

Brief communication

DETERMINATION OF MILK SOMATIC CELLS USING
FLUORESCENCE MICROSCOPY

(A preliminary report)

Milk somatic cell counts have up to the last few years been performed by the method of Prescott and Breed only. This method involves the preparation of a smear of 0.01 ml on 1 cm² of a microscopic slide, defatting, staining, and counting within an arbitrary fixed area of the smear using ordinary microscopy. Electronic counting methods involving Coulter counters represent a development of comparatively recent date. The principle of these methods involves the counting of all elements of the size of the somatic cells. Both methods are rather time-consuming.

Due to the extensive use of cell-counting on milk a more rapid but still accurate method would be of great value. Investigations of the principles of fluorochromation of cells in milk have led to the following method for the preparation of a milk sample for counting: One ml of milk and 2 drops of an aqueous solution of ACRIDINEORANGE (0.1 %) are mixed. Using a Breed syringe, 0.01 ml of the mixture is placed on a slide and covered with a cover glass (32 × 24 mm). The cover glass is heated slightly before application. The size of cover glass indicated gives a thickness of the preparation of 13 μ.

Counting is performed by fluorescence microscopy (Reichert Fluorpan) with a halogen lamp as source of light.

The thin layer of the preparation and an adequate combination of primary and secondary filters give a dark background on which the fluorescent cells appear as yellow-green luminous elements. At a magnification of 200 it is possible to distinguish the cell structure.

With this preparation method the same random distribution of the cells is obtained as by the Prescott and Breed method.

Comparisons between the above-mentioned principle of fluorochromation and the ordinary direct microscopic counting (Stain: Methylene blue) show the same average cell count in the milk samples. A regression coefficient of 1.01 is found between the results from the two methods on examination of 564 samples.

As outlined, an almost black background is achieved by the described technique. Investigations have revealed that the difference between the light intensity of the background and the cells is sufficiently great to affect a phototube. Accordingly it should be possible to utilize an optoelectronic counting method.

An optoelectronic counter is at present under development. For reasons of economy such components are chosen that the time needed to count one sample should be approximately 10 sec. whereby 300—400 determinations per hr. could be achieved.

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