Similar donors—similar blood?

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BACKGROUND: Red blood cell (RBC) storage lesions have been suggested as contributing factors to suboptimal clinical outcomes. While undesirable effects of storage are well documented, their clinical relevance is still debated. Focus on storage time as the sole determinant of RBC quality ignores the variability in cell properties that may depend on factors other than age. Mechanical fragility (MF) aggregately reflects many storage-related functional and structural changes. This study evaluates interdonor versus intradonor variability, throughout storage, of both MF and autohemolysis (AH).

STUDY DESIGN AND METHODS: Thirteen uniformly manufactured RBC units were collected initially as whole blood from nonsmoking, group A+, male Caucasian research donors. Mechanical stress was applied using a bead mill with oscillation at 50 Hz over durations varying from 0.5 to 60 minutes. MF profiles were described in terms of percent hemolysis after stresses of specified durations. Two months later, 11 of the 13 donors returned and assays were performed using the same protocol to allow comparison of intradonor versus interdonor variation.

RESULTS: At 5 days postcollection, RBC MF profiles exhibited marked interdonor variability (up to twofold) overall. Both autolysis and MF across all units increased during storage—with rates of these increases varying by up to 10-fold for certain MF variables. Especially high AH and MF were observed for an outlier donor (with $p < 0.05$), for whom follow-up revealed previously undisclosed hereditary hypertriglyceridemia (levels exceeding approx. 1000 mg/dL).

CONCLUSIONS: RBCs, even from similar donors, vary significantly in levels and changes of both AH and MF, the clinical significance of which must still be ascertained. While further study is needed, donors with severe hypertriglyceridemia may not be appropriate as blood donors due to the unacceptable level of hemolysis observed during storage of our affected study subject.

Modern-day blood banking and transfusion services were made possible by the development of effective blood collection, processing, and storage methods. However, degradation during storage is increasingly suspected as contributing factors to posttransfusion complications and suboptimal clinical outcomes. Undesirable effects of storage lesion on red blood cells (RBCs) are well documented; however, the extent of their clinical relevance is still a subject of considerable debate.[1,2] Notably, the current discussion predominantly focuses on product age—that is, the question whether "new" blood provides better transfusion outcomes than the "old" blood.[1-4] This focus on the RBC storage time (ST) as the sole quality metric is exemplified by the common use of first in, first out in blood bank inventories and by being the centerpiece of numerous clinical studies including ABLE,[5] RECESS,[6] and ARIPI.[7,8]

However, focusing on ST as the sole marker of storage lesion, and thus on cells' viability and prospective transfusion outcomes, ignores the variability in RBC properties that depend on factors other than age. Early reports indicated that some donors' RBCs may be poorly storable,[9] and interdonor variability had been identified as one of the major determinants in variability of RBC posttransfusion ^{51}Cr 24-hour in vivo recovery.[10,11] Individual donor variability in ATP levels had also been reported.[12] Part of the reason for such interdonor variability may be differences in metabolic age of donor RBCs upon

ABBREVIATIONS: AH = autohemolysis; Hem = hemolysis (as a variable); MF = mechanical fragility.

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collection,[13,14] although other donor-specific factors could be involved. Such factors may include metabolomic changes in RBCs that occur during storage.[15] The issue is further complicated by the RBC properties' dependence on component manufacturer.[16] A recent study by Pieracci and colleagues,[17] which tracked the performance of more than 100 transfused units, reported a small ($r = 0.18$) but significant ($p = 0.04$) correlation between the duration of storage of transfused blood and the increase of posttransfusion patient hematocrit (Hct). At the same time, the study demonstrated potentially more significant variability in the incremental increase in Hct between RBC units of the same age. Overall, inherent unit-to-unit differences are likely to be a significant confounder to ST as an indication of transfusion efficacy—a fact that can have significant implications not only to transfusion practice, but also to regulation of RBC products.[18]

Mechanical fragility (MF)[19,20] and related flow properties[21,22] have been proposed as a more physiologic candidate to supplement ST as an aggregate metric of RBCs' functional and structural storage lesions. It was hypothesized that MF would vary significantly among RBC units in part due to interdonor differences and that such variability will be further exacerbated by blood storage. This study aims to evaluate interdonor variability throughout storage of both MF and autohemolysis (AH) of packed stored RBCs.

