Potassium as a Surrogate Marker of Debris in Cell-Salvaged Blood

Dale F. Szpisjak, MD*, David S. Edgell, CCP†, and Bruno Bissonnette, MD*

Departments of *Anesthesia and †Perfusion, The Hospital for Sick Children, University of Toronto, Toronto, Ontario, Canada

Centrifuge-based cell salvage systems have decreased the use of homologous blood transfusions. Although the evidence is anecdotal, the risk associated with the use of salvaged erythrocytes seems related to cellular and chemical contaminants. We sought to determine if potassium can be a surrogate marker for cellular debris and to measure the residual heparin level. Four units of expired whole blood were heparinized and concentrated with a Sequestra®1000 (Medtronics®, Parker, CO) cell salvage device. The potassium, free hemoglobin, leukocyte, and platelet counts were sampled after each 250-mL normal saline wash aliquot, to a total wash volume of 1500 mL, whereas the heparin samples were obtained at wash volumes 0 and 1000 mL. Potassium, leukocyte, and platelet concentrations at wash volumes 0 and 250 mL were significantly greater than at all other volumes ($P < 0.001$). After 500 mL of saline wash, the change in these values was not significant. The mean (± SD) heparin levels (units/mL) at wash volumes 0 and 1000 mL were 10.2 (±3.1) and 0.11 (±0.02), respectively ($P < 0.007$). The r^2 values for free hemoglobin, leukocytes, and platelets versus potassium were 0.006, 0.992, and 0.995, respectively. No convenient test has been validated as an indicator of salvaged erythrocyte cleanliness. This *in vitro* study suggests that residual potassium concentration seems to be a good indicator of quality after washing with a contemporary intraoperative salvage system.

(Anesth Analg 2000;91:40–3)

Centrifuge-based cell salvage and washing systems, available since the 1970s, have decreased the use of homologous blood transfusions (1), which carry the risk of infectious (HIV, hepatitis, cytomegalovirus, Epstein-Barr virus, bacterial contamination) and immunologic complications (hemolytic transfusion reactions, alloimmunization, immunomodulation), which are avoided by the use of autologous blood (2,3). However, the use of salvaged erythrocytes is not without risk and may cause a disseminated intravascular inflammatory response (4).

The American Association of Blood Banks states that assessing the quality of washed erythrocytes may be of value (5). However, the time required to process many assays (such as residual albumin, heparin, free hemoglobin, leukocyte count, bacterial cultures, and complement fractions) precludes withholding the washed erythrocytes during acute hemorrhage. Although the information obtained from these tests may allow retrospective review, contributing to the improvement of the salvage and washing process, a quickly available assay would indicate the need for further processing or withholding poor quality blood.

To compensate for the lack of a specific and quickly available quality check, potassium assays have been suggested as a surrogate (P. S. Potter, written communication, April, 1995). As erythrocytes hemolyze during collection and storage in the reservoir, they release both free hemoglobin and potassium. Not only can free hemoglobin lead to acute tubular necrosis (6), but the association of massive transfusion and hyperkalemia in children is also a well known danger (7,8). In theory, the elimination of potassium should correlate with the reduction in free hemoglobin, leukocytes, and platelets, but this remains an unproven hypothesis.

This study had two goals: to determine the correlation of residual potassium with cellular debris (measured as free hemoglobin, leukocytes, and platelets) and to determine the residual heparin level after a standard wash volume.

This manuscript was written by LCDR Dale F. Szpisjak, MC, USN, while a fellow at the University of Toronto/Hospital for Sick Children training in pediatric anesthesiology. The views expressed in this manuscript are those of the author and do not reflect the official policy or position of the Department of the Navy, Department of Defense, or the United States Government.

Accepted for publication March 7, 2000.

Address correspondence and reprint requests to Dr. Dale F. Szpisjak, Department of Anesthesiology, The National Naval Medical Center, 8901 Wisconsin Ave., Bethesda, MD 20889-5600.

