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Stored Platelet Functionality is Not Decreased After Warming with a Fluid Warmer

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Abstract

Background—Warming of IV administered fluids and blood products is routinely performed in the operating room to help maintain normothermia. Current guidelines recommend against the warming of platelets (PLTs), although there is no evidence for this prohibition in the literature. Our goal in this pilot study was to determine if the warming of stored PLTs had any effect on their function.

Methods—Ten units of three-day-old PLT rich plasma-derived whole blood PLTs were acquired from the transfusion service. A 5 mL aliquot was taken from each unit before warming (control samples). The remainder of the unit was then passed into a blood warming device and held there for two minutes. Post-warming (warmed) PLT samples were then collected from the effluent end of the warming device. PLT aggregometry assays with adenosine diphosphate, collagen, and arachidonic acid as agonists were performed on the control and warmed samples.

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DISCLOSURES:

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Contribution: This author performed conduct of the study, data analysis and manuscript preparation.

Attestation: Gerhardt Konig attests to approving the final manuscript and to the integrity of the original data and the analysis reported in this manuscript. He is also the archival author.

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Thromboelastography (TEG®) tests were also performed on the control and warmed samples from six of the 10 PLT units.

Results—The mean temperature of the control and warmed samples was $22.4 \pm 0.5^{\circ}\text{C}$ and $37.8 \pm 2.3^{\circ}\text{C}$, respectively. There was no significant difference (all $P = 0.13$) in any of the PLT aggregometry assays or in the maximum amplitude of the TEG® test between the control and the warmed samples. The observed mean of only one parameter decreased (PLT aggregometry with $5 \mu\text{M}$ adenosine diphosphate), by 5% (95% CI: -115% to 105%). The maximum change observed was PLT aggregometry with arachidonic acid as agonist, which increased by 116% (95% CI: -91% to 323%).

Conclusion—Although small in size, the results of this study do not support the prohibition against mechanical PLT warming. Studies of PLT activation after warming are also warranted.

Introduction

Perioperative hypothermia is associated with increased postoperative morbidity.¹ Warming IV-administered fluids and blood products with an approved blood warming device is routinely performed to help maintain normothermia during surgery. The current guidelines from the AABB (formerly known as the American Association of Blood Banks) recommend against the mechanical warming of platelets (PLTs),² although there are no data to support this recommendation in the literature.³ The main reason for this prohibition is derived from the contraindication listed in the operator's manual for the Level 1 fluid warmer, which follows from the manufacturer's decision not to perform the testing required by the Food and Drug Administration to license their product as suitable for PLT warming.^a Given that the current practice of many anesthesiologists is to warm PLTs despite the AABB's recommendation, the goal of this study was to determine if warming PLTs in a routinely used blood warmer had any effect on their function.

Methods

This protocol was approved by the Total Quality Council, a division of the IRB of the University of Pittsburgh. Ten units of three day old PLT rich plasma-derived whole blood PLTs were acquired from the Central Blood Bank (Pittsburgh, PA). Five mL aliquots were taken from each unit before warming and served as the control samples. The temperature of the units was measured immediately before warming. The units were then individually drained into a blood warming device (Ranger Blood/Fluid Warming System, Arizant Healthcare Inc, Eden Prairie, MN) and held in the disposable for two minutes. Since the volume of each PLT unit was approximately 50 mL, the warming disposable had to be altered to accommodate the small volume. This was accomplished by removing all of the extraneous tubing as well as the air trap. This particular warming device will warm fluid up to 42°C and is Food and Drug Administration-approved for warming packed red blood cell units and plasma. Post-warming samples were collected when the PLTs emerged from the effluent end of the warming device (warmed samples) and their temperature measured.

PLT aggregometry (PAP-4, Bio/Data Corporation, Horsham, PA) with three different agonists was performed on both the control and the warmed samples: adenosine diphosphate at $20\mu\text{M}$, $10\mu\text{M}$, and $5\mu\text{M}$ concentrations; collagen at 1.9mg/mL ; and arachidonic acid at 5mg/mL . The maximum amplitude of the clot was measured using thromboelastography (TEG®) on 6 of the 10 PLT units using kaolin as an activator in a Thrombelastograph Hemostasis Analyzer (Model 5000, Haemonetics, Braintree, MA).

^aE-mail communication between Jonathan Waters and Kayne Ryan, US Marketing Manager of Smiths Medical, the company which now produces the Level 1(R) rapid transfuser. August 7th, 2012

A study size of ten PLT units was selected as pilot data to determine the normality of the data and standard deviations of the measurements. A power analysis was performed, showing that at least 300 units of PLTs would have to be tested to achieve 80% power to detect the small differences in the observed means. It was decided to report the data from the ten units, and not divert an additional 300 units of PLTs, a scarce resource, away from patient care. The results of the control and warmed samples were compared with an unpaired two-tailed t-test after all of the data distributions were shown to be normal using the Shapiro-Wilk test.⁴ A P-value less than 0.01 was considered statistically significant. All statistical analysis was performed using GraphPad Prism software (Version 5.04, GraphPad Software Inc., San Diego, CA).