MATERIALS AND METHODS

Donor recruitment and RBC collection

Under institutional review board approval, 13 paid research donors between the ages of 24 and 59 were recruited and consented to participate in this study. All donors recruited were Caucasian males of the same blood group (A+) to avoid any contribution of race, different blood groups, or sex into variation in RBC fragility as measured by the MF assay. To avoid confounding the results by the potential effects of smoking on RBC membrane properties,[23] only nonsmoking donors were selected.

A whole blood unit was collected from each research donor in a CPD-A2 bag. Nonleukoreduced RBCs were prepared per standard protocol from each whole blood unit by centrifugation. The RBCs were placed in the refrigerator at 1 to 6°C within 30 minutes of centrifugation. The units were shipped overnight on ice to Dr Tarasev's laboratory in Ann Arbor, Michigan.

After approximately 2 months, 11 of the 13 research donors returned and donated a second unit of whole blood. The second units were collected and prepared using the same processes as the initial units.

MF test

Samples were sterilely obtained from the RBCs at 5 days postcollection and again after 14, 28, and 42 days of storage (2, 4, and 6 weeks postcollection). These samples were diluted to a total hemoglobin (Hb) concentration of 1.6 to 1.7 g/dL (corresponding to approx. 4% Hct) using a HbB system from HemoCue (Ängelholm, Sweden) with AS-3 storage buffer, pH 5.75, containing 40 g/L albumin (Sigma, St Louis, MO). The diluted sample was gently agitated and aliquotted into 2-mL low-retention centrifuge tubes at 330 µL per tube. Mechanical stress was applied to RBC samples with the use of a vertical bead mill (TissueLyser LT, Qiagen, Hilden, Germany) (at an oscillation oscillation frequency of 50 Hz) in the presence of one 7-mm-diameter stainless-steel ball for a predetermined duration. Samples from each RBC unit were subjected to such stress at 10 different durations (ranging from 30 sec to 60 min) to ensure a wide range of achieved hemolysis levels. The sample holder of the TissueLyser was modified to allow air cooling while in operation, which resulted in sample temperature stabilization to within 2 degrees of the 22°C start temperature. Unlysed cells were sedimented by centrifuging the samples for 5 minutes at $157 \times g$ on a centrifuge (Model 5417C, Eppendorf, Hamburg, Germany). Supernatant samples were collected and used for spectral analysis.

Hemolysis assessment

Hemolysis (Hem), both in untreated sample (AH) and as induced by the bead mill, was determined based on the difference in absorbance at 576 nm, a wavelength of oxygenated Hb maximum, and absorbance at 700 nm. It was expressed as a fraction of free Hb (Hb^F) relative to total Hb concentration (Hb^T) according to Equation (1), which included the correction for sample Hct as detailed by Sowemimo-Coker.[24]

$$Hem = \frac{Hb^F_{576} - Hb^F_{700}}{Hb^T_{576} - Hb^T_{700}} * (1 - Hematocrit). \quad (1)$$

Total Hb concentration for each diluted RBC sample was determined by subjecting a small (30-40 µL) aliquot to repeated (5×) rapid freeze-thaw using liquid nitrogen. In control experiments, such treatment was shown to fully lyse RBC. For AH (hemolysis before the application of mechanical stress) determination, small (20 µL) samples of undiluted segments' content were centrifuged for 5 minutes at $157 \times g$, supernatants were collected, and Hb content was measured spectrophotometrically. Spectroscopic measurements were performed with a spectrophotometer (Model N100, NanoDrop, Wilmington, DE).