0003-2999/00

Methods

After institutional approval, 4 units (530 mL/U) of expired whole blood were obtained from the blood bank. After priming a 20-micron cardiotomy reservoir (Medtronics®, Parker, CO) with 175 mL of heparinized (30,000 U/L) saline solution, a unit of blood was added. The blood was concentrated with a Sequestra®1000 (Medtronics®) cell salvage device (225-mL bowl, centrifuge speed 5600 rpm, fill and wash speed 300 mL/min.) and washed with 1500 mL of normal saline. The wash aliquot was 250 mL. After each aliquot, the blood was transferred to the reinfusion bag for sampling, then returned to the centrifuge. The potassium, free hemoglobin, leukocyte, and platelet counts were sampled after each aliquot, whereas the heparin samples were obtained at the beginning of the study (wash volume 0 mL) and after 1000-mL wash. These samples were assayed with a NOVA® Stat Plus 5 (Waltham, MA), Coulter® Max-M (Burlington, Ontario, Canada), Beckman® DU-70 Spectrophotometer (Mississauga, Ontario, Canada), and the Stago® Anti-Xa heparin assay (Asnieres, France). (Factor Xa was added to the sample to allow determination of the amount of heparin in solution.)

Among-group comparisons were analyzed by using repeated-measures analysis of variance and the Student-Newman-Keuls test for multiple comparisons. The coefficients of determination (r^2) was determined by using linear regression analysis. Heparin assays were compared by using paired Student's *t*-tests. A $P < 0.05$ was accepted to express statistical significance.

Results

The mean (±SD) age for the expired blood was 39 ± 3.4 days. The reductions in potassium, platelet, and leukocyte concentrations were significantly different at all wash volumes 500 mL and larger when compared with wash volumes 0 and 250 mL ($P < 0.001$; Figures 1, 2, and 3). However, there were no further reductions in these markers beyond wash volume 750 mL. The mean (±SD) heparin levels (U/mL) at wash volumes 0 and 1000 mL were 10.2 (±3.1) and 0.11 (±0.02), respectively ($P < 0.007$). (Heparin was below the detectable limit in three samples. The lowest detection limit was used for statistical analysis.) The r^2 values for the relationship between free hemoglobin, leukocyte, and platelet concentrations versus potassium were 0.006, 0.992, and 0.995, respectively. The free hemoglobin curve was fit to a cubic polynomial equation, although excluding the value at wash volume 0 demonstrated a curve with exponential decay. At least 750 mL of volume wash was required to significantly reduce the concentration of free hemoglobin ($P < 0.01$).

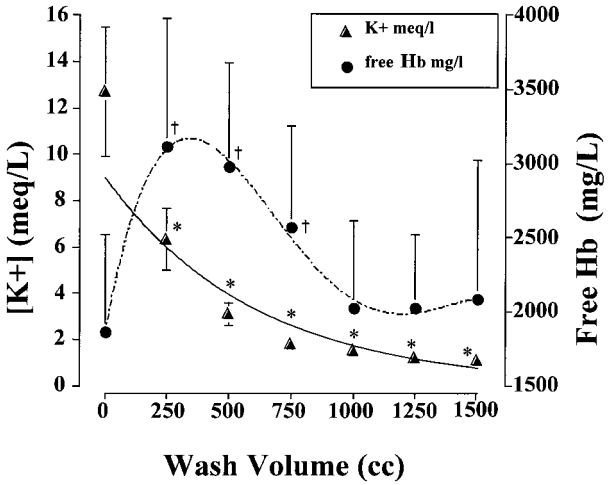

Figure 1. The concentrations of potassium (K^+) and free hemoglobin (Hb) in expired whole blood versus cell-saver wash volume. Results are means ± SD. *$P < 0.001$ for K^+ concentrations compared with that at wash volume 0 mL. †$P < 0.001$ for free Hb concentrations compared with that at wash volume 0 mL. After 750 mL of wash volume, there were no further significant reductions in the concentrations of free Hb or K^+.

