Study determining the BactoScan conversion factor for the United Kingdom

Area of research interest: Foodborne pathogens
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Three commercial laboratories using BactoScan equipment, and accredited to ISO17025 for their use, undertook duplicate analysis of 1,800 routine samples of raw milk from farms across the UK to determine IBC values.

TVC results, as log10 values, gave the equation: Log10 (TVC) = 0.9151x Log10 (IBC) – 0.5696 (r^2 =0.6694).

Background

In EU legislation referring to the microbiological standard of raw bovine milk the units ‘colony forming units (cfu)’ are cited when describing numbers of bacteria, as enumeration is based on conventional microbiological plate counting (Regulation (EC) No 853/2004 Annex III, Section IX). Such plate counts yield a total viable count (TVC) of bacteria which is normally expressed as cfu/cm3.

Therefore, there is a need for a conversion factor to convert IBC to TVC, to demonstrate that legislative standards are being met. The UK lacks such a conversion factor, which is a requirement of EC legislation (Regulation (EC) No 2073/2005), and this was noted in the recommendations of the EU report ‘Final Report of An Audit carried out in the United Kingdom from 08 To 19 April 2013’ ref. DG(SANCO)/2013-6872).

Approach

It was essential to use the equipment in commercial premises, which process high numbers of samples, in order to obtain valid results. Routine samples of raw milk were analysed in duplicate by the BactoScan, and using conventional plate counting as described in BS EN ISO 4833:2003 (Microbiology of food and animal feeding stuffs -- Horizontal method for the enumeration of microorganisms -- Colony-count technique at 30 degrees C) (Anon. 2003). All three laboratories had ISO 17025 accreditation for the determi¬nation of IBC, hence they participated in external quality assurance (EQA) programmes, and the results of these were provided to AFBI to verify their competence.

Overall, 1,799 valid results were obtained and statistical analysis of these was undertaken, Simple linear regression analysis of the data, as Log10 values, was undertaken and the conversion equation was determined as (standard error in brackets): The mean value of the duplicates was then analysed, using regression analysis, to give the conversion factor. Overall, 1,799 samples were analysed and regression analysis of the dataset gave the equation: Log10

(TVC) = 0.9151x Log10 (IBC) – 0.5696 (r2 =0.6694).

This equation constitutes the conversion factor for the UK. This result was compared to the results of a pan-EU study undertaken by the European Union Reference Laboratory (Guillier et al, 2016), and was seen to fall within the confidence interval (95%) of the EU harmonised conversion equation, supporting its validity. All statistical analyses were undertaken by the Biometrics & Informa¬tion Systems Branch, AFBI, using Genstat Release 18.1 for Windows (VSN International Ltd, Hemel Hempstead, HP2 4TP, UK). Note that enumeration results were transformed to Log10 to meet the require¬ments of BS ISO 21187:2004 [9] for data analysis.

Raw milk is legally required to have a TVC (30°C) value less than 100,000 cfu/mL, as defined in Regulation (EC) No 853/2004 of the European Parliament [5] and of the Council laying down specific rules on the hygiene of foodstuffs.

Results

Three commercial laboratories using BactoScan equipment, and accredited to ISO17025 for their use, undertook duplicate analysis of 1,800 routine samples of raw milk from farms across the UK to determine IBC values. In ad¬dition duplicate bacterial enumeration of all samples, using BS EN ISO 4833:2003, was undertaken to determine TVCs.

Overall, 1,799 valid results were obtained and regression analysis of the IBC against the TVC results, as log10 values, gave the equation: Log10 (TVC) = 0.9151x Log10 (IBC) – 0.5696 (r^2 =0.6694).

Raw milk samples (n = 1,799) were analysed in three laboratories locat¬ed in Northern Ireland, England and Scotland and the bacterial count at 30°C determined using conventional microbiology and flow cytometry, the latter using BactoScan equipment.

The resulting data was analysed to determine a BactoScan conversion factor. The BactoScan conversion therefore been adopted to provide automated routine microbiological analyses. In the UK flow cytometry is used to determine the bacterial quality of almost all commercially produced raw milks, and this takes place in three laboratories: one in England, one in Scotland and one in Northern Ireland.

All of these laboratories use BactoScan equipment (Foss UK Ltd, Birchwood, Warrington WA3 6AE, UK), and the laboratories all have enumeration procedures accredited to ISO17025.

The EU Food and Veterinary Office (FVO) requested that a UK conversion factor for the calculation of TVC results from IBC vales was determined and the Food Standards Agency of the UK commissioned the UK National Reference Laboratory for Milk and Milk Products (NRLMMP), based in Food Microbiology Branch, the Agri-Food and Biosciences Institute (AFBI), to obtain this conversion factor.

Total viable counts (TVC) of bacteria in raw milk at 30°C were determined using conventional plate counts as described in BS EN ISO 4833:2003 and individual bacterial counts (IBC) using BactoScan equipment, as recommended by the manufacturers. Samples of raw milk submitted for routine determination of individual factor for the United Kingdom of Great Britain and Northern Ireland was determined to be: Log10 (TVC) = 0.9151 Log10 (IBC) - 0.5696.

This determination meets the requirement of EU Food and Veterinary Office (FVO) for the UK to determine this conversion factor, and there¬fore commercially determined IBC values can be converted to legally required TVC values using this factor.

Research report
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