Buffy coat pooled platelet concentrate: A new age platelet component

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Abstract:
BACKGROUND: Buffy coat pooled platelet concentrate (BCPP) is a new blood component mainly used in Europe, which has good attributes of both random donor platelets and apheresis platelets in terms of high platelet count, leukoreduced and available in emergency. We planned this study to compare quality parameters and biochemical activation markers among buffy coat pooled platelets and apheresis platelet concentrate (AP-PC) to establish the quality and safety of this new blood component during storage.

MATERIALS AND METHODS: Three different preparations of BCPP were prepared: Nonleukoreduced (BCPP Part A), leukoreduced (BCPP Part B), and leukoreduced with platelet additive solution (PAS) (BCPP Part C) using a pool of 15 ABO-matched, nonreactive buffy-coats in each experiment to avoid any donor-related variations. Ten such experiments were done. Each BCPP was equivalent to 5 buffy coat units. Ten apheresis platelets were taken as control. Serial samplings were done on day 0, 3, and 5 of collection and were assessed for: volume, platelet count, white blood cell count, swirling, pH, sterility, glucose, lactate, soluble p-selectin, Interleukin (IL)-6, IL-1β, and tumor necrosis factor alpha (TNF-α).

RESULTS: BCPP Part C (leukoreduced with PAS) maintained the best quality parameters in terms of maintenance of pH, least lactate accumulation, least sP-selectin levels, and least accumulation of inflammatory mediators (IL-6, IL-1β, and TNF-α) than the other groups >BCPP part B >AP-PC and >BCPP Part A. On day 5 of storage pH for BCPP Part A, Part B, Part C, and AP-PC was: 6.33, 6.42, 6.64, and 6.29, respectively, and soluble p-selectin (ng/ml) was 201 ± 22, 186 ± 11, 149 ± 18, 200 ± 23, respectively. BCPP Part B and AP-PC had comparable quality parameters and activation markers.

CONCLUSIONS: Buffy coat pooled platelet has comparable and even better-quality control parameters (especially leukofiltered with PAS) than conventional platelet preparation and is a good alternative for meeting platelet transfusion requirements of critical patients during emergency hours in resource-constraint setting.

Keywords: Buffy coat derived platelet component, Buffy coat pooled platelet concentrate, Leukofiltered platelets, Platelet additive solution

Introduction

Platelet transfusion therapy is vital in the management of a variety of clinical conditions, especially in extreme emergencies such as postpartum hemorrhage, intracranial bleeds, and massive transfusion. Types of platelet concentrates currently licensed for use in our country are platelet concentrates prepared from whole blood called random donor platelets (RDP) and apheresis platelet concentrate (AP-PC).

AP-PC contains platelets equal to a single therapeutic adult dose and is prestorage leukoreduced. However, AP-PC involves considerable cost, the turnaround time after the requisition is around 4-5 h, so this is not an ideal product for extreme emergencies.

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This product contains about 200 ml of plasma, so it has to be transfused to ABO-compatible recipients.

RDPs are the by-product of packed red cell preparation, and the cost is very low as compared to apheresis platelets, which can be issued in an emergency and can be transfused across the ABO barrier. However, a single RDP unit contains insufficient platelet dose for an adult, so multiple units need to be given, thereby increasing cost, and these are usually nonleukoreduced.

Buffy coat pooled platelets (BCPP) is another attractive option having merits of both the conventional platelet concentrates like platelet dose in the unit is equal to single adult dose platelets, are leukoreduced, can be issued immediately after requisition and is cost-effective as compared to apheresis platelet concentrate. If platelet additive solution (PAS) is used in BCPP, these products can be used across ABO barrier and exhibit a lower incidence of allergic reactions, including anaphylaxis, febrile nonhemolytic transfusion reactions, and increased volume of plasma in fresh-frozen plasma.\(^{[2-6]}\)

BCPP is being used in European countries, Denmark, Finland, and the Netherlands, and Canada.\(^{[7-9]}\) In India, BCPP is recently included as a new blood component for clinical use in our country as per the drugs and cosmetics (Second Amendment) Rules, 2020.