RBC fragility profiles

RBC fragility profiles are defined here as the cumulative incremental (beyond AH) hemolysis resulting from applied stress of varying durations. Unlike single-point measurements that use a single stress duration at a single

stress intensity, as implemented, for example, by Raval and colleagues,[20] MF profiles allow ascertainment of the propensity for RBCs to hemolyze over a range of applied stress magnitudes. These include levels of duration and/or intensity resulting in minimal hemolysis, ranging up to those resulting in nearly total hemolysis of cells in the tested sample—thereby allowing multiple fragility-based indexes to be interpolated for separate analyses.[19] Here, profiles were described by the hemolysis variables (Hem variables) representing the extent of hemolysis achieved as a result of small (1-2 min), medium (5-10 min), and large (30-60 min) stress durations identified by a subscript number for the Hem variable, at a fixed stress intensity, thus corresponding to overall small, medium, and large total applied stress magnitude. Fragility variables at particular stress durations were obtained from best fit second-order polynomial regression to the experimental data. For curves exhibiting significant deviations from a simple polynomial, raw data were subdivided into low and high hemolysis subsets and the fits were obtained independently for each subset of the data.

Statistical analysis

Data are presented in the form of mean ± standard deviation (SD), range, and coefficients of variability (CV) where appropriate. Student t testing with a two-tailed p value of 0.05 was used to test for significance.

The longitudinal data collected in the study were described by a linear mixed model (Eq. 2), which combines subject-independent (fixed) and subject-specific (random) variables. Such an approach is better suited for describing longitudinal data than ordinary linear regression. Ordinary linear regression would underestimate the variance of regression variable estimates (thus overestimating model's significance) due to correlation between repeated measurements from the same subject.

$$\mathbf{Y} = \beta_0 + \beta_1 \times \text{time} + \beta_2 \times \text{Hem}_{1,5j} + \beta_3 \times \text{Hem}_{5,5j} + \gamma_{1j} + \gamma_{2j} \times \text{time} + \varepsilon_{ij}. \quad (2)$$

Here the outcome $\mathbf{Y} = \{y_{ij}\}$, $i = 1, \ldots, p$; $j = 1, \ldots, n$ is the data vector of any selected response variable with components of response measured on n subjects at p times; β_0 is the fixed-effects intercept; β_1 is the fixed coefficient assuming time as a linear predictor; β_2 and β_3 are fixed-effects variables for $\text{Hem}_{1,5j}$ and $\text{Hem}_{5,5j}$, MF measured at Day 5 for Patient j; γ variables introduce donor-specific component with γ_{1j} standing for the random intercepts and γ_{2j} for random slopes with the index j defined as above. ε_{ij} is the error vector (or residuals). Specific response variables evaluated by this model included Hem_0 (AH value) and Hem_1 and Hem_5 (MF variables associated with small and medium amounts of total applied stress). The limited number of observations allowed constructing models with only a minimal number of variables describing "intradonor" effects. Selection and fitting of the best fit for each response variable was performed as recommended by Littell and colleagues[25] with the estimation of variance components performed using the model with random intercept and random slope only (see SAS Institute[26]). For ease of interpretation, fixed-effects variables were centered: time by its value at first measurement (Day 5) and induced hemolysis variables by their mean values. Donor 5 was excluded due to an underlying medical condition (see discussion to follow) established at poststudy check-up. The analysis was performed using computer software (SAS 9.2, SAS Institute, Cary, NC).[26]

RESULTS

Donors

All donors were nonsmoking males, aged 24 to 59 years, of the same blood type, and qualified to donate based on the current AABB and FDA eligibility criteria. No existing pathology or chronic condition was disclosed at the time of donation and all donors appeared to be in good health. Units and samples were prepared from whole blood donated by 13 donors (11 of which donated again in 2 months) to ensure maximal uniformity. Total Hb concentration in collected RBC units was 20.6 ± 0.7 g/dL.

