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THE USE OF AUTOTRAK FOR THE RAPID ENUMERATION OF MICRO-ORGANISMS ON MEAT SURFACES

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SUMMARY

The ability of AUTOTRAK to count the number of micro-organisms on the surfaces of meat carcases was compared with that of the total viable count (TVC) obtained using the standard pour plate method. Cultural counts were carried out after 1,3 and 7 days incubation. Regression values were 0.710, 0.820 and 0.840 respectively against AUTOTRAK on the 110 samples examined.

It was concluded that AUTOTRAK is a successful method for the rapid non-destuctive assessment of the microbial quality of meat surfaces.

INTRODUCTION

Considerable variability in the organoleptic and keeping quality of meat has always been apparent to the consumer. Microbiological standards for fresh meat have been proposed but great controversy remains regarding specifications. The sampling of fresh meat for microbiological analysis provides problems as there is no standard accepted procedure. Rapid non-cultural assessment of the microbial load on meat is a priority.

The sampling of meat surfaces may be performed in two ways:

a) non-destructive
b) destructive

a) Non-destructive Methods

i) Swab/rinse method. This is the most widely used technique whereby a sterile swab is applied to a unit area of surface. The swab is then broken into sterile diluent agitated and plated using an appropriate culture media. A modification of this method comprises the Wet and Dry swab technique which involves swabbing unit area with a moistened swab and then with a dry swab. This results in a higher recovery of contaminant micro-organisms.

Alginate swabs have been used in place of cotton swabs since they will dissolve in 1% sodium hexametaphosphate releasing entrapped micro-organisms and thereby improving recovery.

ii) Rinse method. The sample of meat is immersed in a sterile fluid or the fluid is brought into contact with the surface being examined. Clark (1965) introduced a pressure spray device based on the rinse method. This method is independant of the texture of the surface.

iii) Agar contact method. Sterile agar is pressed on to the surface, removed and incubated (Gabis and Silliker, 1975). This method is unsuitable if the surface of the meat is uneven as in the case of carcases, or if the surface is contaminated with spreading bacteria.

iv) Direct surface agar plating method. Sterile melted agar is poured on to the surface to be sampled and left to solidi under a sterile cover. After incubation colonies at the interface are counted (Angelloti and Foter, 1958).

b) Destructive Methods

This method of sampling is not popular w the meat industry since it reduces the retail value of the carcase or joint but does achieve a higher recovery than non-destructive methods.

i) Blender method. A specific weight or is asceptically removed and macerated us a mechanical blender. This is then suspen in diluent, mixed and filtered (Baldock, 1

ii) Cork borer method. A cylinder of mea removed using a sterile cork borer, macer and suspended in diluent as in the blend method (Haines, 1931).

iii) Tissue removal method. A stainless steel plate with an oval hole in the cen is pressed on to the sample of meat and projected area removed and mecerated (Yokawa and zulzke, 1975).

However, it is recognised that with the most efficient of techniques only a proportion of the micro-organisms will b recovered. It is impossible to wash all attached micro-organisms into the diluen Therefore, any method of assessment of microbial load can only produce results good as the sampling method employed.

AUTOTRAK, a rapid automated system for t enumeration of micro-organisms in the li menstruua developed by the Centre for Bio-Medical Instrumentation, University o Strathclyde, Scotland, is a microprocesso controlled epifluorescence microscope sy Samples are taken up automatically by a probe and applied to a specially treated moving tape prior to fixation and staine a fluorochrome. The specimen passes unde microscope objective where the fluoresce signals emitted by the micro-organisms a detected by a photomultiplier. The resul are processed and can be stored by the integral computer and hard copy printed Results are expressed as the number of organisms per millilitre of original spe

AUTOTRAK has a throughput of up to 120 samples per hour and produces a result w 60 seconds of uptake of specimen.