There is only one study by Chatterjee et al.\(^{[10]}\) from AIIMS New Delhi showing that BCPP and AP-PC have similar quality parameters such as platelet count, volume, pH, Glucose consumption, and lactate production. This study has not discussed the role of PAS and the platelet activation markers and interleukin (IL) levels during storage. There are no other studies from India on the use of PAS in buffy coat pooled platelets to the best of our knowledge.

We did a comparative assessment of quality parameters, and biochemical activation markers among buffy coat pooled platelets (nonleukoreduced vs. leukoreduced vs. leukoreduced with PAS) versus apheresis platelet concentrate to establish the quality and safety of this new blood component during storage. These data may provide scientific evidence to promote the use of this product in our country.

**Material and Methods**

We conducted this experimental study for 1 year from April 2016 to March 2017 after taking Institutional Ethical Committee approval. In this study, we prepared three different variations of buffy coat pooled platelets in a paired experiment; these were (1) BCPP Nonleukoreduced (Part A), (2) BCPP Leukoreduced (Part B), and (3) BCPP Leukoreduced with PAS (Part C).

These were prepared using a pool of 15 ABO-matched transfusion transmitted infection (TTI) nonreactive buffy coats in each experiment to avoid any donor-related variations. For the availability of TTI results on the same day, we have chosen days where we have blood donation camps with good collection by 11 AM, and TTI results are available by 5 PM. The predonation platelet count of donors was not checked.

As the pool of 15 was divided into three parts, virtually each BCPP unit was equivalent to 5 buffy coat units [Figure 1]. As per the European guideline,\(^{[11]}\) recommendations, 4–6 bags of buffy coats need to be pooled. As we were comparing the product with apheresis platelet which usually have platelet counts of \(>3 \times 10^{11}\), we chose to pool five buffy coat bags to get comparable platelets in both products so that other parameters can be compared.

A convenient sample size of ten such paired experiments was kept as this was a feasibility study for BCPP (BCPP was not licensed product in India at that time of planning of study) and to minimize the wastage of scarce resource as this was an in vitro study, BCPP units were discarded after the last day of storage.

Ten apheresis platelets were taken in the control group. These were collected from voluntary apheresis donors on the apheresis system (Amicus®, Fenwal Inc., USA) following the departmental standard operating procedure. The target yield for apheresis procedures was kept at \(3 \times 10^{11}/\text{bag}\).

**Preparation of buffy coat pooled platelet**

Whole blood was collected in Top and Top 450 ml quadruple bags (HLL Donato, Thiruvananthapuram, Kerala, India). These bags were subjected to hard centrifugation (5000 g × 5 min at 22°C) using a refrigerated centrifuge (Cryofuge 6000i, Thermofisher, USA) and around 40–50 ml buffy coat was expressed into buffy coat transfer bag with minimal plasma.

Fifteen of ABO-matched buffy coat bags as per inclusion criteria were prepared by the methods described above. After hold overtime of around 1 h, these buffy coat bags were pooled in pairs and finally into a 1 L transfer bag using the sterile connecting device (TCD: GenesisBPS, New Jersey, USA). After mixing the contents of the bag, the pooled buffy coat was equally divided into three parts in transfer bags marked as A, B, and C.

Plasma components from any two of these fifteen donors were pooled, and 160 ml each was added to Part A, and B. The same amount of PAS (SSP + ™, Macopharma,
Mouvaux, France) was added to Part C (PAS: Plasma was maintained approximately in a ratio of 70:30 in part C as per literature.\textsuperscript{[12,13]} This ratio was calculated based on the total volume of the product, and the PAS volume added).

A one liter capacity transfer bag was attached to part A. An integral leukofilter with transfer bag one liter capacity (Bio P, Fresenius Kabi, Germany) was attached to part B and C. All three bags were subjected to soft centrifugation (520 g × 9 min at 22°C) followed by transfer of platelet product directly into transfer bag in part A and through leukofilter in Part B and C, leaving behind the residual white blood cell (WBC) and red blood cell (RBC). Hence, BCPP products with a volume range of 200–250 ml were prepared for further evaluation. This range was kept identical to the volume of AP-PC as per DGHS\textsuperscript{[14]} and AABB\textsuperscript{[15]} guidelines to facilitate comparisons. European guidelines were used as quality control criteria for BCPP as it is mostly licensed in Europe.\textsuperscript{[11]}

**Storage and sampling**

Both BCPP and AP-PC were stored for 5 days in a flat-bed agitator at 22°C ± 2°C with continuous agitation. A 5 ml sample was collected from each of these bags on days...
Each platelet unit was assessed for the following parameters: Volume, platelet count, WBC count, swirling, pH, Sterility, glucose, lactate, soluble p-selectin, IL-6, IL-1β, and TNF-α.