Results

The mean temperature of the 10 control and the warmed samples was $22.4 \pm 0.5^{\circ}\text{C}$ and $37.8 \pm 2.3^{\circ}\text{C}$, respectively. Results of aggregometry and TEG® tests are shown in Table 1 and Figure 1. There was no significant difference ($P = 0.13$) between the control and warmed samples in any of the PLT aggregometry tests, or in the TEG® maximum amplitude measurement. The observed mean of only one parameter decreased (PLT aggregometry with $5 \mu\text{M}$ adenosine diphosphate), by 5% (95% CI: -115% to 105%). The maximum change observed was PLT aggregometry with arachidonic acid as agonist, which increased by 116% (95% CI: -91% to 323%).

Discussion

In our study, a maximum decrease of 5% (95% CI: -115% to 105%) was seen in PLT function as measured by PLT aggregometry and TEG® between the unwarmed control and the warmed samples, although the inter-test variability was quite large for both the unwarmed and warmed PLTs. PLT aggregation becomes abnormal during storage,^{5,6} and our results suggest that warming the PLTs with this device does not cause further degradation in their function. However, a larger sample size would be required to definitively establish whether warming induced further degradation of PLT function. Also, as activated PLTs contribute to the pathogenesis of transfusion-related acute lung injury,⁷⁻¹⁰ perhaps through the formation of neutrophil extracellular traps,¹¹ studying the impact of transit time through the warmer, flow rate and temperature on the activation status of the warmed PLTs will also be important in determining their safety.

Some previous clinical studies which compared post-transfusion PLT counts demonstrated higher PLT counts (corrected for the dose of transfused PLTs and the patient's body surface area) after transfusion of warmed PLTs,¹²⁻¹⁴ while others showed no difference in counts.^{15,16} The suggested mechanism for the improvement is that the spherical shape that the PLTs tend to adopt during routine storage at room temperature is reversed upon warming, thereby facilitating longer *in vivo* survival and higher counts.^{17,18}

Warming of PLTs before transfusion in the operating room will help to reduce the risk of hypothermia in the recipient. Typically, transfusing 4-5 PLT units has a volume of approximately 250-300 mL, which is similar to the volume of a single donor (apheresis) PLT unit. This volume is also approximately the same as that of a unit of red blood cells, or 5-6% of the circulating blood volume of the average patient. Hence by mass and temperature balance, assuming a total circulating blood volume of 5L and assuming the specific heat of PLT units are the same as circulating blood, transfusing a 5-unit whole blood PLT dose which is stored at 22°C into a patient with a core body temperature of 37°C would decrease the temperature of the circulating blood volume by almost a full degree, to 36.1°C .

A limitation of our study is the small sample size. However, after analysis of the data from the first ten PLT units, and the lack of a clear difference in any of the measured variables, the decision was made to terminate the study so as not to divert more PLT units away from patient care. It is also possible that warming the PLTs caused damage to the PLTs in a way that was not measured in our tests; however, PLT aggregometry is the “gold standard” for PLT function measurement and no difference between the control and warmed samples was observed. Our *in vitro* findings, as well as those from future *in vitro* studies as outlined above, can help inform the design of a clinical trial whereby the safety and efficacy of warmed PLTs are evaluated in patients undergoing surgery.

The purpose of this study was to analyze the effect of warming on PLT function. As no significant decrease was observed in any of the measured variables after warming in this small study, we suggest that the current practice of warming the PLTs before infusion to an intraoperative patient is safe, although studies on PLT activation after warming are needed.