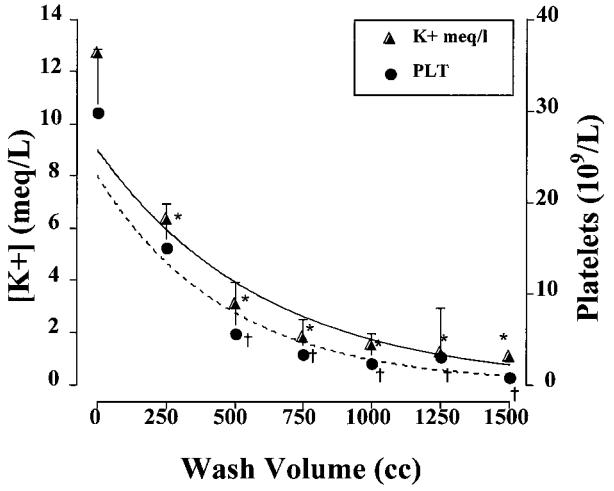

Figure 2. The concentrations of potassium (K^+) and platelets (PLT) in expired whole blood versus cell-saver wash volume. Results are means ± SD. * $P < 0.001$ for K^+ concentrations compared with that at wash volume 0 mL. †$P<0.001$ for PLT concentrations compared with those at wash volumes 0 and 250 mL. After 750 mL of wash volume, there were no further significant reductions in the concentrations of PLT or K^+.

Discussion

Potassium was an excellent marker for the level of nonerythrocyte cellular debris (leukocytes and platelets). However, the correlation between the elimination of free hemoglobin during the wash process was not indicated by the residual concentration of potassium. The initial increase in the free hemoglobin level from wash volume 0 to 250 mL was unexpected and has not been reported. The reason for this increase is unknown; however, there may be two explanations

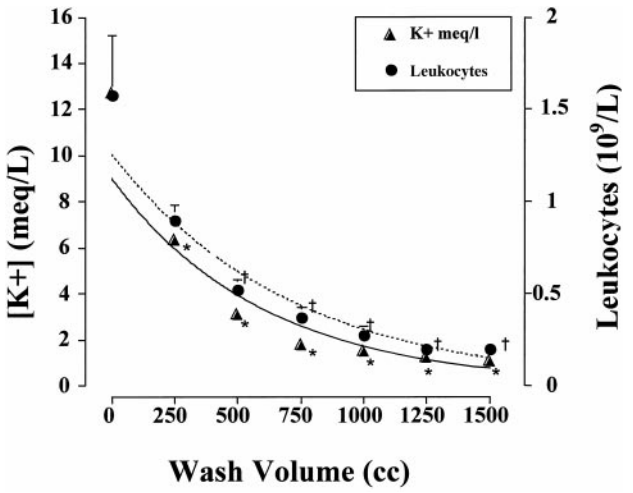

Figure 3. The concentrations of potassium (K^+) and free leukocytes in expired whole blood versus cell-saver wash volume. Results are means ± SD. *$P < 0.001$ for K^+ concentrations compared with that at wash volume 0 mL. †$P<0.001$ for leukocyte concentrations compared with those at wash volumes 0 and 250 mL. After 750 mL of wash volume, there were no further significant reductions in the concentrations of leukocytes or K^+.

for this pattern. First, the combination of centrifugal force applied to the blood and the consequential shearing effect during the initial 250-mL saline wash could have contributed to hemolysis. Second, the age of the (expired) erythrocytes might have predisposed them to hemolysis and release of free hemoglobin. Although one may believe that potassium should have paralleled this free hemoglobin increase, it has been well demonstrated that the release of potassium occurs continuously during storage and that the amount left within the cell at this stage is decreased (9). It is also likely that the elimination of potassium (atomic weight = 39 d) is more efficient than the larger hemoglobin molecule (molecular weight = 64 kd).

The free hemoglobin level after a 1000-mL wash volume (2020 ± 594 mg/L) is consistent with salvage data in patients undergoing total hip arthroplasty, a procedure associated with erythrocyte damage during salvage (10,11). Furthermore, considering that the level of free hemoglobin did not decrease further with supplemental washing to 1500 mL may support the hypothesis that shear forces applied during the process might continue to hemolyze cells.

In contrast to free hemoglobin, the removal of leukocytes and platelets correlated well with the removal of potassium. Although cell-salvaged blood is autologous and without risk of graft versus host disease, platelets and leukocytes can deposit on the bowl wall and release cytokines (12), theoretically placing the patient at risk for disseminated intravascular coagulation and adult respiratory distress syndrome, termed the "salvaged blood syndrome"(13). Although the change in potassium, leukocyte, and platelet concentrations was not statistically significant beyond a 500-mL wash volume, the results of this study show a progressive decrease in debris supporting the manufacturer's recommended wash volume of 1000 mL. A minimal acceptable potassium level of 2 mEq/L has been suggested (P. S. Potter, written communication, April, 1995). This correlates with a 78% elimination of leukocytes and 87% elimination of platelets. Finally, the efficiency of heparin elimination in this study is consistent with the 99% elimination rate reported elsewhere (14).