The assessment of swirling in platelets concentrates was done using visual inspection and scoring\textsuperscript{[10]} on a scale of 0–3.

### Sample processing

Two milliliter sample was transferred to ethylenediaminetetraacetic acid (EDTA) tube for platelet count, WBC count, and pH evaluation. The 3-mL sample was taken in a plain plastic tube and subjected to centrifugation of 3000 rpm for 10 min in the laboratory centrifuge. Platelet poor plasma (PPP) was stored in cryovials below-80°C in the deep freezer for testing of glucose, lactate, soluble p-selectin, and Interleukins.

Platelet count and WBC count (PART A) was done using a calibrated automated hematology analyzer (Orion 60, Ocean Medical, India), WBC count (Part B and C and ACPC) was done using Nageotte chamber.\textsuperscript{[11]} pH was done using a thin probe pH meter (Adwa, Romania, Europe). Glucose and lactate concentrations were measured using an automated biochemistry analyzer (Backman AU55811, Indianapolis, USA).

Soluble p-selectin, IL-6, IL-1β and tumor necrosis factor alpha (TNF-α) were done on stored PPP using enzyme-linked immunosorbent assay (ELISA) kits (Diaclone, Besancon, France) with their sensitivity/minimum detectable limit as 3.6 ng/ml, 2 pg/ml, 6.5 pg/ml, respectively. For testing sterility, culture was done on day 5 of storage with 10 ml of platelet sample, using an automated blood culture system (BACTEC 9240, BD Diagnostics, USA) using standard aerobic culture vials. After doing all the tests and taking appropriate samples, the platelet units were discarded according to Biomedical Waste Management guidelines 2016.\textsuperscript{[18]}

Collected data were analyzed using SPSS (IBM Statistical product and service solution version 22.0 (SPSS, Chicago, IL, USA) and online Graph Pad software (Prism 5 for Windows) version 5.01. Pearson’s Chi-square test was used to evaluate differences between groups for categorized variables. Paired and unpaired t-test and analysis of variance (ANOVA), including repeated-measures ANOVA, were used to calculate the difference of means for quantitative variables. All tests were performed at a 5% level of significance. Thus an association was considered statistically significant if the $P < 0.05$.

### Results

#### Physical characteristics of buffy coat pooled platelets

**Physical appearance**

All platelet components showed normal physical appearance throughout the storage period. None of the platelet components showed leakage, RBC contamination, precipitates, gas formation, or discoloration. The volume of BCPP Part A, Part B, part C, and AP-PC was 225 ± 6 ml, 226 ± 5 ml, 226 ± 5 ml, and 223 ± 4 ml, respectively on day 0 of storage. In the BCPP Part C ratio of PAS: Plasma was 68%: 32%.

Swirling was maintained at “Score 3” in both AP-PC and all three types of BCPP on day 3 of storage. On day 5 of storage, swirling was maintained at “Score 3” in 100% Part B and C BCPP, but 30% (3 out of 10) of BCPP part A and 50% (5 out of 10) of AP-PC platelets showed a swirling of “Score 2.”

**Cellular counts**

**Platelet count**

Platelet content was found to be the highest ($P < 0.001$) in BCPP part A, followed by BCPP part B and C, followed by apheresis platelets [Table 1]. All platelet components met the quality control criteria for platelet count. There was around 10% and 12% decrease in platelets due to leukofiltration in BCPP part B and part C, respectively. BCPP part B suspended in plasma contained more platelets than part C suspended in PAS ($P = 1.000$).

There was a decreasing trend in platelet count during storage and decrease was maximum in BCPP part A (nonleukoreduced), and platelet count was most stable in BCPP part C [Leukoreduced with PAS; Figure 2].

**White blood cell counts**

WBC content was variable in different platelet preparations and ranged from 10⁰ in BCPP part A, 10² in apheresis PC, and 10⁵ in leukofiltered components: BCPP part B and C [Table 1].

