.27*		
2.6	*1.26*		
2.8	*1.25*		
2.9	*1.24*		
3.2	*1.23*	*2.16*	*2.25*
3.4	*1.22*	*2.21*	*2.32*
3.6	*1.21*	*2.27*	*3.38*
3.8	*1.20*	*2.32*	*2.44*
4.0	*1.19*	*2.37*	*2.50*
4.2	*1.18*	*2.43*	*2.56*
4.5	*1.17*	*2.51*	*2.66*
5.0	*1.16*	*2.64*	*2.81*
5.3	*1.15*	*2.72*	*2.90*
5.5	*1.14*	*2.77*	*2.96*
6.0	*1.13*	*2.90*	*3.12*
7.0	*1.11*	*3.16*	*3.43*
8.0	*1.10*	*3.43*	*3.74*

These values only hold for the Leja 20 micron chambers (SC-20-01-02-B and SC-20-01-04-B)