AH

Thirteen RBC units varied in their 5-day AH with the coefficient of variability (CV) of 0.58. Exploratory analysis of the results indicated that RBCs collected from one of the subjects (Donor 5) exhibited unexpectedly high AH—up to 4% at the end of storage. The CV of the remaining 12 units remained unchanged after 42 days of storage, with CV of 0.43 both on Day 5 and at 6 weeks postcollection (Table 1). While all Hem_0 values increased with ST, the actual donor-to-donor rates of change, expressed in percentage of Hem_0 per day, varied significantly among donors (CV, 0.96 for 13 subjects; CV, 0.53 excluding the "Donor 5" outlier).

The rate of change in Hem_0 was expressed as percent hemolysis per week. For the outlier unit, this rate was significantly different from the mean: Donor 5 at 7.7×10^{-3} compared to a mean of 1.45×10^{-3} percent hemolysis per week based on all 13 units. This difference was significant with p values less than 0.0001. The AH value on Day 5 postcollection for this unit was close to the mean of all 13 units. However, as a result of significantly increased rate of change, this unit also exhibited a significant difference in the Hem_0 values on Day 42 of storage compared to the mean Hem_0 value (Table 1 and Hem_0 panel of Fig. 1).

Linear mixed models can be used to describe the longitudinal data, as detailed under Materials and Methods. In this study, collected RBC units were tested repeatedly

TABLE 1. AH and induced hemolysis values

	Induced hemolysis					
	Hem_0 (AH)		Hem_1 (low stress)		Hem_5 (medium stress)		Hem_{30} (high stress)	
Parameters	Mean ± SD (range)*	Unit 5	Mean ± SD (range)*	Unit 5	Mean ± SD (range)*	Unit 5	Mean ± SD (range)*	Unit 5
5 days (hemolysis, %)	0.07 ± 0.03 (0.03-0.14)	0.185	6.8 ± 1.7 (4.8-10.2)	6.8	15.2 ± 4.6 (11.6-25.5)	18.4	38.7 ± 4.7 (29.4-47.3)	53.1
42 days (hemolysis, %)	0.89 ± 0.38 (0.37-1.65)	4.25	11.1 ± 2.8 (6.8-15.7)	18.7	19.2 ± 2.8 (14.2-23.8)	38.3	39.0 ± 4.1 (32.3-47.7)	60.2
Rate of change† (% hemolysis /week)	0.15 ± 0.08 (0.06-0.28)	0.77	0.81 ± 0.44 (0.21-1.68)	2.3	0.71 ± 0.69 (–0.28 to 1.82)	3.6	0.02 ± 1.3 (–2.4 to 2.5)	0.56

* Mean ± SD and range data are based on 12 units, excluding the Donor 5 outlier.
† Rate of change was calculated from a best-squares linear fit to experimental data collected at 5 days and 2, 4, and 6 weeks of storage.