**pH changes over storage**

pH was maintained better in BCPP part C as compared to part A, Part B, and AP-PC on each day of storage, and the difference was significant across all three products. However, pH differences among part A, part B, and AP-PC were comparable [Table 1 and Figure 3].

**Glucose and lactate concentration**

There was a progressive decrease in glucose on storage in all different types of platelet components. However, on the last day of storage, glucose levels were best preserved in BCPP Part B [Table 1]. The
Table 1: Comparative chart of all quality parameters for Buffy coat pooled platelet concentrate Part A, B, and C and apheresis platelet concentrate during storage

| Quality control parameter | Day of storage | BCPP Part A (n=10) | BCPP Part B (n=10) | BCPP Part C (n=10) | AP-PC (n=10) | P   |
|---------------------------|----------------|-------------------|-------------------|-------------------|--------------|-----|
| Volume (ml)               |                | 225±6 (218-236)   | 226±5 (220-234)   | 226±5 (218-234)   | 223±4 (220-230) |     |
| Platelet count (x10^11/Bag) | 0              | 3.95±0.14 (3.85-4.04) | 3.58±0.08 (3.49-3.68) | 3.49±0.10 (3.40-3.58) | 3.33±0.20 (3.24-3.42) | <0.001* |
|                           | 3              | 3.83±0.12 (3.73-3.92) | 3.55±0.13 (3.46-3.65) | 3.45±0.10 (3.35-3.54) | 3.25±0.21 (3.16-3.35) |     |
|                           | 5              | 3.42±0.09 (3.33-3.51) | 3.34±0.12 (3.25-3.43) | 3.31±0.09 (3.23-3.40) | 3.03±0.21 (2.95-3.12) |     |
| WBC count (x10^5/Bag)     | 0              | 3214±200.0 (3076-3360) | 2.7±0.2 (2.55-2.79) | 2.7±0.2 (2.47-2.82) | 39.7±2.4 (37.9-41.4) |     |
|                           | 3              | 3038±179.7 (2906-3164) | 2.5±0.1 (2.34-2.67) | 2.5±0.2 (2.28-2.64) | 38.3±2.4 (36.6-40.0) |     |
|                           | 5              | 2819±197.8 (2670-2964) | 2.3±0.2 (2.11-2.40) | 2.2±0.3 (2.03-2.39) | 35.1±3.0 (32.9-37.2) |     |
| pH (mean±SD) range        | 0              | 0.67±0.09 (0.67-0.68) | 0.67±0.09 (0.66-0.78) | 0.68±0.04 (0.63-0.69) | 0.72±0.07 (0.67-0.78) | <0.001* |
|                           | 3              | 0.61±0.11 (0.64-0.73) | 0.65±0.10 (0.65-0.71) | 0.64±0.05 (0.67-0.69) | 0.62±0.07 (0.66-0.68) |     |
|                           | 5              | 0.63±0.07 (0.62-0.67) | 0.64±0.05 (0.63-0.66) | 0.64±0.06 (0.60-0.68) | 0.65±0.07 (0.62-0.63) |     |
| Glucose (mg/dl)           | 0              | 366±18 (349.2-383.6) | 367±18 (350-384.4) | 166±9 (149.4-183.6) | 306±45 (307.6-384.4) | <0.005* |
|                           | 3              | 291±27 (272.4-309.6) | 293±26 (274.3-311.6) | 136±10 (117.4-154.6) | 238±42 (219.2-256.4) |     |
|                           | 5              | 230±41 (210.7-252.8) | 239±33 (215.5-261.6) | 109±10 (86.6-131.0) | 165±45 (162.8-207.7) |     |
| Lactate (mg/dl)           | 0              | 65±14 (43.4-51.6) | 60±11 (42.6-79.2) | 38±9 (22.2-51.6) | 42±10 (23.6-79.8) | <0.001* |
|                           | 3              | 131±30 (87.2-181.5) | 124±22 (85.4-152.2) | 70±17 (32.4-89.6) | 131±18 (99.4-153.4) |     |
|                           | 5              | 172±25 (128.3-201.4) | 173±26 (121.6-197.2) | 99±23 (55.4-132.6) | 182±11 (163.2-196.8) |     |

