FOOD ANALYSIS

Serial Dilutions in Cost Effective Single-Use Sterile Bags

Microbiology testing for food and beverage safety and quality is mainly done by determining viable cell counts in homogenized and diluted samples. Instead of going through the labour intensive process of preparing test tubes for serial dilutions, users can now benefit from the sterile, single-use PE bags. Verifications by method comparison confirm the equivalence of the new method which relieves staff and increases the productivity of the testing laboratory.

Food can become contaminated at any point during slaughtering or harvesting, processing, storage, distribution, transportation and preparation. Lack of adequate food hygiene can lead to foodborne diseases and death of the consumer. Important information about the hygiene and quality of foodstuffs is the total number and types of viable cells, mainly bacteria, detected in food samples.

The standard plate count method for viable cell counting is applied for more than 100 years, dating back to the pioneering microbiologist Robert Koch (1843–1910). Samples to be analysed are diluted in ten-fold steps several times with a diluent solution (e.g. sterile saline). Aliquots of the dilution steps are then plated on appropriate culture medium, incubated for several hours to allow colony forming and counting for the calculation of the number of viable cells present in the original food sample. Although simple and reliable, the method is very time-consuming in its preparation and execution. Large amounts of test tubes have to be cleaned, filled, sealed and sterilized each time before they can be used. This consumes a lot of time, energy, water and space. The fast execution of the test tube method also requires manual skills and can cause in routine physical discomfort or pain known as repetitive strain injury (RSI).

Instead of going through the labour intensive process of preparing serial dilution tubes, laboratories can benefit from the many advantages of sterile, single-use polyethylene (PE) bags. The sample for dilution is simply added into the bags and sterile diluent is added by just pressing a button of the Serial Diluter. Thereby a perfect homogeneous mixing of the sample with the diluent is achieved. The manual mixing of each test tube with a mixer/vortexer is thus completely eliminated.

After finishing sample dilution and plating, the used bags are removed from the Serial Diluter and disposed *[Figure 1]*. Therefore, the preparation and examination of test tubes is no longer necessary.

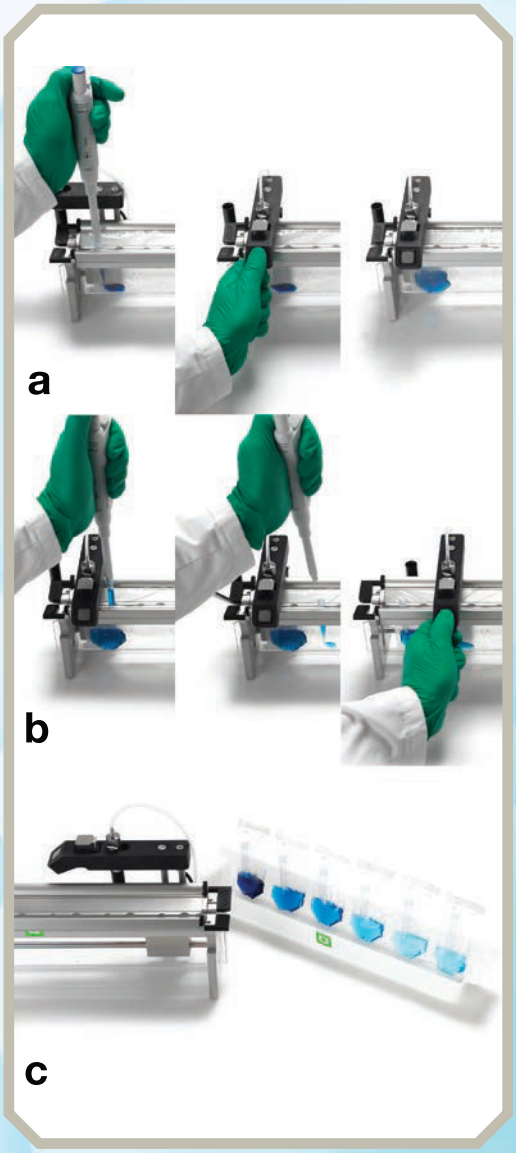
Product development and comparisons of the Inlabtec dilution method with the classical test tube method according to the international standard ISO 6887-1: 1992 were done in collaboration with the Zurich University of Applied Sciences (ZHAW), Institute of Food and Beverage Innovation supported by the Swiss Commission for Technology and Innovation (CTI).

