

# Differential cell count methods for measuring leukocytes in bovine milk samples

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## Background

Intramammary infection results in influx of leukocytes into the udder tissue and milk. To control milk quality, ensure food safety, and monitor udder health, the leukocyte content is continuously measured as the somatic cell count (SCC). Alternatively, the leukocytes can be differentiated and measured separately by the differential cell count (DCC)<sup>1-6</sup>. The aim of this study is to review methods used for DCC of bovine milk, and assess how this parameter can be used to monitor udder health.

## Measuring the leukocyte content in bovine milk

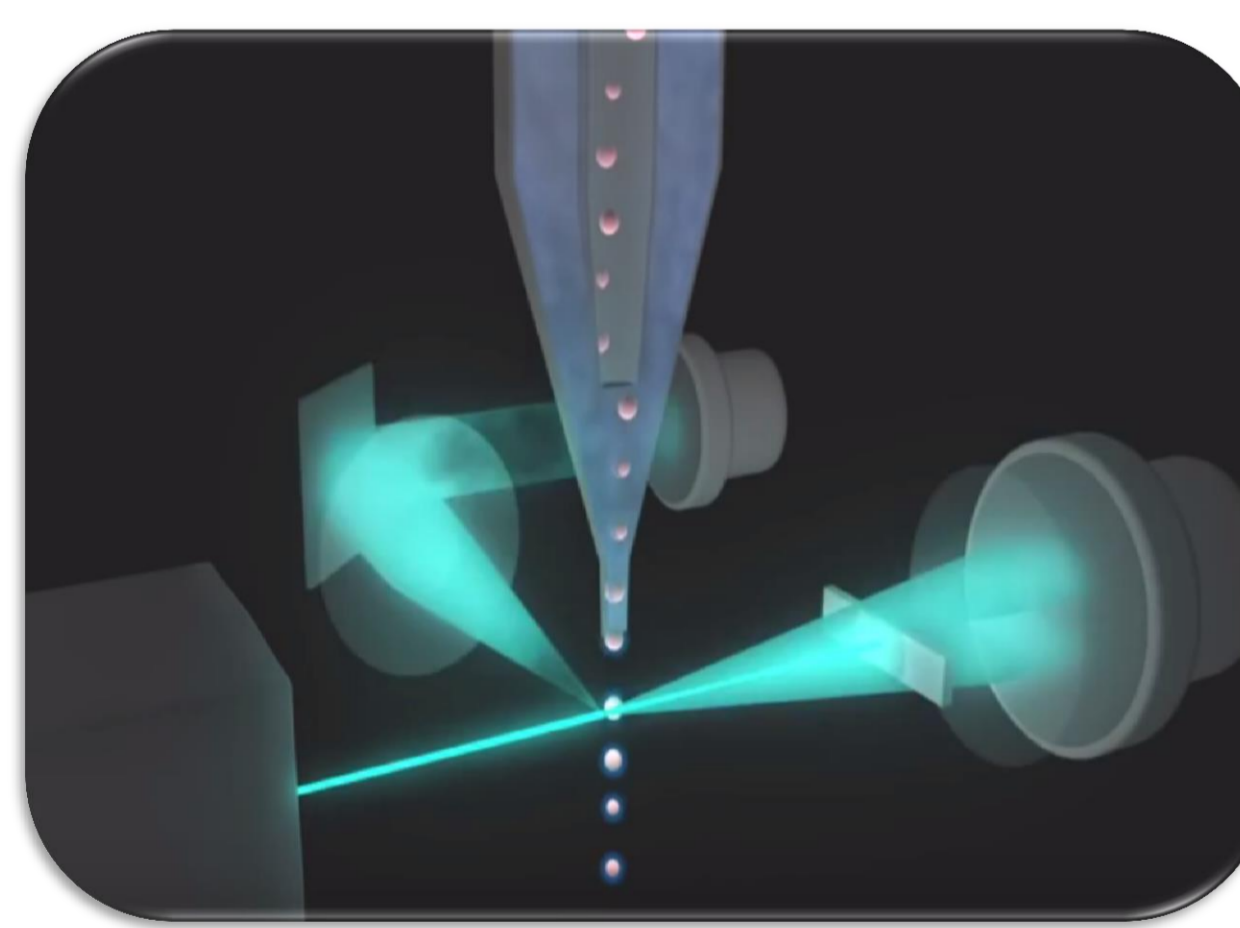
DCC measures macrophages, lymphocytes, and polymorphonuclear neutrophil cells (PMN) separately<sup>4,5</sup>.

- Made by light microscopy or flow cytometry, no standardized sample preparation or protocols, large discrepancies are present in the literature<sup>1-6</sup>
- Changes in leukocyte levels are detectable at low SCC levels<sup>2, 4</sup>



### Light microscopy

Manual differentiation of 100-200 cells by light microscopy. The cells are stained for easier differentiation, different staining protocols are used<sup>5, 6</sup>. Subjective and slow method.



### Flow cytometry

Laser based technology where one leukocyte at a time is passed through a laser beam. Scattered light is measured and correlates with cell size and granularity. In this manner, cell populations can be distinguished. Cells can also be stained by fluorescent antibodies targeted towards particular cell proteins<sup>3</sup>, or by DNA labeling<sup>2, 4</sup>. This is an objective, precise, and high throughput method that can count thousands of cells in one sample.

## Perspectives

The DCC provides a promising tool for early detection and monitoring of intramammary infections. As it unveils changes in the leukocyte composition already at low SCC levels, it has great potential as a complementary parameter for monitoring udder health.

1. Pilla et al., 2013, Differential cell count as an alternative method to diagnose dairy cow mastitis. J. Dairy Sci.
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3. Piepers et al., 2009, Technical note: Flow cytometric identification of bovine milk neutrophils and simultaneous quantification of their viability. J. Dairy Sci.
4. Dosogne et al., 2003, Differential Leukocyte Count Method for Bovine Low Somatic Cell Count Milk. J. Dairy Sci.
5. Pilla et al., 2012, Microscopic differential cell counting to identify inflammatory reactions in dairy cow quarter milk samples. J. Dairy Sci.
6. Schwarz et al., 2011, Microscopic differential cell counts in milk for the evaluation of inflammatory reactions in clinically healthy and subclinically infected bovine mammary glands. J. Dairy Res.

