Bacterial Contamination of Donor Blood and Blood Components from a Tertiary Care Hospital in North India

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ABSTRACT

Screening of blood donors for viral pathogens has greatly improved the safety of donated blood. However, transfusion associated bacterial sepsis, remains an important public health concern, which has received very little attention. Therefore this study was carried out to determine the prevalence of bacterial contamination in donor blood and blood products, to find the commonly contaminated blood product and to identify the microorganisms involved. The present study was conducted on 136 random blood samples received in the Department of Microbiology, GMC, Jammu for a period of 1 year i.e. April 2017-2018. Bacteria were identified using standard bacteriological and biochemical methods. The overall prevalence rate was 12.50% (Packed cells, 21.21%; Platelets, 10.41%; Whole blood 9.09%). The most commonly isolated bacteria were Klebsiella sp, Staph. aureus and CONS. Most of the contaminated samples had 3-7 days of storage time. Maximum number of contaminated samples was from Blood Bank, GMC, Jammu. This concludes that bacterial contamination of donor blood and blood components is common in our hospital setting. Active surveillance methods to improve the safety of transfusion, regular monitoring and educating the clinical staff can help in reducing the contamination of transfusion blood.

Keywords
Monitoring, Prevalence, Sepsis, Surveillance, Transfusion.

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Introduction

Transfusion transmissible infection (TTI) is defined as the infection resulting from the introduction of a pathogen into a person through blood transfusion and such infections remain a leading cause of post-transfusion mortality and morbidity (Damgaard et al., 2015). For the past three decades, attention has been focused on the risk of transmission of viruses through the transfusion of blood and blood components. Technological advances in screening for TTIs such as human immunodeficiency virus (HIV), hepatitis B virus (HBV), hepatitis C virus (HCV), and syphilis have greatly improved the safety of donated blood. However, the problem of bacterial contamination of blood and blood products remains the same as it was 50 years ago.

Approximately 57% of all TTIs and 16% of transfusion-related deaths have been associated with bacterial contamination.
The bacteria implicated in the transfusion of blood and its products are Gram-negative bacilli such as *Yersinia enterocolitica*, *Pseudomonas fluorescens*, and *Pseudomonas aeruginosa*. Other species are Gram-positive species including *Staphylococcus* and *Streptococcus* spp. (Adjei *et al.*, 2009)

The bacterial contamination of whole blood and its various components can occur at several points including production of blood bags, donor venepuncture, blood donor bacteraemia, blood component separation or at the time of transfusion. (Bolarinwa *et al.*, 2011) Contaminated blood units may contain a numbers of virulent bacteria as well as endotoxins that are considered to be fatal to the recipient. Although the initial concentration of bacteria in blood components may be very low, these few viable pathogens can grow over time and achieve transfusion-relevant concentrations prior to transfusion. Further warm temp (20-35°C), high relative humidity (80%-90%), and unreliable refrigeration favors bacterial contamination of blood and blood components.

Bacterial contamination of transfusion blood is an important but overlooked health hazard which may lead to hospital acquired infection in the recipients. With this objective in mind, this study was undertaken to evaluate:

1. The prevalence of bacterial contamination of donor blood and blood components received in our hospital for sterility testing.
2. To determine blood products most likely to be contaminated.
3. To identify the microorganisms involved.

**Materials and Methods**

**Study design**

This was a retrospective study conducted in the Department of Microbiology, Government Medical College, Jammu. This study was carried over a period of 1 year from April 2017- April 2018.

**Sample collection**

All the random samples of stored whole blood and blood components (Packed cells and platelets) meant for transfusion received in the Microbiology laboratory for sterility testing. Samples from blood products that tested positive for routinely tested TTIs (HIV, HBV, HCV, and syphilis) were not included in the study.

**Sample processing**

Sample processing was done using standard aseptic precautions. Stored blood in bags was thoroughly mixed, and the end of the tied tubing was disinfected using 70% isopropyl alcohol. It was then cut with sterile scissors to discard any clotted blood in the line. Some of the mixed blood from the main bag was allowed to seep into the line. A sterile syringe was used to withdraw 5 ml of sample (whole blood, packed cell, platelets) from the line and was dispensed into 50 mL of liquid broth in blood culture bottles. The end of each line was then sealed to prevent blood from flowing back into main bag.

**Bacterial isolation and identification**

After overnight incubation, sterile loopful of broth were sub-cultured on to Blood agar and MacConkey agar plates and incubated aerobically for 18-24 hours at 37°C. All plates were examined for visible growth.

The colonies were identified as per standard microbiological procedures. The bacteria were identified by their colony morphology, Gram staining, biochemical and sugar fermentation tests. The bottles were incubated 37°C up to 7 days before they were discarded.
Results and Discussion

A total of 136 random samples (55 whole blood, 48 platelets and 33 packed cells) were received in the Microbiology for sterility testing. Of the 136 samples tested, 17 (12.50%) were found to be contaminated with bacteria.