Platelet counts significantly higher in BCPP part A than Part B, C, and AP-PC on day 0 to 5, *pH of BCPP Part C significant higher as compare to Part B, C, and AP-PC on day 0 to 5, $Glucose levels were significantly higher in BCPP part A & B than C and AP-PC on day 0 to 5, $$Lactate levels were significantly lower in BCPP part C as compared to A, B and AP-PC on day 0 to 5 of storage. BCPP=Buffy coat pooled platelet concentrate, AP-PC=Apheresis platelet concentrate, SD=Standard deviation, WBC=White blood cell

Platelet activation markers and cytokine accumulation

**Soluble p-selectin level**

On day zero of storage, levels of soluble p-selectin was the maximum in AP-PC followed by BCPP part A, part B, and followed by a significant difference seen in part C, respectively ($P < 0.001$) [Table 1]. On the last day of storage, the soluble p-selectin level was maximum in BCPP-part A, followed by apheresis platelets followed by BCPP part B and followed by a significant difference with part C, respectively ($P < 0.001$).

**Concentrations of inflammatory cytokines**

A steady rise in the level of all inflammatory mediators (IL-6, TNF-α, and IL-1 β) was observed in BCPP part A during storage and the change from day 0 to day 5 was statistically significant ($P < 0.0001$). The level of IL-6 was below the detectable limit of the kit (<2 pg/ml) on all days of storage for BCPP part B, part C, and AP-PC level of TNF-α was below the detectable limit of the kit (<6.0 pg/ml) on day 3 and 5 of storage for BCPP part B and C. The decrease in TNF-α levels from day 0 to 5 was statistically significant in AP-PC ($P < 0.02$) [Table 2].

**Interleukin-1 beta**

Rise in IL-1 β levels in BCPP Part A as compared to part B and C was significant ($P < 0.05$) on all days of storage. BCPP Part B and Part C had comparable IL-1 β levels.

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**Figure 2:** Trend of platelet count during storage

**Figure 3:** Trends of pH during storage
levels ($P < 0.824$). IL-1β levels were higher in AP-PC than BCPP part B and C on all days of storage ($P < 0.0001$) but lower than BCPP part A ($P < 0.125$) [Table 2].

**Bacterial sterility testing**

In our study, “two BCPP out of 30 BCPP failed sterility check.” Both cultures were positive in the BCPP part C. The bacterial strains identified were Staph. Hominis and lactobacillus.

**Discussion**

There is a need to develop platelet concentrates that are available in emergency and have good quality in terms of platelet count, leukoreduction, and minimum platelet activation.

All platelet components met the quality control criteria for platelet count, which is $>2 \times 10^{11}$ platelets per bag for BCPP as European guidelines.[11] As we pooled five buffy coat bags, the platelet content was $>3 \times 10^{11}$ and was comparable with apheresis platelet concentrate so that other quality parameters can be compared. Platelet content was found to be the highest in BCPP part A, followed by BCPP part B and C due (loss during leukofiltration) followed by apheresis platelets. A study by Chatterjee et al.[10] in which four buffy coats were pooled and leukofiltered, the mean platelet count was $3.38 \times 10^{11}$ per bag, similar to the component in our study (BCPP Part B) had a mean platelet count of $3.58 \times 10^{11}$ per bag (equivalent to 5 buffy coat units).

All platelet bags were meeting QC criteria for WBC count as per the European guidelines. A study by Kaur et al.[9] showed similar WBC content in nonleukofiltered, leukofiltered buffy coat platelets and apheresis platelet concentrates.

In our study, pH was best maintained in leukofiltered BCPP with PAS (6.64) at day 5 of storage as compared to apheresis platelets (6.29). On day 5 of storage, none of the apheresis platelets, 20% BCPP part A, 60% of BCPP part B, and 100% of BCPP part C were meeting the QC criteria as per European guidelines.[11] A similar finding was observed by Alhumaidan and Sweeney,[20] where they found that platelet suspended in PAS had maintained better pH (6.50) than with plasma (<6.30) on day 5 of storage. Chatterjee et al.[10] observed a higher pH of 6.9 on 5th-day leukofiltered BCPP, which may be attributed to lower platelet count (four vs. five buffy coat units) and more volume in BCPP as compared to our study. A study by Jain et al.[21] observed a mean pH of 6.4 in AP-PC at day 5 of storage, which is comparable to the present study.

