

bacteria since also fungi can be demonstrated as exemplified applying the yeast fungus *Candida albicans*.

P-693

TIME COURSE OF pH IN PLATELET CONCENTRATES AFTER BACTERIAL CONTAMINATION

Montag T¹, Nicol S-B¹, Schurig U¹, Brachert J¹, Sauer A¹, Hermanns J¹, Sicker U¹, Muller TH², Schneider CK¹

¹Paul-Ehrlich Institute, Langen, Germany ²German Red Cross Blood Transfusion Service, NSTOB, Springe, Germany

Background: Among the methods for Platelet Bacteria Screening, lowering of pH in Platelet Concentrates (PC) is considered as one indicator for bacterial contamination. There are long lasting critical discussions regarding the sensitivity of this approach (e.g. Wagner and Robinette, 1996). Recently, the BCSI pH1000 system has been developed allowing non-invasive pH measurement(s) in PCs at any point in time during storage until the moment of transfusion.

Aims: The novel tool cited above has been used in the current study for precise characterisation of pH development in PCs after defined artificial bacterial contamination.

Methods: The following PEI Bacteria Standards (bacteria strains especially characterized regarding their growth properties in PC) were used: PEI-B-20-05 (*Streptococcus pyogenes*), PEI-B-23-06 (*Staphylococcus aureus*), PEI-B-06-07 (*Staphylococcus epidermidis*), PEI-B-07-22 (*Bacillus cereus*), PEI-B-19-05 (*Escherichia coli*), PEI-B-08-09 (*Klebsiella pneumoniae*). Additionally, the yeast fungus *Candida albicans* (PEI-B-21-03) was applied. All together, 35 PCs were contaminated (5 each for the respective microbial species) with 0.03 CFU (Colony Forming Units) per mL (10 CFU per platelet bag). Sterile PCs served as controls. The PCs were stored under usual blood bank conditions; bacteria count and pH were monitored over up to 8 days in parallel.

Results: As expected, the pH in contaminated PCs started to decrease after the bacteria had reached a count of 10exp6 to 10exp7 followed by a further lowering down to 6.7 to 6.5. Thereafter, a re-increase of pH values could be observed in all contaminated PCs. In several cases, the pH returned to its initial value three days after contamination and increased further up to 7.3–7.4 on day 7.

Conclusions: Generally, pH measurement in PCs is not acceptable to be a solitary approach for Platelet Bacteria Screening because of its low sensitivity. On the other hand, there are time points at which the pH provides a warning signal to prevent transfusion of bacterially contaminated PCs. Additionally, the BCSI pH 1000 System allows pH measuring immediately before transfusion without opening the bag which gives a clear effort in comparison with pH estimation by strips or dip sticks (as recommended, for instance, by AABB in 2004). There is a need for further studies regarding pH changes in bacterially contaminated PCs.

P-694

RELIABLE RAPID STERILITY TESTING OF CELL THERAPEUTICS BY COMBINATION OF SHORT PRE-CULTURING AND BACTERIA DETECTION USING FLOW CYTOMETRY

Montag T, Nicol S-B, Schurig U, Brachert J, Sauer A, Friedrich AF, Sicker U, Schneider CK

Paul-Ehrlich Institute, Langen, Germany

Background: In principle, Cell Based Medicinal Products (e.g. stem cells, monocyte derived dendritic cell tumour vaccines) have to be tested for sterility following pharmacopoeial regulations. In the classical test, each sample must be exposed, in parallel, to aerobic and anaerobic incubation conditions, in liquid media, for a period of at least 14 days. The main disadvantage of the pharmacopoeial sterility test is the long incubation time of 14 days up to result. This is owing to the parameter for bacterial growth in classical microbiological cultivation, i.e. examination of turbidity which can be observed after the bacteria have reached a count of around 10⁸ CFU/mL. On the other hand, cell therapeutics show, in many

cases, a very short shelf life from cell harvest until administration to the patient; often <2 days. The sterility test can, therefore, be significantly improved if each contaminant can be identified in <14 days with a more sensitive detection method that does not rely upon high bacterial cell density for visualisation.

Aims: In this paper, it is demonstrated that the incubation time necessary for reliable bacteria detection can be significantly reduced if macroscopic observation of turbidity is replaced by bacteria detection using flow cytometry. All together, 12 bacterial species and two fungal species were involved in the study.

Results: The vast majority (13/14 micro-organisms) could be detected within 24 hours; all of them were detected within 42 hours. The exception concerns the mould fungus *Aspergillus niger* which was detectable after 42 hours. The experiments intentionally included the six microbial strains which are recommended in the international pharmacopoeias (e.g. European Pharmacopoeia 2.6.1. Sterility) for Growth Promotion Test and Validation Test in sterility testing. All of them could be detected within 42 hours at the latest.

Conclusions: The results demonstrated in this paper should be seen as a pilot study on feasibility on the novel Rapid Sterility Testing. There is a need for further evaluation of the principle. On the other hand, the method should be rapidly implemented into sterility testing of cell based medicinal products since there is no reliable approach which considers their extraordinary circumstances.

P-695

SENSE AND NON-SENSE IN PLATELET BACTERIA SCREENING

Montag T

Paul-Ehrlich Institute, Langen, Germany

Since the impressive reduction of transfusion-transmitted virus infections, bacterial infections by blood transfusion are representing the most important infection risk. The current focus of attention are platelet concentrates as they are stored under temperature conditions which allow growth of contaminating bacteria up to 10¹⁰ and more microbes per platelet bag. This paper does not consider the Pathogen Reduction Methods but will assess suitable Screening Methods: Beside conventional microbiological approaches or surrogate markers, several efficient methods able to detect bacterial contamination in platelets are available on the market. They have to be divided into two different methodological principles: The cultivation methods and the rapid methods. Cultivation or incubation methods require some time for signal production as they depend on growth of microbes. Thus, they have to be combined with early sampling, i.e. the sample to be examined has to be drawn from the blood component 1 day after donation. Their advantage is the relatively uncomplicated implementation into the logistics of blood banks. Because of the initially very low count of bacteria after donation, there remains a certain small sampling error in application of that strategy. Rapid methods are able to produce the diagnosis within a short time. Therefore, they allow postponing of sample drawing, ideally up to the time immediately before transfusion. However, that procedure causes logistical complications. On the other hand, late sampling combined with a rapid method will prevent the transfusion of highly contaminated platelet concentrates leading to acute septic shock up to the death of the patient. Considering the sum of different aspects including the supply of patients, the potential improvement of microbial safety of platelet concentrates is comparable in both strategies.

P-696

EVALUATION OF CURRENT TOOLS FOR ENDOGENOUS INFECTIVITY REMOVAL FROM BLOOD PRODUCT IN A SHEEP TSE EXPERIMENTAL MODEL

Coste J¹, Segarra C¹, Grassi J¹, Andreoletti O²

¹Etablissement Français du Sang de Pyrénées-Méditerranée, Montpellier, France ²ENVT, Toulouse, France

Background: Since the recent identification of three vCJD cases who received a blood transfusion (use of Red Blood Cell Concentrate) from