For the verification of the dilution methods ZHAW scientists used overnight cultures of bacterial strains and determined the count of aerobic mesophilic bacteria in salad, milk and minced meat.

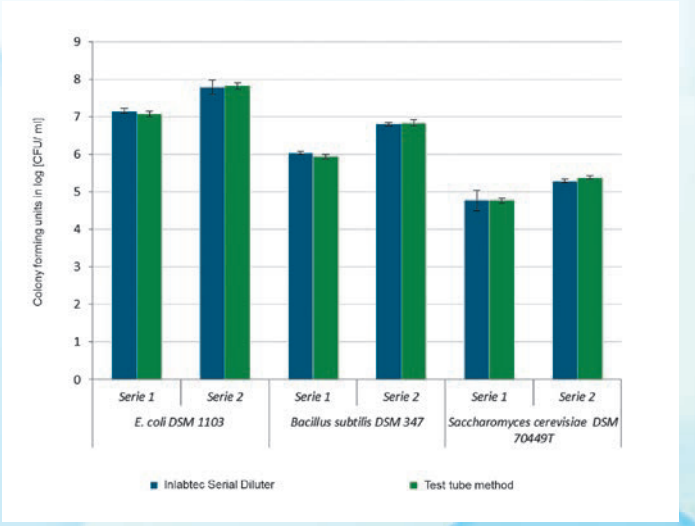
The results of replicated decimal dilutions of the cultivated strains by the classical test tube method and the Inlabtec method are shown in *Figure 2* and the results of the foodstuffs in *Figure 3*. The results by both methods are equal and within the standard measurement uncertainty of ±0.5 log10. Therefore, it can be concluded that the results obtained by using the Inlabtec Serial Diluter are equal to the results obtained by the standard test tube method.

Work in the labs of the ZHAW also confirmed that the entire dilution process is now much less labour-intensive (no more laborious cleaning of test tubes) and time-consuming. Also set up of the instrument is both simple and quick in addition to the Serial Diluters speed and reliability, ease of use and its ability to eliminate traditional sources of dilution errors.

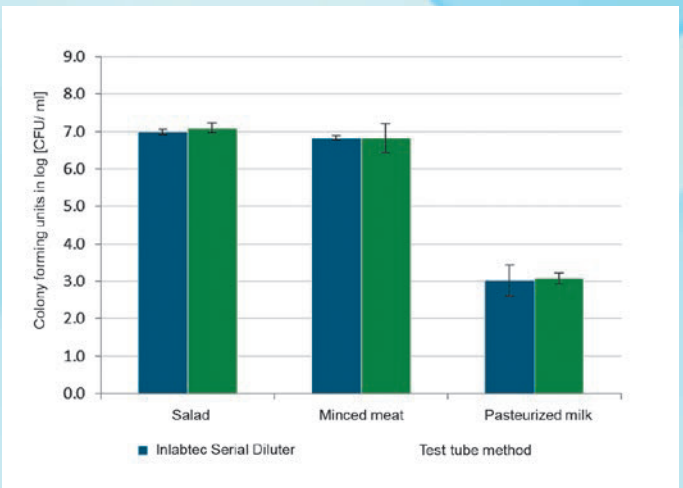
For detailed information about the Inlabtec Serial Dilution System please visit www.inlabtec.com.



01 Serial dilution in single-use sterile bags. A 1 ml sample together with the pipette is added into the first bag. The dosing arm is connected to the tip and the sample tenfold diluted by adding 9 ml diluent by pressing the button (a). Diluted sample is taken out for plating and for the next dilution step (b). After finishing the serial dilution and the plating of the diluted samples used bags are removed for disposal (c). Watch video under www.inlabtec.com



02 Test results with *E. coli* DSM 1103, B. subtilis subsp. spizizenii DSM 347 and S. cerevisiae DSM 70449T. Shown are the mean of colony forming units of three replicates per independent series. Results of the serial dilutions with the Inlabtec Serial Diluter in blue and with the test tube method in green. Chart from the ZHAW project report.



03 Total viable cell counts in samples of salad, minced meet and pasteurized milk determined by using the Inlabtec Serial Diluter (blue) and the test tube method (green). Shown are the mean of colony forming units of three replicates per independent series. Chart from the ZHAW project report.