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References

1. Kurz A, Sessler DI, Lenhardt R. Perioperative normothermia to reduce the incidence of surgical-wound infection and shorten hospitalization. Study of Wound Infection and Temperature Group. *N Engl J Med.* 1996; 334:1209–1215. [PubMed: 8606715]
2. Guidelines for the use of blood warming devices: 2000–2001 Scientific Section Coordinating Committee. Bethesda, MD: American Association of Blood Banks; 2002.
3. Thomas D, Wee M, Clyburn P, Walker I, Brohi K, Collins P, Doughty H, Isaac J, Mahoney PM, Shewry L. Blood transfusion and the anaesthetist: management of massive haemorrhage. *Anaesthesia.* 2010; 65:1153–1161. [PubMed: 20963925]
4. Diehr P, Martin DC, Koepsell T, Cheadle A. Breaking the matches in a paired t-test for community interventions when the number of pairs is small. *Stat Med.* 1995; 14:1491–1504. [PubMed: 7481187]
5. Norfolk DR, Ancliffe PJ, Contreras M, Hunt BJ, Machin SJ, Murphy WG, Williamson LM. Consensus Conference on Platelet Transfusion, Royal College of Physicians of Edinburgh, 27–28 November 1997. Synopsis of background papers. *Br J Haematol.* 1998; 101:609–617. [PubMed: 9674730]
6. Rinder HM, Murphy M, Mitchell JG, Stocks J, Ault KA, Hillman RS. Progressive platelet activation with storage: evidence for shortened survival of activated platelets after transfusion. *Transfusion.* 1991; 31:409–414. [PubMed: 1710840]
7. Caudrillier A, Looney MR. Platelet-neutrophil interactions as a target for prevention and treatment of transfusion-related acute lung injury. *Curr Pharm Des.* 2012; 18:3260–3266. [PubMed: 22621270]
8. Corash L, Lin JS, Sherman CD, Eiden J. Determination of acute lung injury after repeated platelet transfusions. *Blood.* 2011; 117:1014–1020. [PubMed: 20935256]
9. Middelburg RA, Borkent-Raven BA, Janssen MP, van de Watering LM, Wiersum-Osselton JC, Schipperus MR, Beckers EA, Briët E, van der Bom JG. Storage time of blood products and transfusion-related acute lung injury. *Transfusion.* 2012; 52:658–667. [PubMed: 21950582]
10. Shaz BH, Stowell SR, Hillyer CD. Transfusion-related acute lung injury: from bedside to bench and back. *Blood.* 2011; 117:1463–1471. [PubMed: 20944069]
11. Caudrillier A, Kessenbrock K, Gilliss BM, Nguyen JX, Marques MB, Monestier M, Toy P, Werb Z, Looney MR. Platelets induce neutrophil extracellular traps in transfusion-related acute lung injury. *J Clin Invest.* 2012; 122:2661–2671. [PubMed: 22684106]

12. Fijnheer R, Pietersz RN, Huijgens PC, de Korte D, Roos D, Reesink HW. Beneficial effect of pre-transfusion warming of platelets prepared from buffy coats. *Lancet*. 1990; 335:1524. [PubMed: 1972448]
13. Hutchinson RE, Kunkel KD, Schell MJ, Jackson CW, Nelson EJ, Wang WC, Fischl SJ, Taylor LB, Garcez RB, Pui CH. Beneficial effect of brief pre-transfusion incubation of platelets at 37 degrees C. *Lancet*. 1989; 1:986–988. [PubMed: 2565517]
14. Shanwell A, Wikman A, Ringden O. Pretransfusion incubation of apheresis platelets at 37 degrees C improves posttransfusion recovery. *Transfusion*. 1992; 32:715–718. [PubMed: 1412678]
15. Hussein MA, Schiffer CA, Lee EJ. Incubation of platelet concentrates before transfusion does not improve posttransfusion recovery. *Transfusion*. 1990; 30:701–703. [PubMed: 2219256]
16. Slichter SJ, Christoffel T, Corson J, Jones MK, Pellham E, Bolgiano D. Effects of pretransfusion warming of platelets to 35 degrees C on posttransfusion platelet viability. *Transfusion*. 2009; 49:2319–2325. [PubMed: 19624609]
17. Fijnheer R, Pietersz RN, de Korte D, Roos D. Monitoring of platelet morphology during storage of platelet concentrates. *Transfusion*. 1989; 29:36–40. [PubMed: 2492127]
18. Slichter SJ, Harker LA. Preparation and storage of platelet concentrates. II. Storage variables influencing platelet viability and function. *Br J Haematol*. 1976; 34:403–419. [PubMed: 10956]

Figure 1.
Platelet aggregometry and thromboelastography maximum amplitude (TEG® MA) results for the platelet units

Table 1

Platelet aggregometry and thromboelastography (TEG®) results for the control and warmed samples, reported as mean $\pm$ SD and difference between means $\pm$ SD [99% confidence intervals]. P values are for unpaired two-tailed T test. The maximum change in observed means was a 116% increase in platelet aggregometry with arachidonic acid as agonist (95% CI: -91% to 323%). The only observed decrease was platelet aggregometry with 5 μ M adenosine diphosphate (ADP) as agonist, the mean of which decreased by 5% (95% CI: -115% to 105%); MA = maximum amplitude

	Control	Warmed	Difference between means	p
20 μ M ADP (%)	10.0 $\pm$ 7.7	11.3 $\pm$ 8.0	1.3 $\pm$ 3.5 [-8.8 to 11.4]	0.71
10 μ M ADP (%)	6.9 $\pm$ 5.1	7.5 $\pm$ 6.2	0.6 $\pm$ 2.5 [-6.7 to 7.9]	0.82
5 μ M ADP (%)	4.0 $\pm$ 3.2	3.8 $\pm$ 3.6	-0.2 $\pm$ 1.5 [-4.6 to 4.2]	0.90
Collagen (%)	50 $\pm$ 17	56 $\pm$ 22	6 $\pm$ 9 [-20 to 31]	0.52
Arachidonic Acid (%)	8.2 $\pm$ 9.1	17.7 $\pm$ 16.4	9.5 $\pm$ 5.9 [-7.5 to 26.5]	0.13
TEG® MA (mm)	66.4 $\pm$ 9.0	68.0 $\pm$ 8.7	1.5 $\pm$ 5.1 [-14.7 to 17.8]	0.77