A limitation of this study was the use of expired blood. Although this may have led to artifactual increases in free hemoglobin, this blood may contain levels of debris exceeding those in the clinical arena, placing a greater burden on the cell washing process. A further limitation is that Coulter counters miscount fractured cells as platelets (15). Although the leukocyte and platelet counts may not be as accurate, they are still measuring debris. The poor correlation of free hemoglobin elimination may be related to the use of expired, rather than fresh, blood. The initial increase in free hemoglobin concentration may have occurred because frail, older erythrocytes hemolyzed. It is also possible that the initial wash aliquot detached hemoglobin molecules that had become weakly bound to the erythrocyte during the storage process.

These data support the hypothesis that the potassium concentration of cell-salvaged blood is a surrogate marker of cellular debris contained in stored blood as measured by platelet and leukocyte counts. As whole blood assays of potassium are rapidly available with blood gas machines, there is now evidence supporting prospective, rather than retrospective, quality control. However, applying laboratory data generated with expired whole blood to the clinical arena might be premature. Recommending a definitive potassium end point as a surrogate marker of cellular debris requires proof that elimination of free hemoglobin, leukocytes, and platelets correlates with potassium in fresh blood.

References

1. Goodnough LT. Intraoperative cell salvage in patients undergoing elective abdominal aneurysm repair: an analysis of cost and benefit. J Vasc Surg 1996;24:213–8.
2. Crosby ET. Perioperative hemotherapy. II. Risks and complications of blood transfusion. Can J Anaesth 1992;39:822–37.
3. Landers DF. Blood transfusion-induced immunomodulation. Anesth Analg 1996;82:187–204.
4. Bull BS. Hypothesis: disseminated intravascular inflammation as the inflammatory counterpart to disseminated intravascular coagulation. Proc Natl Acad Sci USA 1994;91:8190–4.
5. Guidelines for blood salvage and reinfusion in surgery and trauma. Bethesda, MD: American Association of Blood Banks, 1993:7.
6. Toshiharu Y. Haptoglobin therapy for possible prevention of renal failure following thermal injury: a clinical study. J Trauma 1985;25:281–7.

7. Brown KA, Bissonnette B, McIntyre B. Hyperkalemia during rapid blood transfusion and hypovolemic cardiac arrest in children. Can J Anaesth 1990;37:747–54.
8. Brown KA, Bissonnette B, MacDonald M, Poon A. Hyperkalemia during massive blood transfusion in pediatric craniofacial surgery. Can J Anaesth 1990;37:401–8.
9. Wallas CH. Sodium and potassium changes in blood bank blood stored human erythrocytes. Transfusion 1979;19:210–5.
10. Roberts DRD, Mah ET, Nyman TLM, Davis RW. Quality of blood prepared for autotransfusion in primary cemented total hip replacement. Anaesth Intensive Care 1991;91:382–7.
11. Mortelmans Y, Vermaut G, Van Aken H, et al.Quality of washed salvaged red blood cells during total hip replacement: a comparison between the use of heparin and citrate as anticoagulants. Anesth Analg 1994;79:357–63.
12. Sieunarine K, Lawrence-Brown MM, Goodman MA, et al. Plasma levels of the lipid mediators, leukotriene B4 and lyso platelet-activating factor, in intraoperative salvaged blood. Vox Sang 1992;63:168–71.
13. Bull B, Bull M. The salvaged blood syndrome: a sequel to mechanochemical activation of platelets and leukocytes? Blood Cells 1990;16:5–23.
14. Kling D, Börner U, v Bermann B, Hempelmann G. Heparinelimination und freis hämoglobin nach zellseperation und waschen autologen blutes mit dem Cell-Saver 4. Anasth Intensiuther Notfallmed 1988;23:88–90.
15. Morris M, Williams W, Nelson D. Automated blood cell counting. In: Beutler E, Lichtman M, Coller B, Kipps T, eds. Hematology. 5th ed. New York: McGraw-Hill, 1995:L3–11.