Prospective Crossover Study of the Effect of Phlebotomy and Intravenous Crystalloid on Hematocrit

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ABSTRACT

Objective: To compare the changes in hematocrit (Hct) between phlebotomized and nonphlebotomized individuals given IV crystalloid.

Methods: A prospective, crossover volunteer study was performed comparing Hct changes immediately and 30 minutes after IV crystalloid bolus in 20 healthy adults with and without prebolus phlebotomy. In the control portion, volunteers were given a 15-mL/kg bolus of normal saline over 30 minutes with Hct determination before (H1), immediately after (H2), and 30 minutes after (H3) crystalloid infusion. At least 7 days later, the same subjects were phlebotomized 1 unit of blood and then administered a 15-mL/kg IV bolus of normal saline 30 minutes later. Hcts were obtained before (H4) and 30 minutes after (H5) phlebotomy (immediately prior to crystalloid infusion). Hcts were also obtained immediately after (H6) and 30 minutes after (H7) crystalloid infusion. A post-hoc test performance analysis was then performed to determine the Hct drop thresholds that would yield the maximal sensitivity and specificity for 500 mL of blood loss (via phlebotomy) in this population.

Results: The Hct (%) drops in the nonphlebotomized individuals receiving IV fluids averaged 4.5 ± 1.3 immediately and 3.2 ± 1.3 30 minutes after infusion. These drops were different ($p < 0.05$) from the Hct drop in individuals receiving IV fluids after phlebotomy, which averaged 6.6 ± 1.5 and 5.7 ± 1.1, respectively. Post-hoc analysis revealed that Hct drops of 5.4 immediately, or 4.3 at 30 minutes after infusion, had a sensitivity of >90% and a specificity of 75% for identification of patients in the phlebotomy group.

Conclusions: The practice of measuring serial Hcts may be helpful to identify trauma patients with occult blood loss. A prospective clinical trial is needed to validate these Hct drop thresholds (immediate and 30 minutes postinfusion) in crystalloid-resuscitated trauma patients.

Key words: hemorrhage; hematocrit; blood volume; crystalloid solution.

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Victims of blunt trauma and suspected internal bleeding are often evaluated with, among other modalities, serial hematocrits (Hcts) in an attempt to detect or monitor internal blood loss.[1–3] Patients with suspected blood loss are given IV crystalloid to maintain an adequate volume status. Previous studies[4,5] have demonstrated that infusion of IV crystalloid to healthy (and presumably euvolemic) adults results in a significant (4.5–6.3-point) drop in Hct, secondary to hemodilution.

This volunteer study was designed to assess whether the conventional practice of measuring serial Hcts in trauma patients to detect blood loss is valid and to suggest potential Hct drop thresholds (immediate and 30 minutes postinfusion of crystalloid).

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METHODS

Study Design: A prospective, volunteer crossover study was performed to evaluate the effect of phlebotomy and IV crystalloid infusion on Hct. This study was approved by the Institutional Review Board of Albany Medical Center, and informed consent was obtained from all volunteers prior to enrollment in the study.

Setting and Subjects: Infusion of IV crystalloid was performed in the ED of the Albany Medical Center Hospital, and phlebotomy was performed at the American Red Cross Donation Center in Albany, NY, from June 19, 1995, through October 24, 1995. Volunteers eligible for enrollment included healthy adults 18–65 years old who had not donated blood, had surgery, or suffered injury necessitating medical attention in the preceding 90 days. Standard Red Cross criteria for exclusion from phlebotomy also applied.