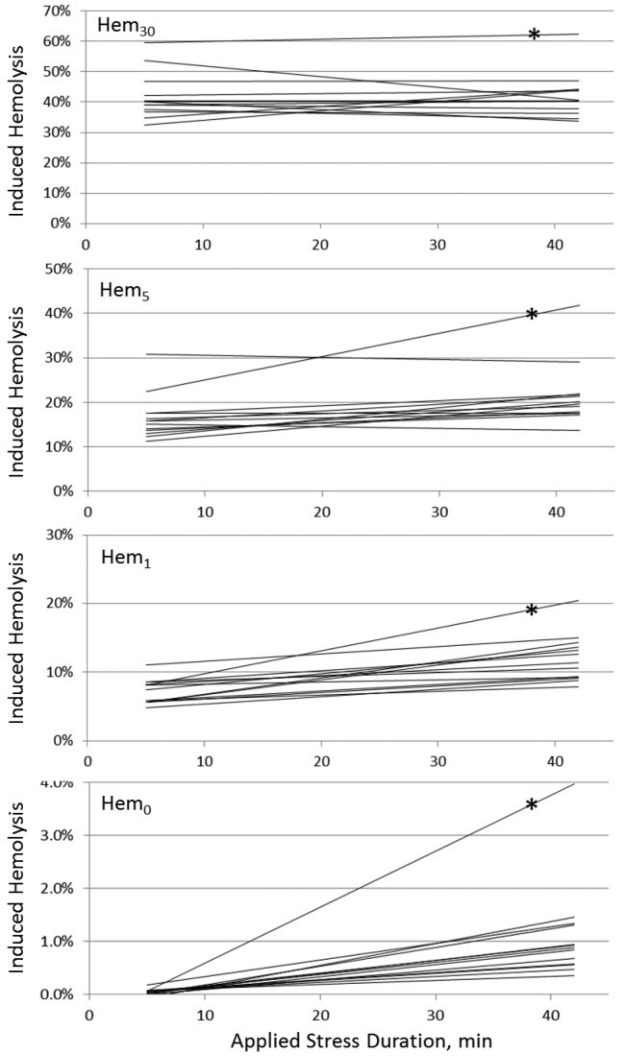

Fig. 1. Changes in AH (Hem_0) and induced hemolysis (Hem_1, Hem_5, and Hem_{30}) as a function of ST. Presented are linear trend lines for the experimental data from the 13 donors measured at 5, 14, 28, and 42 days after blood collection. *Trend lines calculated from the Donor 5 data.

over time. Although the individuals themselves are independent of each other, observations obtained from the same donors are correlated by sharing characteristics from the same subject. Linear time was a significant fixed-effect predictor of Hem_0. Variability of the rate of change in Hem_0 among subjects is included as a random effect (random slope). This allows reflecting the observed increase of variance over time. Inclusion of Hem_5 at baseline (5 days postcollection) as an additional fixed-effect predictor reduced the variance of unexplained residual by 30%—thus significantly improving the model. Estimated variables of the model are shown in Table 2. Controlling for other variables in the model, adjusted estimated mean values of Hem_0 at baseline ($\beta_0 = 0.06\%$) are similar to the

TABLE 2. Linear mixed model for longitudinal changes in Hem variable

Model coefficients	Hem_0	Hem_1	Hem_5
Variance of random effects			
Intercept (γ_1)	NA	0.730	5.958
Time (γ_2)	0.004	0.106	NS
Residual (ε)	0.012	1.440	9.029
Fixed model variables			
Intercept (β_0)	0.060	7.134	16.081
Time (β_1)	0.153	0.795	0.694
Hem_1 at baseline (β_2)	NS	NS	1.675
Hem_5 at baseline (β_3)	0.029	0.371	NS

All variable estimates are restricted maximum likelihood estimates with $p < 0.05$.
NA = not applicable; NS = not significant.

observed mean values of Hem_0 on Day 5 postcollection (0.07%). The rate of change of Hem_0 over time varied across individual donors, with the adjusted mean β_1 of 0.16% (SD, 0.064%) per week, which is similar to the observed values of 0.15 ± 0.08% hemolysis per week.

MF

Variables describing RBC MF at low (Hem_1), medium (Hem_5), and large (Hem_{30}) applied stress varied significantly among the donors (Table 1). At 5 days postcollection, Hem variables exhibited up to twofold interdonor variability. This variability increased over time, with the increase more pronounced for Hem variables associated with lower applied stress magnitudes. Mean MF values also increased over the ST, with such changes being progressively less pronounced for those MF variables involving higher overall stress magnitude (Fig. 1). Statistically, relatively small observed storage time–dependent changes in Hem_{30} values were not significantly different from zero.

The full scope of MF changes during RBC storage is described by both the absolute values of the MF variables and the rates of their change over time. Donor 5's unit, which exhibited abnormally high AH values, also exhibited significantly ($p < 0.0001$) elevated MF variables (except as measured by Hem_1 at 5 days postcollection; Table 1), as well as the rates of change of said variables. None of the other units exhibited changes that were as pronounced as that by Donor 5 (see Fig. 1).