Glucose is used as a source of energy by platelets, so glucose concentration decreases during storage with a rise in lactate concentration. This accumulation of lactate leads to a decrease in pH and activation of platelets. To prevent the accumulation of lactate, the PAS used sodium acetate as a source of energy instead of glucose. This was evident in our study that platelet activation was least in BCPP part C with better pH maintenance due to the least lactate accumulation as compared to other platelet groups.

Soluble p-selectin, which is a sensitive marker of platelet activation, is also in concordance with other results. On the last day of storage, the soluble p-selectin level was minimum in BCPP part C (149 ng/ml) versus (200 ng/ml) in apheresis platelets.

**Table 2: Comparative chart of platelet activation markers for buffy coat pooled platelet concentrate Part A, B, and C and AP-PC during storage**

| Platelet activation marker | Day of storage | BCPP Part A ($n=10$) | BCPP Part B ($n=10$) | BCPP Part C ($n=10$) | AP-PC ($n=10$) | $P$ |
|----------------------------|----------------|----------------------|----------------------|----------------------|----------------|-----|
| Soluble p-selectin (ng/ml) | 0              | 137±10 (127.6-147.5) | 128±21 (117.4-137.8) | 86±18 (75.3-96.6) | 152±12 (141.3-162.6) | <0.001* |
| (mean±SD) range            | 3              | 165±14 (152.9-177.9) | 154±16 (140.6-165.6) | 125±14 (112.5-137.6) | 158±30 (145.1-170.6) |     |
|                            | 5              | 201±22 (188.9-213.6) | 187±11 (174.2-198.8) | 149±18 (136.6-161.5) | 200±23 (187.5-212.3) |     |
| IL-6 (pg/mL)               | 0              | <2.0*                | <2.0*                | <2.0*                | <2.0*          |     |
| (mean±SD) range            | 3              | 10±2.6 (6.1-14.5)    | <2.0*                | <2.0*                | <2.0*          |     |
|                            | 5              | 18±5.3 (13.6-30.5)   | <2.0*                | <2.0*                | <2.0*          | <0.01* |
| IL-1β (pg/mL)              | 0              | 61±5.7 (56.4-66.7)   | 47.9±10.2 (43.5-53.7) | 41.0±5.5 (36.6-45.1) | 54±5.8 (49.4-59.6) | <0.05** |
| (mean±SD) range            | 3              | 66±5.5 (60.5-73.9)   | 46.1±14.4 (39.6-52.8) | 46.0±7.6 (39.6-53.3) | 56±5.8 (50.6-62.4) |     |
|                            | 5              | 76.6±9.8 (69.5-84.6) | 53.5±14.4 (46.1-61.6) | 51.6±9.0 (43.2-58.4) | 67.9±11 (60.3-75.3) |     |
| TNF-α (pg/mL)              | 0              | 12.0±4.0 (9.3-15.8)  | 8.9±6.0 (6.2-13.4)   | 7.8±5.8 (0-11.8)    | 11.1±4.7 (7.4-15)  | <0.001# |
| (mean±SD) range            | 3              | 21.0±4.7 (18.1-24.4) | <6.0*                | <6.0*                | 8.9±7.2 (0-12.6)  |     |
|                            | 5              | 27.7±4.1 (26.2-30.6) | <6.0*                | <6.0*                | 6.1±3.0 (0-8.8)   | <0.001# |

