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## **GROWTH CURVES AND CORRELATION OF OD600 TO COLONY FORMING UNITS PER MILLILITER FOR SEVENTEEN MICROBES MOST COMMONLY KNOWN TO CONTAMINATE PLATELETS**

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### **Background:**

Bacterial contamination of blood components is currently the most frequent infectious complication of blood component use, with an estimated incidence of 1000 to 3000 platelet units, which is associated with 100-150 deaths per year, and 1 in 30,000 RBC units in the U.S. The risk of receiving a bacterial-contaminated blood product is estimated to be 50- to 250- fold higher than the combined risks of transfusion-mediated infectious of HIV 1/2, HCV, HBV and HTLV VII. Due to the risk of bacterial contamination, the shelf life of platelets is limited to five days in the U.S. In 2003, the AABB recommended to implement a bacteria detection test for platelet components by March 2004. To develop a test for bacterial detection in platelets, platelet samples are usually artificially spiked with known amounts of bacteria as the study materials to model the contaminated platelets. In order to better control the desired amounts of each species of bacteria spiked into the modeled platelet samples, we need to have a very accurate method to predict and calculate the microbial loads for each species prior to spiking into platelet samples. In this study we investigated and established methods to accurately predict the correlation of optical density at 600nm (OD600) to the actual plate counts, colony forming units per milliliter (CFU/ml), for each of the 16 Gram positive, Gram negative bacteria and the yeast *Candida albicans* that are most prevalent to contaminated platelets.

### **Materials & Methods:**

Bacterial strains were obtained from the Chiron Master Culture Collection or the ATCC. Culture media was prepared using DIFCO reagents. OD600 measurements were made using an Eppendorf Bio Photometer. Cells were grown at their optimal growth conditions. Cultures were sampled at time intervals consistent with their doubling times, and OD600 was measured. Immediately following OD600 measurements, samples were diluted and plated on sheep blood agar plates to obtain countable colonies. The data were

plotted as OD600 verses time for the growth curves, and OD 600 verses colony counts for OD600 to CFU/ml curves.

### ***Results & Conclusion:***

Among the 17 microbes investigated, the correlation of OD600 to the actual plate counts (CFU/ml) for some microbial species are significantly different. Using the method we developed, we are able to consistently predict plate counts (CFU/ml) within 1-2 fold variations from OD600 measurements for each of the microbes studied.

1. Hillyer, C., et al. Bacterial Contamination of Blood Components: Risks, Strategies, and Regulations. Hematology 2003. American Society of Hematology.