

New Italian conversion line for the enumeration of total bacteria in raw milk with Bactoscan FC

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The total bacterial count at 30 °C is performed on raw milk as part of official controls (Reg. EC 853/2004). This analytical parameter is the better indicator of hygienic production and preservation of milk and, among the others, it is the basic tool for programming the raw milk heat treatment process, with obvious and direct impact on the final product quality.

Reg. EC 2074/2005 establishes that the reference method for the total bacterial count at 30 °C in raw milk is EN ISO 4833. In addition, it establishes that the use of alternative methods is acceptable when these are validated against the reference method in accordance with the protocol set out in EN/ISO standard 16140 or other similar internationally accepted protocols.

EN ISO 4833 counts the colonies grown in defined conditions after 72 h of incubation at 30 °C, whereas the flow cytometry instruments count, in about 10 min, presumably each cell independently from its physiological status or its capability to develop a colony in the conditions defined by EN ISO 4833. The count is obtained from electrical impulses that must then be converted into CFU/ml being this the regulatory unit of measure. Hence, the use of the alternative method is conditioned by the availability of a conversion relationship.

Unfortunately the use of different conversion lines strongly affects the reproducibility obtained by the instrumental method, impairing the accuracy of the results with major consequences both on economic and food safety aspects. However, due to the undisputable better repeatability, rapidity and cost effectiveness of the alternative method respect to the reference one, it is indispensable in the routine milk control.

The EURL-MMP has suggested for years that each member state has an its own single conversion line.

In Italy, a project for the elaboration of a single conversion line for Bactoscan FC was started in 2008 by National Reference Center BMQ of IZSLER with a group of 18 laboratories. A first conversion line was calculated as:

$$\text{Log (CFU/ml)} = \text{Log (IBC/}\mu\text{L)} \times 0.911 + 2.599$$

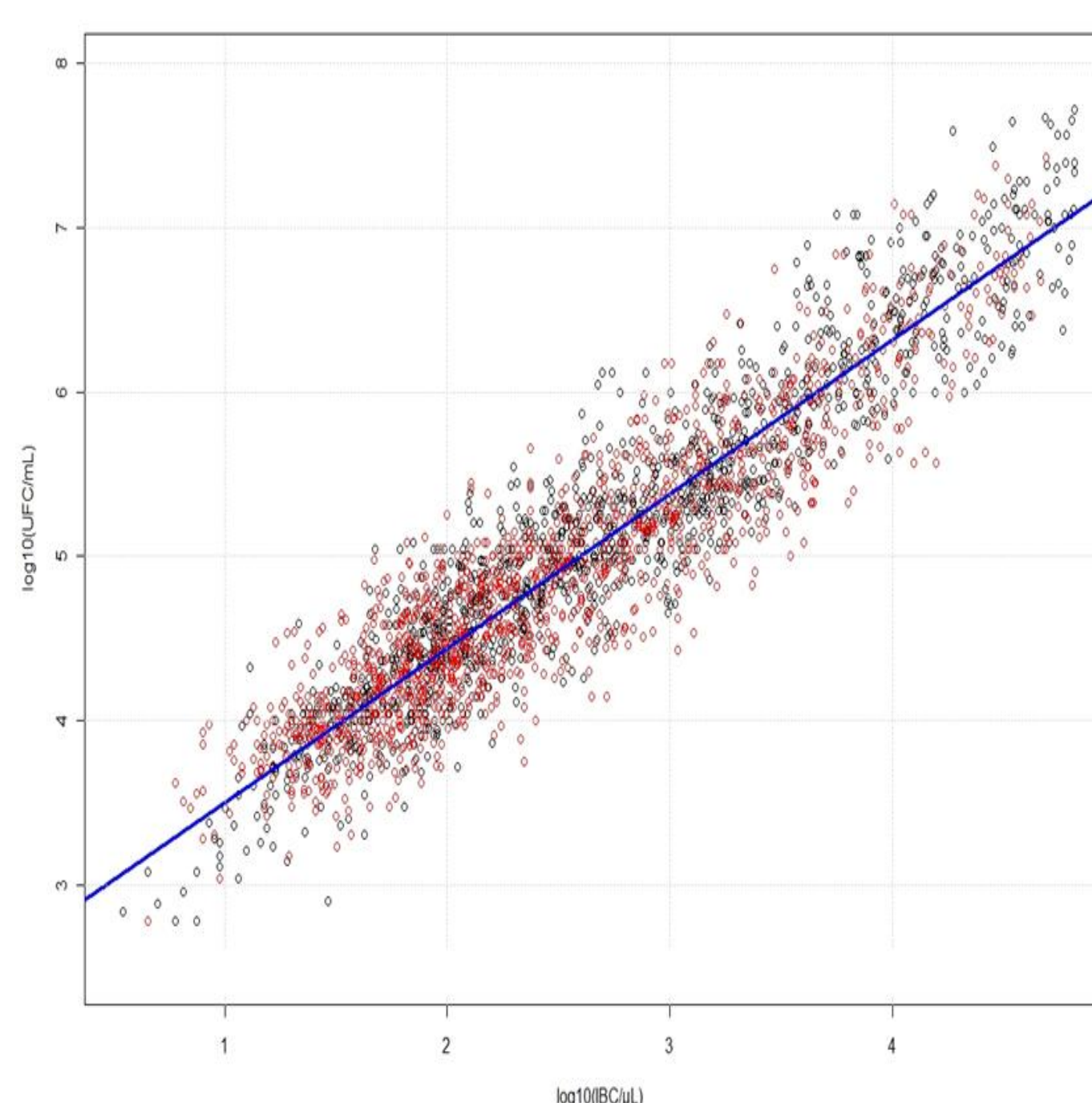
with Syx = 0.279 and an application range up to 50,000 IBC/ μ L.

This line was adopted on a voluntary basis by the laboratories participating to the initial study.

In 2012 a second round was started with the coordination of the NRL-MMP involving almost all the Italian laboratories using the instrumental method (private laboratories, breeder association network and IIZZSS) and leading to an updated conversion line based on a total of 2,732 valid samples from 29 labs, calculated as:

$$\text{Log (CFU/ml)} = \text{Log (IBC/}\mu\text{L)} \times 0.939 + 2.559$$

with Syx = 0.282 and an application range up to 70,000 IBC/ μ L.



data of 2008 round
data of 2012 round

The new conversion line, adopted in Italy on indication of NRL-MMP from 2013, gave a significant improvement of method reproducibility at national level. Such effects are regularly monitored through interlaboratory trials, questionnaires, data evaluations and periodic verifications of the conversion relationship.