Linear time remained a significant fixed-effect predictor for MF variables Hem_1 and Hem_5. However, for smaller stress, represented by the variable Hem_1, variability in rates of change in MF fragility over time among subjects was much higher than for larger stress as represented here by the variable Hem_5. Thus for the low-stress variable, the model includes random intercepts and slopes, while for medium stress the variance of the slope does not have significance (Table 2). Addition of another Hem variable measured at collection as a fixed-effects predictor improved the models. In particular, for Hem_1 it reduced the variance in unexplained residuals by 27% and in random-effect intercept by 74%, while for Hem_5 the variance in random effect intercept was reduced by 59%. Reduced variance of random effects indicates a better fit when fixed-effects variables are included in the model.

Controlling for other variables in the model, adjusted mean values of Hem_1 (7.13%) and Hem_5 (16.08%) measured at collection (β_0) varied among the donors, with SDs of 0.85 and 2.44%, respectively. These are similar to the experimentally observed values for Hem_1 and Hem_5 (11.1 ± 2.8 and 19.2 ± 2.8%, respectively; see Table 1). The rate of change of Hem_1 differs for each donor, with the adjusted mean of 0.79% (SD, 0.33%) per week, while the rate of change of Hem_5 is approximately the same for all the donors, with the adjusted mean of 0.69% hemolysis per week. These values correspond well with the experimentally observed means of the rate of change of 0.81 and 0.71% per week for Hem_1 and Hem_5, respectively. Overall correspondence of observed values with those derived from the mixed-effects model indicate that such model adequately describes experimentally observed changes and variability in AH and MF over ST.

Second draw

To evaluate interdonor versus intradonor contributions to the variability of MF properties of stored RBCs, 11 donors (of the original 13) donated blood again approximately 2 months after the original draw. Overall, the mean values of AH (Hem_0) and MF variables (Hem_1, Hem_5, and Hem_{30}) and their variability at 5, 14, 28, and 42 days postcollection were similar between the two draws. Notably, Donor 5 remained a marked outlier in both AH and MF. Mean values, however, do not account for possible significant differences between the draws for individual donors (see Fig. 2). When comparing the values of Hem variables at the beginning of storage (Day 5 postcollection) and their rates of change over the 42-day storage, paired t test analysis shows that Hem_1 was the variable most varied between the draws. It remains to be determined what might be the underlying causes for this observation; likely such inquiry will require larger donor selection combined with observation over a larger number of repeated donations, as well as tracking additional factors, which could potentially affect RBC properties (donor demographic, blood physiology, etc.). While rigorous statistical analysis of intradonor variability is not possible on only two data points per donor (two blood draws), it can be noted that overall, some donors appeared to exhibit a noticeable difference in AH and MF between the two draws, while for many (six of 10, outlier excluded), such differences were minimal for all variables.