MATERIALS AND METHODS

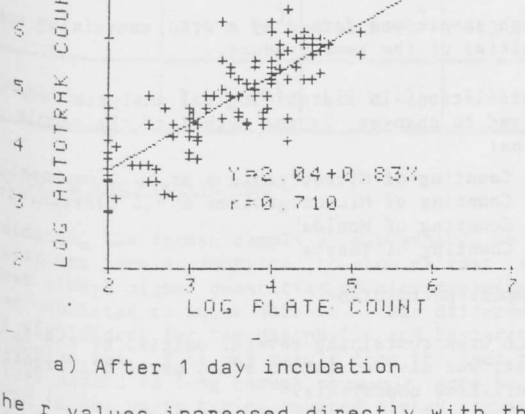
The skin surface of 110 samples of beef flank was sampled using the swab/rinse method. Disposible sterile cotton wool s (Exogen Ltd., Clydebank, Scotland) were us to swab an area of 4cm^2. The swab was th transferred to 10ml of ¼ strength Ringer solution (Oxoid Ltd., Basingstoke, England The sample was vortex mixed for 1 minute ambient temperature. A 1ml aliquot was removed and serial dilutions carried out Ringer solution to cover the range from 0 to 10E6 organisms/ml. Pour plates were prepared using Plate Count Agar (oxoid and incubated at 30°c for up to 7 days.

Counts were carried out of the number of colonies appearing after 1,3 and 7 days incubation.

2ml aliquots of the sample were dispensed into an analyser cup and analysed using the AUTOTRAK system.

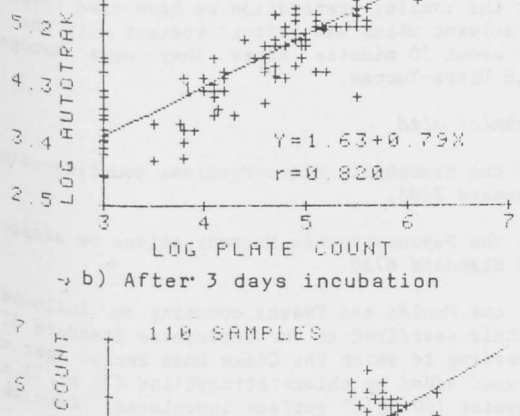
RESULTS

The results from the cultural method following 7 days incubation were plotted against the AUTOTRAK results in logarithmic form and linear regression calculated

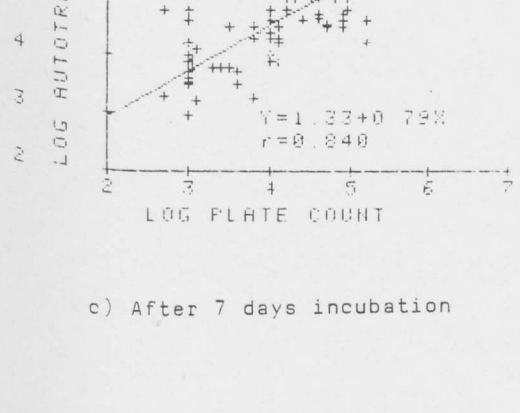


a) After 1 day incubation

The r values increased directly with the number of days incubation. 1,3 and 7 days incubation gave r values of 0.71,0.82 and 0.84 respectively, AUTOTRAK correlating best with an incubation period of 7 days.



b) After 3 days incubation



c) After 7 days incubation

DISCUSSION

The basic principle of AUTOTRAK involves the use of Direct Fluorescence Microscopy (D.F.M.). Using the developed protocol the fluorochrome will stain organisms which may not be recovered by the cultural procedure. As can be seen from the correlation values, AUTOTRAK results best correlate with the cultural counts obtained following 7 days incubation. Therefore, AUTOTRAK eliminates the delay normally required for quantitative indication of the microbiological status of a meat sample.

Any automated method must fulfill certain criteria. These include:-
a)low overall cost per test
b)ease of use
c)high throughput of samples
d)minimum amount of pretreatment
e)rapid reporting of results

Using AUTOTRAK the approximate cost per sample for meat surface evaluation would be 10p,compared with cultural methods where the costs exceed 50p per sample.

An automatic sampler integral to AUTOTRAK allows batches of 50 samples to be analysed on a "load and go" basis. AUTOTRAK has applications for the microbiological screening of medical, food and industrial specimens and represents the first of a new generation of "real time" automated microbial screening systems.

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