Calculation for Sperm Concentration using the haemocytometer. Reference WHO5 pages 32+



On the haemocytometer, there are 9 grids each containing 100nl (0.1ul).



In the central grid there are 25 squares (each with a volume of 4nl-25x 4=100nl).

If you count 1 sperm in 1 square, the concentration is

- 1 sperm/4nl •
- 0.25 sperm/nl
- 2.5 sperm/10nl •
- 25 sperm/100nl
- 250 sperm/1000nl (ul) •
- 250sperm/ul •
- 250,000 sperm/ml.

By extension

If you count 4 sperm/1 square, the concentration is

- 4 x 250,000 sperm/ml •
- 1 million/sperm/ml

If you dilute the sample 1/10 and count 4 sperm /1 square, the concentration is

- 10x4 x 250,000 sperm/ml
- 10 million/sperm/ml
- If you dilute the sample 1/10 and count 20 sperm /5 square, the concentration is
 - 10x4 x 250,000 sperm/ml
 - 10 million/sperm/ml •

Therefore the equation for estimating the concentration by haemocytometer is

A=(the number of sperm counted/number of squares) [average number/square] B=A/4nl [number sperm per unit volume of 1 nl] Concentration=B x the dilution

OR (wait for it)

Concentration= (number of sperm counted x Dilution)/(number of squares x 4) in million/ml

Examples...

- 1. sample diluted 1/10 had a count of 50 sperm in 5 squares a. Concentration=(50 * 10)/(5 * 4)=500/20=25 million/ml
- 2. sample diluted 1/10 had a count of 20 sperm in 5 squares a. Concentration=(20 * 10)/(5 * 4)=200/20=10 million/ml
- 3. undiluted sample had a count of 50 sperm in 5 squares a. Concentration=(50 * 1)/(5*4)=50/20=2.5 million/ml
- 4. undiluted sample had a count of 20 sperm in 5 squares a. Concentration=(20 * 1)/(5*4)=20 /20=1.0 million/ml
- 5. sample diluted 1/20 had a count of 500 sperm in 5 squares
 - a. Concentration=(500 * 20)/(5 * 4)=10,000/20=500 million/ml