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Blood Bank

Blood and Beyond

Frequency of False-negative Cultures in Screening of Platelet Concentrates for Bacterial Contamination

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INTRODUCTION

- Bacterial screening in NL since 2001
  - 500,000 units 5BC-PC tested (+ 50,000 Apheresis PC)
  - Started with 0.9% initial positive
  - After implementation pre-donation sample:
    - 0.44 % initial positive
  - Since pre-donation: 4 reported serious transfusion reactions with BacT/Alert negative TC
    - Imputability
    - Severity

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INTRODUCTION

- International discussion on false-negatives
  - **PASSPORT study in US**
  - **data ARC**
  - **Irish and Welsh “outdated” studies**
- In these studies
  - **False-negative rate similar or higher than initial positive**
- Sanquin relatively high initial rate
  - **Fear for high false-negative rate**
  - **Not to be expected based on clinical data**

INTRODUCTION

- Sanquin started also “Outdated” study
  - Outdated units (>7/5 days) to Amsterdam (R&D unit)
  - Re-cultured in BacT/Alert
  - Aerobic and anaerobic bottle, 7.5 ml/bottle
  - 1 - 5 days (mean 2) after expiration

Study Rules

- Check for contamination:
  - **Per day of inoculation 5 saline samples included**
- Positive in “outdated” culture
  - **Initial culture negative**
  - **Repeat “outdated” culture also positive**
  - **Both bottles positive (if applicable)**
  - **Organism derived from positive culture bottles and bag should be the same**
- At least 4000 5BC-PC to be included

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Results

- 4021 5BC-PC were tested
- 4 real positives (2 plasma, 2 PAS-II):
  - **Both bottles positive, as well as repeated cultures**
  - **Same microorganism from bottles and bags**
  - **All Staphylococcus Coagulase Negative (*S.epi*)**
  - **Rapidly positive: 5-10 h, at least 10^6 CFU/ml (PCR)**
- 1 indeterminate:
  - **One bottle positive (20 h), repeats and bag positive**
    - **Staphylococcus Haemolyticus**
  - **No signs of lower platelet quality**
  - **Very low contamination at day 9**

Results

#	Product	Age of PC	Sequencing results	TTP	AFLP typing	Ct	Bacterial concentration (PCR)
1	PC in plasma	8 days	*S. epidermidis*	(I) 4.1h (II) 4.3h	Identical isolates	17.78	> 10^7
2	PC in plasma	8 days	*S. epidermidis*	(I) 4.7h (II) 8.2h	Identical isolates	12.24	> 10^8
3	PC in PAS II	7 days	*S. epidermidis*	(I) 11h (II) 10.6h	Identical isolates	18..83	> 10^6
4	PC in plasma	9 days	*S. haemolyticus*	(I) 20.4h (II) 5.5h	Identical isolates	NA	Not tested in PCR
5	PC in PAS II	7 days	*S. hominis*	(I) 4.3h (II) 5.8h	Identical isolates	17.85	> 10^7

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Indeterminate: at day 9 ~ 1 CFU/ml

Results

35 false positives

- **Only one bottle positive and/or late**
- **Repeat cultures negative**
- **No bug isolated from positive bottles, except 2/35**

- 2 positives, appeared to be screenings positive
- Controls: about 1700, 5 false positives (early phase)
  - False positive rate (no bug isolated from positive bottle)
    - Controls: 1/400 bottles – 1/2000 bottles
    - PC screening 1/2400 bottles
    - PC outdated: 1/480 bottles

Results

with 4021 5BC-PC tested

- 4 false negatives:
  - **4/4021: 95% CI 0.03 – 0.25 (0.10%)**
- Frequency similar to those reported sofar
  - **Murphy (Ireland): 7/8282 = 0.08% (mainly BC-PC)**
  - **Dumont (PASSPORT): 4/6039 = 0.07% (apheresis)**
  - **Pearce (Welsh): 6/6438 = 0.09% (mainly BC-PC)**

Results

- Also outdated apheresis PC were tested
  - **In total > 1700 different units,**
    - **261 full units**
    - **1449 units spliced for pediatric use**
  - **1-8 aliquots per unit (mean 3; high sensitivity)**
- 1 positive sample
  - **Split product, 4 aliquots, 3 tested**
  - **For 1 split: anaerobic bottle repeatedly positive**
  - **3 different Coagulase Negative Staphylococci from bottles and unit**
  - **Difficult interpretation; not a fals negative**

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Discussion

- For the Netherlands:

  Frequency of false-negatives: ~ 0.10 %
- Frequency similar to other studies, not related to initial rate of positives

Discussion

- False-negatives detected: serious contamination, but in NL not reflected in clinical problems
  - **About 20% transfused after 5 days: 10,000/yr**
  - **Expected: 10 cases/yr**
  - **Haemovigilance: two cases reported last 3 years**
  - **During storage below critical level for patient harm?**
  - **Reports of CNS containing TC:**

    10^5 – 10^8 CFU/ml tolerated in most cases

    Jacobs et al. Clin.Infect.Dis. 2008;46:1214

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Possible consequences

- Reduction of shelf life
- Additional test at release if longer stored
  - PCR would be sensitive enough
  - Fast enough?
- Additional test at day 4/5 if longer stored
  - BacT/Alert culture, one bottle?
  - Negative to date?
- All skin flora (CNS)
  - More attention for skin desinfection?
- Pathogen reduction method
  - Pro’s and con’s

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