



## Specification – Handling instructions Cell Count Standards (Calibration solution)

### Composition

Raw milk (no pure *Leucocyt – Standard!*); preserved with Bronopol\*).

### Samples

Cell count milk “low”	: ~ 150.000 - 200.000 cells/ml
Cell count milk “high”	: ~ 350.000 - 450.000 cells/ml
Cell count milk “extra high”	: ~ 650.000 - 750.000 cells/ml
Goat milk “low”	: ~ 800.000 - 1.000.000 cells/ml
Goat milk “high”	: ~ 1.400.000 - 1.600.000 cells/ml
Goat milk “extra high”	: ~ 1.800.000 - 2.200.000 cells/ml

Only raw  
goat milk

### Application – delivery

**a)** every 4 weeks

– interlaboratory studies – evaluation of the measuring results

**b)** Control milk for cell count measuring instruments – control of the correctness.

\*) Dosage: 0,2% (0,0002% of 2-Bromo-2-nitropropane-1,3-diol)

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## Accuracy, precision

- ❖ repeatability (Standard deviation of repeatability/arithm. mean \* 100) : 2,4-4,7%
- ❖ reproducibility (Standard deviation of reproducibility/arithm. mean \* 100) : 4,0-5,0%
- ❖ uncertainty of measurement (calculated for each Batch) : 0,04-0,05 log  
~ +/-10% (2xVK% R)


## Correctness – Calculation of standard value

- ❖ The reference value is determined on the basis of two different fluorescence optical measurement systems, such as BacSomatic TM (Foss, DK) and De Laval Cell Counter DCC.
- ❖ NEW: Since 2021, the so-called "gold standard" (ERM-BD001) of the EU has also been measured to ensure the measurement level. It has been shown that the measurement level of this "Goldstandard" correlates very well with the reference values of the "Hüfner" cell count standards. Any differences are mostly due to the relatively difficult sample preparation of the ERM-BD001 standard.
- ❖ The reference value has an accuracy of 10% (the measurement uncertainty is calculated for each sample). However, the measurement results show that the variance is usually significantly lower for this type of cell count sample.

## Portioning – Storage - Stability

The Cell Count Milk should be portioned in smaller units (of 40 ml each) and deep-frozen on the day of its arrival or the day after. The arrival time and temperature of the standards should be noted. The temperature should be lower than 10°C. Temperatures of more than 15°C are critical. If the Cell Count Standards are not in a satisfactory condition, we will send a replacement.

Due to the refrigerating accumulators, the local forming of ice is possible.

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It is essential to ensure that the standards do not contain any ice lumps during portioning and freezing, as this can lead to crystallization of the fat (during thawing). The filling temperature should be between 5°C and 10°C.

The cell count milk bottles have a free head space. The 1-liter bottles must be swirled approx. 25 times before filling. Excessive stirring/shaking must be avoided. The small vials used must be clean and dry and must not contain any detergent residues.

The filled vials must be placed at a distance to achieve rapid freezing.


**Important:** Make sure that the milk - before filling into smaller units - is completely thawed and does not contain any ice particles etc.. Please ensure that the portioned bottles are placed in the freezer within 4-6 hours. To ensure rapid freezing, pre-chill the filled vials to <6°C (4°C). Freezing temperatures of -18°C to -30°C are sufficient. Under these conditions, the cell count milk is stable for a period of ~6 months. Temperatures that are too low (below -30°C) should be avoided.

## Examination

Please thaw up the samples in water (~ 20°C)– with occasional shaking. The measuring should be made according the manual of the instrument (warm up to 40°C!).

Please investigate as soon as possible. If there is something wrong with the cell count milk you will get a new delivery free of charge immediately!

After thawing, the Cell Standards, especially samples with high (> 600,000) counts, should be cooled immediately (<4 ° C). Otherwise, a decrease in the number of cells can be expected. In general, long (> 60 min.) warming phases (at room temperature) should be avoided in the cell count samples.

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# Labor Dr.Hüfner GmbH

If the samples reach the laboratory late (> 4 days) and at an elevated temperature (> 10 ° C), as a rule, the measurement results rather lower (cell disintegration!). In this case you will receive a free replacement. The tests must therefore be carried out immediately after the sample arrives!

## Results report - evaluations:

You should take 5-10 measurements per sample.

The results are to be entered in .xls Formpapers (form can be downloaded from the Internet at [www.mih-huefner.de](http://www.mih-huefner.de)).

The results are evaluated monthly - with regard to:

1. Correctness of the measurement results (deviation - z-score - to total mean and target value)
2. Repeatability (coefficient of variation for repeatability)
3. Reproducibility (coefficient of variation for reproducibility)

Hergatz, Nov. 2021

Dr. Josef Hüfner

Reference material manufacturer accredited by DAkKS according to DIN EN ISO 17034



The accreditation is only valid for the scope of accreditation listed in annex RM-21321-01-00

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