In the present study, packed cells 7 (21.21%) had a significantly higher level of bacterial contamination compared to the rest of the blood products i.e. platelets 5 (10.41%) and whole blood 5 (9.09%). Table 1 shows the level of contamination of the various blood products.

20 isolates were obtained from 17 culture positive samples i.e. 14 (82.35%) yielded single bacterial isolate, 3(17.64%) yielded 2 bacterial isolates.

In our study, both gram positive and gram negative bacteria were equally isolated. Among the gram positive isolates, the leading blood contaminants were *Staphylococcus aureus* 3(15%) and CONS 3(15%) followed by *Bacillus sp.* 2(10%) and *Enterococcus sp.* 2(10%). *Klebsiella sp.* 7 (35%) was the predominant gram negative bacteria to be isolated. 1(5%) isolate each of *Citrobacter sp.*, *Pseudomonas sp.* and *Escherichia coli* were obtained (Figure 1).

The length of storage of the blood samples ranged from 0 to 10 days. 2(11.76%) contaminated samples had <3 days of storage. Most of the contaminated samples ie. 11 (64.70%) had 3-7 days of storage as compared to 4 (23.52%) which had >1 week of storage (Figure 2 and Table 2).

Among the 17 contaminated samples obtained, 10 (58.82%) were received from Blood Bank, GMC Jammu and 7 (41.17%) from SMGS Hospital, Jammu.

Of the blood group types, blood Group O had significantly higher (52.94%, 9/17) prevalence of bacterial contamination compared to blood Group A (23.52%, 4/17), blood Group B (17.64%, 3/17), and blood Group AB (5.8%, 1/17).

The transfusion of bacterial contaminated blood and blood products is of public health concern which may lead to severe or even fatal consequences. In the present study, prevalence of 12.50% was reported which was in accordance with (12%) (Ethopia) (Esmael *et al.*, 2014) and (9%) (Ghana) (Adjei *et al.*, 2009). In contrast to it, developed countries reported very low prevalence such as (0.19%) (United Kingdom) (Love *et al.*, 2002), (0.2%) (United States) (Kuehnert *et al.*, 2001) and (0.1%) (France) (Perez *et al.*, 2001). It may be due to efficient infection control protocols, strict donor selection and screening procedures and thorough care during blood collection which is poor in developing countries.

| **Table 1** Level of contamination of blood products |
|---------------------------------------------|
| **Whole blood** | **Platelets** | **Packed cells** | **Total** |
| No. of Blood bags tested | 55 | 48 | 33 | 136 |
| No. of Blood bags contaminated (%) | 5 (9.09%) | 5 (10.41%) | 7 (21.21%) | 17 (12.50%) |
**Table 2** Isolated organism and time of storage

| Microorganism        | Time of Storage |
|----------------------|-----------------|
| *Staph aureus*       | Day 4           |
| *Staph aureus*       | Day 3           |
| *Staph aureus*       | Day 3           |
| CONS                 | Day 9           |
| CONS                 | Day 4           |
| CONS                 | Day 5           |
| *Bacillus sp.*       | Day 4           |
| *Bacillus sp.*       | Day 2           |
| *Klebsiella sp.*     | Day 6           |
| *Klebsiella sp.*     | Day 8           |
| *Klebsiella sp.*     | Day 2           |
| *Klebsiella sp.*     | Day 6           |
| *Klebsiella sp.*     | Day 6           |
| *Klebsiella sp.*     | Day 5           |
| *Klebsiella sp.*     | Day 8           |
| *Citrobacter sp.*    | Day 6           |
| *Pseudomonas sp.*    | Day 8           |
| *E.coli*             | Day 9           |

**Figure 1** Percentage distribution of bacterial isolates
The organisms isolated in our study both gram positive (Staph aureus, CONS, Bacillus sp. and Enterococcus sp.) and gram negative (Klebsiella sp., Citrobacter sp., E.coli, Pseudomonas sp) were reported equally. Similar results were reported by (Adjei et al., 2009) and (Opoku-Okrah et al., 2009).

In the present study it was seen that contamination of blood by gram positive bacteria was within few days of storage while contamination by gram negative bacteria was delayed. Our findings are in agreement with (Bolarinwa et al., 2011) and (Sharma et al., 2004) Gram-positive isolates being commensal or transient skin flora, contamination is thought to occur primarily during phlebotomy, as a result of incomplete disinfection and/or skin core removal by the collection needle. Therefore they are isolated soon after donation, whereas Gram negative organisms not usually detectable until after a period of proliferation during storage.

This study concludes that bacterial contamination of donor blood and blood components is common in our hospital setting. This alarms an urgent need to adopt active surveillance methods to improve the safety of transfusion. Systemic and comprehensive donor selection, maintaining a clean working environment, use of 70% isopropyl alcohol for disinfection of phlebotomy sites and temperature monitoring of storage fridges are few essential measures which needs be adopted. Regular monitoring and educating the clinical staff will surely help in reducing the contamination of blood and blood components.

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