

# A point-of-issue system for detecting platelet components contaminated with bacteria

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## Introduction

Platelet (PLT) transfusion-associated bacterial sepsis has remained a substantial patient risk, primarily due to the lack of effective and point-of-issue measures to detect bacterial contamination. Our previous researches using a research type near infrared (NIR) instrument provided information indicating that NIR spectroscopy could detect the number of microorganisms non-invasively in various products, including solutions obtained from cabbage washing, raw milk and PLT components kept in PLT bags. To examine the potential of NIRS in more practical viewpoint, this work evaluated its ability in terms of detection of various bacterial species, and the use of a low-cost portable machine for bedside monitoring.

## Materials and methods

Two series of experiments were conducted. The first series was to collect data from platelets infected with *Klebsiella oxytoca* and *Pseudomonas aeruginosa* with control platelets. The second series was for platelets infected with *Staphylococcus aureus* and *Enterobacter cloacae*. For both series, three apheresis platelets were pooled to allow uniformity among those used as infected and as control samples. NIR spectra were collected using the FQA-NIR Gun (Shizuoka Shibuya Seiki, Hamamatsu, Japan) equipped with a custom-made platelet bag holder for transmittance measurement (Figure 1).

The samples were kept in a platelet storage unit with constant swirling, and temperature control at 24°C. Prior to NIR measurements, samples were kept in a 24°C water bath covered with polyethylene sheet for at least 15 minutes. Spectral acquisitions were performed at 12-hour intervals until the total storage time reached five days, giving a total of 11 measuring points for each sample. Some samples were removed from the calibration due to the natural extinction of bacteria, or because of obvious infected characteristics that were noticed. Classifications were



**Figure 1.** NIR measurement of platelet component using a portable NIR instrument.

performed in such a way as to imitate the actual situation, where the species of bacterial infected could not be identified in advance. The SIMCA analysis on the difference spectra of each sample was calculated by subtracting second derivative spectra measured at  $i$  hours by second derivative spectra of the same sample measured at 0 hour.

## Results and discussion

First, a PCA model was developed from control samples. The detection of infected platelets was performed by using the membership plots: the plots between leverage and distance of that sample from the control model. It was found that the number of days that our system could segregate infected platelets from the control platelets differed, depending on the growth rate of bacteria infected. The membership plots could identify platelets infected with the fast growth bacteria, *S. aureus* and *K. Oxytoca* soon after inoculation; all infected samples were identified within 72 hours after inoculation, due to the high number of bacteria that accumulated since 48 hours after inoculation (Figure 2).

For the slow growing bacteria, our system could detect the contamination only after 4 or 5 days of storage, corresponding to the rises of their population in platelet bags that dramatically increased to the vital level of  $10^7$  CFU  $\text{mL}^{-1}$  at Day 5 and at Day 4 for *E. Cloacae* and *S. aeruginosa*, respectively.

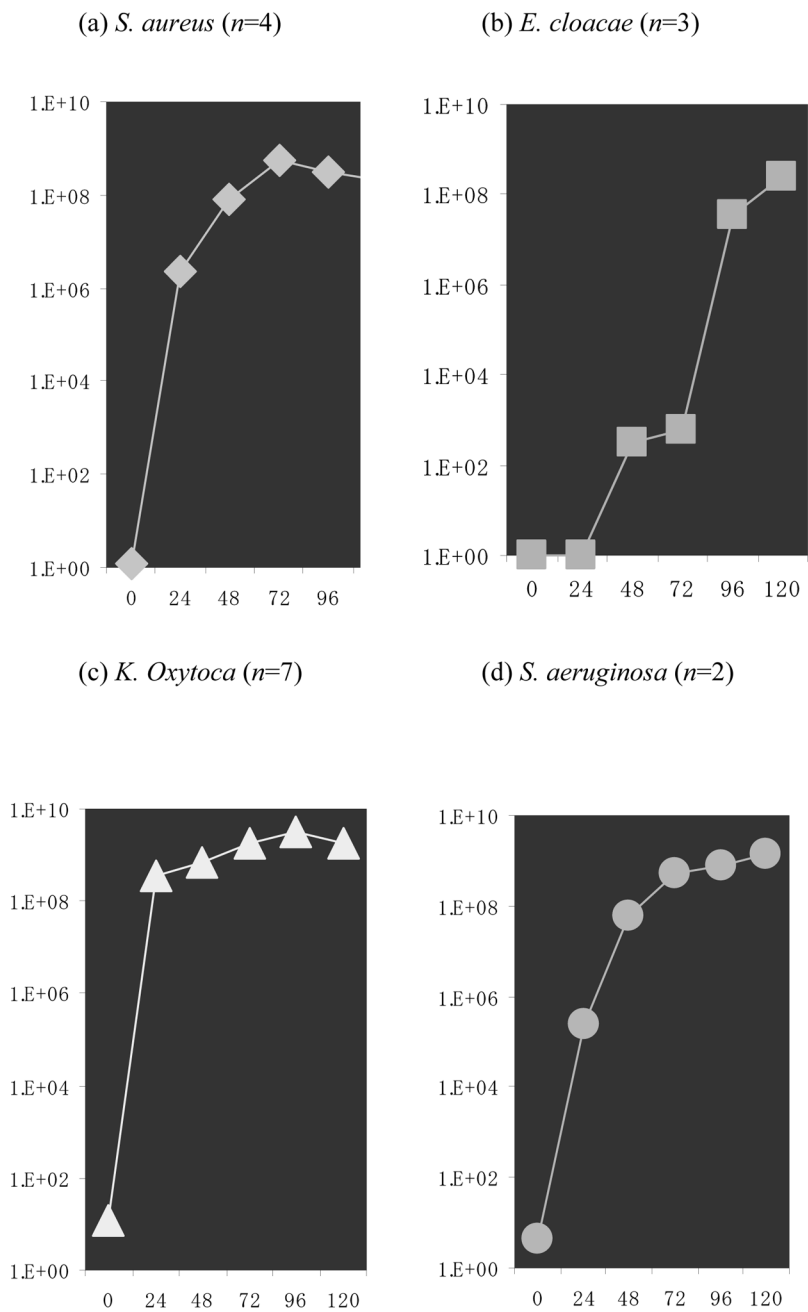


Figure 2. Growth rate in platelet bags of the 4 bacteria examined.