*The level was below the detectable limit of the kit, *sP-selectin concentration was significantly lower in BCPP part C than A, B, and AP-PC on day 0 to 5, **IL-6 levels were significantly higher in BCPP part A than B, C, and AP-PC on day 5, ***IL-1β levels were significantly higher in BCPP part A and AP-PC than B, C on day 0 and 5. #TNF-α levels were significantly higher in BCPP part A and AP-PC than B, C on day 0 and 5. **BCPP with PAS (6.64) at day 5 of storage as compared to apheresis platelets (6.29). On day 5 of storage, none of the apheresis platelets, 20% BCPP part A, 60% of BCPP part B, and 100% of BCPP part C were meeting the QC criteria as per European guidelines.[11] A similar finding was observed by Alhumaidan and Sweeney,[20] where they found that platelet suspended in PAS had maintained better pH (6.50) than with plasma (<6.30) on day 5 of storage. Chatterjee et al.[10] observed a higher pH of 6.9 on 5th-day leukofiltered BCPP, which may be attributed to lower platelet count (four vs. five buffy coat units) and more volume in BCPP as compared to our study. A study by Jain et al.[21] observed a mean pH of 6.4 in AP-PC at day 5 of storage, which is comparable to the present study.
by Kaur et al.,\cite{20} where they concluded that platelet preparations with a WBC count >10^7 had significantly higher levels of cytokines in comparison to preparations with WBC count less than or equal to 10^6. Results of our study are corroborative with the study by Muylle et al.,\cite{21} in demonstrating trends in cytokine levels during storage in both leukoreduced and nonleukoreduced platelet concentrates. Platelet concentrates with WBC count less than or equal to 10^6 have cytokines below detection threshold.\cite{22-24} This could be due to the neutralization of these inflammatory mediators by plasma anti-inflammatory protein.

Root cause analysis for bacterial culture positivity in two BCPP Part C bags showed that it was most probably due to improper sealing, sterile docking, or contamination at the time of sampling, as culture from remaining PAS, BCPP part A and Part B from the same experiment were negative and both the bacterial strains identified were skin commensals. A recent study by Ramirez-Arcos et al.,\cite{25} showed culture positivity inuffy coat platelets as 1 out of 10,000 platelets. A study by Van Der Meer et al.,\cite{26} on, the sterility of plastic tubing weld in the platelet component, showed 2 out of 244 welds having bacterial contamination because of sterile weld failure. This observation should not affect the decision regarding the safety of the BCPP product. As this was an experimental study involving extensive product manipulation, the study reemphasizes the fact that extensive product manipulation poses a risk of bacterial contamination and all precautions to prevent this should be taken.

In a tertiary care hospital like ours, we have to maintain a large inventory of platelets (RDPs and AP-PC) to fulfill the requirements of both emergencies and multiply transfused hematooncology patients. Ours is a public sector institution and primarily caters to the patients from low socioeconomic strata of society who cannot afford apheresis platelet concentrates. These platelets are usually collected as directed donations. Hence, in a resource constraint setting where an inventory of voluntary apheresis platelets is not available, it is need of the hour to develop high-quality platelet concentrate like buffy coat pooled platelet (BCPP) having high platelet content, which can be leukoreduced, available on the shelf for emergency use. The availability of BCPP will further reduce the need for apheresis platelet donations during emergency hours. However, BCPP may lead to increased donor exposure (for TTI transmission) as compared to apheresis platelets. Here, we should also consider the feasibility of NAT testing of these platelet preparations. As BCPP is whole blood-derived hence NAT testing is easily feasible without any additional cost versus NAT testing of apheresis platelets, which is often not feasible due to time constraints, especially in emergency conditions and if feasible, will have an additional cost for NAT. So further studies are required to assess TTI safety of NAT-tested BCPP versus Rapid/ELISA/Chemiluminescence tested apheresis platelets.

Regarding the feasibility of NAT testing, buffy coat pooling can be initiated on the day of the collection after ELISA results and final labeling and inclusion in inventory can be done after the NAT results are available. If NAT results are positive (which will be rare considering the NAT yield), the BCPP can be discarded. Hence, if NAT is done, BCPP will be available on day 1 of storage and will be available on the shelf for the next 4 days for any emergency issue.

RDP can serve the purpose for an emergency issue, but benefits such as the feasibility of leukoreduction and the use of PAS is better in BCPP.

Hence, BCPP has good attributes of both conventional platelet (RDPs and apheresis platelets) components with additional benefits as described above.

As BCPP is recently listed as a blood component in Drugs and Cosmetics (Second Amendment) Rules, 2020, Government of India for use in patients in our country, these data will provide some scientific evidence to promote the use of this product in our country.

Conclusions

Buffy coat pooled platelets (BCPP) have shown equivalent or even better quality parameters (especially Leukofiltered with PAS) and biochemical activation marker profile than apheresis platelets and conventional platelet preparations and could be a good alternative for meeting platelet transfusion requirements of critical patients during emergency hours in a resource-constraint setting.

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Conflicts of interest
There are no conflicts of interest.

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