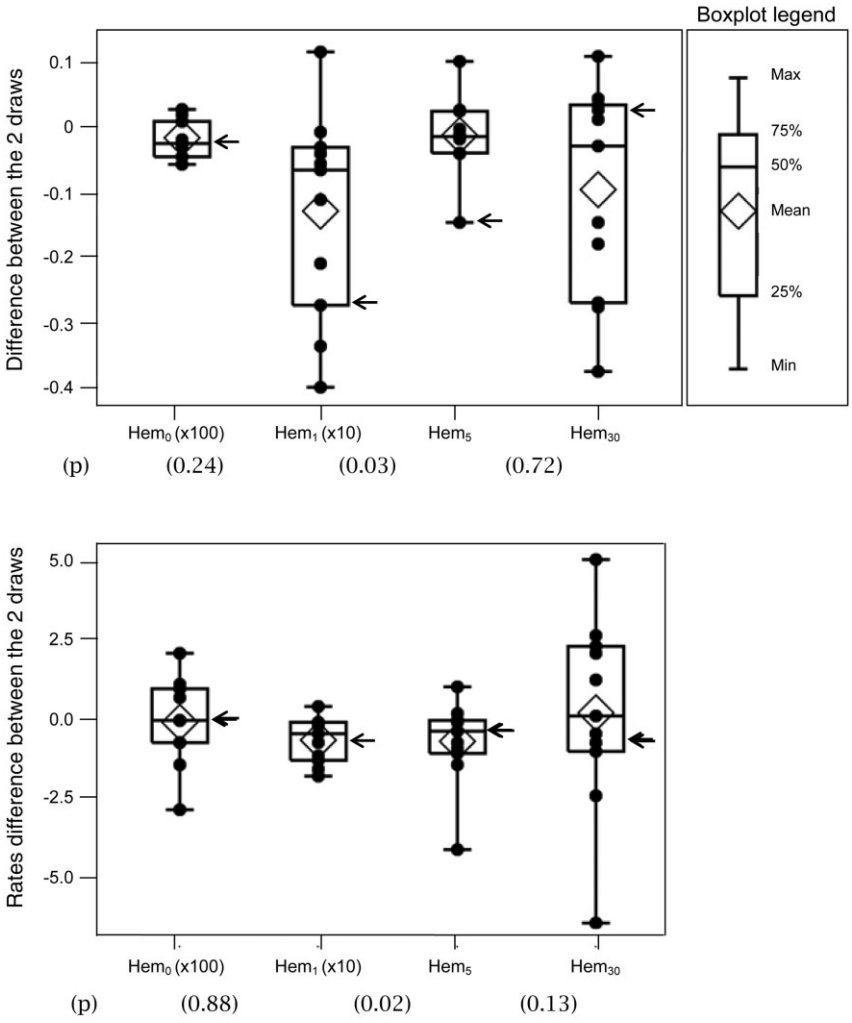

Fig. 2. Box and whiskers plot of the difference between the two blood draws. Difference between the values at Day 5 postcollection (A) and in the rates of change over time (in percent per week); (B) for AH (Hem_0) and induced hemolysis (Hem_1, Hem_5, and Hem_{30}). Arrows indicated the values from the Donor 5. Paired t test p values are shown below the relevant variable.

DISCUSSION

RBC MF is gaining acceptance as a useful tool for assessing clinically relevant cell properties, with common approaches using a single value often called a MF index to describe MF properties of RBC units.[20,27,28] This study used a multipoint profile, which allowed the calculation of multiple MF-defining variables from each RBC sample. These MF variables—reflecting small, medium, and large magnitudes of applied stress—all varied significantly among the donors, even though effort was made to make the RBC units as uniform as possible regarding manufacturing and storage methods as well as donor base. Such variability is likely to be even larger in a typical blood bank inventory sourced from different collection, processing, and storage modalities and a much more diverse donor pool. While the clinical relevance of such variability remains to be demonstrated, it can be anticipated that units with markedly different RBC mechanical fragilities would behave differently in vivo, potentially resulting in different RBC recovery rates and overall efficacy.

Determination of MF profiles allows inferring of multiple MF variables (indexes) to characterize each unit's response to MF stress. Variables Hem_1, Hem_5, and Hem_{30} (responsible for small, medium, and large stress) were calculated in this study. Interestingly, while lower-stress variables were found to be better at describing MF changes due to prolonged blood storage, higher-stress variables were better at discriminating the outlier unit (Donor 5) at Day 5 after collection. This indicates that different profile-derived variables may indeed reflect different RBC membrane-related attributes and thus have potentially different relevance for future clinical applications.

A mixed-effect model (Eq. 2) framework was used to describe longitudinal data collected in the study. It allowed identifying and describing the characteristics of longitudinal behavior of both AH and induced hemolysis variables. Linear time, as expected, was a significant fixed-effect predictor for both AH and induced hemolysis variables. Inclusion of induced hemolysis variables measured at 5 days postcollection as correlated allowed for improved fit of mixed-effect models.

