## Segre Silberberg Effect

Capillary flow into a 20-micron counting chamber (fixed coverslip type) 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 while the velocity at the chamber walls equals 0 micron/second. The sperm cells or beads (further – "particles" for simplicity) in the middle of the chamber height move faster than the ones near the wall. It has been shown that during chamber filling all particles move to two planes at equidistance from each chamber wall with velocity gradient. The distance of these planes from the wall depends on several parameters:

- Development of full Poiseuille flow
- Chamber height
- Surface properties of the counting chamber
- Surface tension
- Fluid viscosity
- Flow velocity
- Particle diameter

Because particles in the two Segre Silberberg planes move faster than the average fluid velocity, there is an accumulation of them at the filling front that impacts their distribution throughout the chamber volume. As sample viscosity is not a constant parameter for semen and beads (semen is much more viscous as compared to beads spread in a water-based media), it impacts their distribution in a fixed coverslip chamber. Correspondingly concentration of the sperm and beads cannot be accurately counted under the SAME conditions. In fact, semen viscosity prevents the even spread of spermatozoa throughout the fixed coverslip volume resulting in accumulation at the filling point, whereas the water-based suspension of beads is loaded faster with minimal accumulation at the filling point.

## **Counting Sperm**

In order to insure accurate manual counting of SPERMATOZOA using a fixed coverslip chamber in the SQA-V or SQA-Vision visualization compartment, the assessment should be performed at ZOOM-OUT (low magnification and wide field of view) which will compensate for the impact of semen viscosity.

## **Counting Beads**

For accurate manual counting of BEADS using a fixed coverslip chamber in the SQA-V or SQA-Vision visualization compartment, assessment should be performed at ZOOM-IN (high magnification and narrow field of view) which will compensate for the very low bead viscosity.

## <u>Notes</u>

- This technical bulletin is related to fixed coverslip slides only.
- It is not related to the other counting chambers which have different loading principles such as the hemocytometer, Makler chamber and standard slides.