Surprisingly, the significance of subject-specific effects varied. Within the model framework, for AH, represented by Hem_0, the variability of hemolysis at collection was not significant. Variability was explained by variable Hem_5 measured at that time, and the rate of change was different for each subject. For the small induced-stress variable, Hem_1, both the variance of values at collection and that of the rate of change over time were significant. For the medium induced-stress variable (Hem_5), only the variance of the starting value had significance, meaning that at baseline each subject had its own starting value of Hem_5, and with time in storage it increased for all subjects at about the same rate. This analysis shows the promise of the utilized approach and indicates what information would likely be required to make predictions about future behavior of AH and MF variables.

Observed AH levels were on average higher that that reported previously,[29] with the difference likely due to the use of nonleukoreduced RBCs in this study[30] and possible interdonor variability in hemolysis levels.[31] Consistent with the previously reported data,[31] interdonor variability was found to be a factor of AH due to significant variance in its rates of change among the individual subjects. Note that in this study we avoided additional sources of variability by using a uniformly processed single ABO group blood from only male sex donors, in an attempt to isolate donor-specific effects. While donor specificity may be a factor for all AH-related variables, additional potential correlates may independently affect postcollection values and RBC stability while in storage. Overall, even within a small group of similar donors some donated using exhibited marked changes in their AH and MF variables after 42 days, compared to that at collection, while others were only minimally affected by the storage.

Current guidelines for RBC hemolysis provide a limit of 1% in the United States and 0.8% in Europe to avoid transfusion of hemolytic products[32] as such products can have detrimental effects on transfused patients.[33,34] At the same time, donor-dependent variability in AH had been previously reported[31] with the authors further stating that some donor blood exhibited particularly "poor storage characteristics."

Blood donors may donate many times a year, and it is possible that lifestyle and dietary changes may be reflected in both plasma composition and RBC properties.[35,36] However, potential intradonor changes between donations in the properties of donated blood remain obscure. The presented data indicate that while for many donors there are no significant changes in AH and MF RBC variables between the two consecutive donations, for some donors and variables that may not be true. Presented results should be considered with caution as two draws is insufficient for a proper statistical analysis. Furthermore, the 11 donors studied may not be representative of the whole blood donor community.

In this study an attempt was made to select donors similar to each other; however, the MF of donated blood was found to be significantly different. The reasons behind this variability remain to be investigated and will be the subject of the further studies. Of particular interest is the fact that blood collected from Donor 5 exhibited significantly elevated levels of AH and MF. While acceptable at 5 days according to the US 1% hemolysis standard, this RBC unit would have failed the standard after only 2 weeks of storage. This particular donor appeared in good health, answered negatively on all screening questions on the standard donor questionnaire on deferral health conditions and drug use, and would have been fully acceptable for routine blood donation. Based on the abnormal results observed, this donor later shared that his triglyceride level was 1056 mg/dL and he has since started on therapy for previously undisclosed hereditary hyperlipidemia. On therapy, his triglyceride level has dropped to 837 mg/dL. He appears to have isolated hypertriglyceridemia, as his cholesterol level is 177 and HDL level is 24. Fasting glucose was 158 with a Hb A1c f 7.8, suggesting that he has concomitant diabetes. He has been deferred from further blood donation.

Published data indicate a potential link between serum cholesterol (LDL/HDL) and triglyceride levels, and RBC membrane properties, including its deformability.[37,38] In addition, decreased RBC deformability was associated with triglyceridemia, cholesterolemia, and hyperglycemia in humans and in animal models.[39,40] Increased hemolysis, potentially due to increased RBC fragility, was reported in patients with hyperlipidemia and particularly with hypertriglyceridemia.[41] However, to our knowledge this report is the first to observe markedly increased AH, concurrent with the increased MF, in a hypertriglyceridemic donor. While further study of this correlation is obviously warranted, these data indicate that individuals with severe genetic hypertriglyceridemia should potentially be excluded as blood donors due to high baseline hemolysis.

CONFLICT OF INTEREST

MT, KA, and SC are employed by Blaze Medical Devices, which develops clinical tests for measuring RBC fragility. KD and JBG have no conflict of interest to disclose.

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