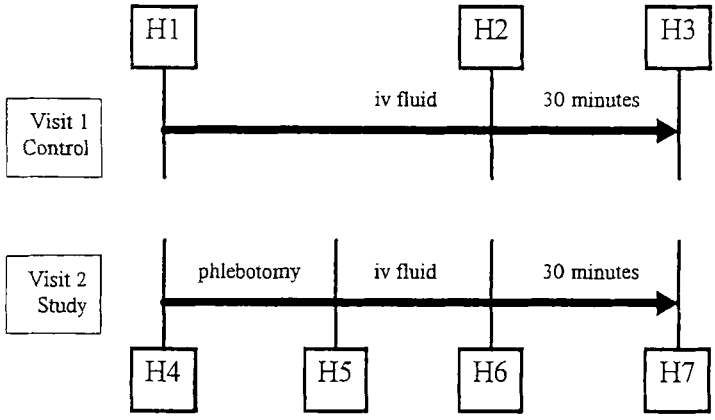

FIGURE 1. Study protocol timeline.

Experimental Protocol (Fig. 1): In the control portion of the study, a 20-gauge catheter was inserted into an upper-extremity vein and the first Hct (H1) was obtained. Fifteen mL/kg of 0.9% normal saline (NS) was then infused via pump over 30 minutes. Immediately following the bolus infusion, the second Hct (H2) was collected. After waiting 30 minutes for equilibration, the third Hct (H3) was obtained.

The experimental portion of the study took place ≥7 days later, when the volunteer returned, had a 20-gauge catheter inserted, and had the fourth Hct (H4) obtained. The subject was then phlebotomized 1 "unit" (about 500 mL) of blood. Thirty minutes after phlebotomy, the fifth Hct (H5) was collected and the patient was again given a 15-mL/kg bolus of NS over 30 minutes. Immediately and 30 minutes after the bolus, the sixth (H6) and seventh (H7) Hcts, respectively, were collected.

All the Hct specimens were obtained through the IV catheter after discarding the first 5 mL withdrawn. All the patients were semisupine and had nothing-by-mouth (NPO) orders during infusion of the 15 mL/kg IV NS bolus. All the patients were supine and had NPO orders during phlebotomy.

Measurements: Hct (%) was determined using a Coulter counter technique (Coulter STKS Analyzer, Coulter Corporation, Miami, FL, coefficient of variance 0.5%) in the hematology laboratory of Albany Medical Center Hospital. Results were communicated via hard copy to the study group.

Data Analysis: Data were first subjected to analysis for normalcy and found to be parametric. They were then analyzed by the paired t-test. All Hcts (%) are reported as mean ± SD.

The drop in Hct immediately after crystalloid infusion without phlebotomy (H1–H2, or Diff1) was compared with the drop in Hct immediately after crystalloid infusion following phlebotomy (H4–H6, or Diff3). Similarly, the drop in Hct 30 minutes after crystalloid infusion without phlebotomy (H1–H3, or Diff2) was compared with the drop in Hct 30 minutes after crystalloid infusion with phlebotomy (H4–H7, or Diff4).

A post-hoc receiver operating characteristic (ROC) curve was generated to determine the Hct drop thresholds that would maximize the sensitivity and specificity for the detection of the 500-mL phlebotomy immediately and 30 minutes after saline infusion.

RESULTS

Twenty-five subjects were enrolled. Five subjects were excluded. Two were subsequently determined to be ineligible to donate blood. One subject was not available to participate in the experimental portion of the study. For the other 2 patients, clotted specimens prevented complete data analysis. Therefore, data from 20 subjects were available for analysis. The subject age ranged from 19 to 41 years, with a mean of 26.4 years. Eight volunteers (40%) were female. Power analysis revealed an 89.9% chance of detecting a 1-SD difference between the 2 groups (β = 0.10).

As shown in Figure 2, the average initial baseline Hct (H1) was 41.6 ± 2.7, which is not different from the average baseline Hct on the second visit (H4) of 42.4 ± 3.1 (p = 0.36). Following phlebotomy, the mean Hct dropped from 42.4 ± 3.1 to 39.9 ± 2.9 (H5).

Immediately after the infusion of 15 mL/kg of saline, the Hct fell to an average of 37.1 ± 2.3 (H2), and then

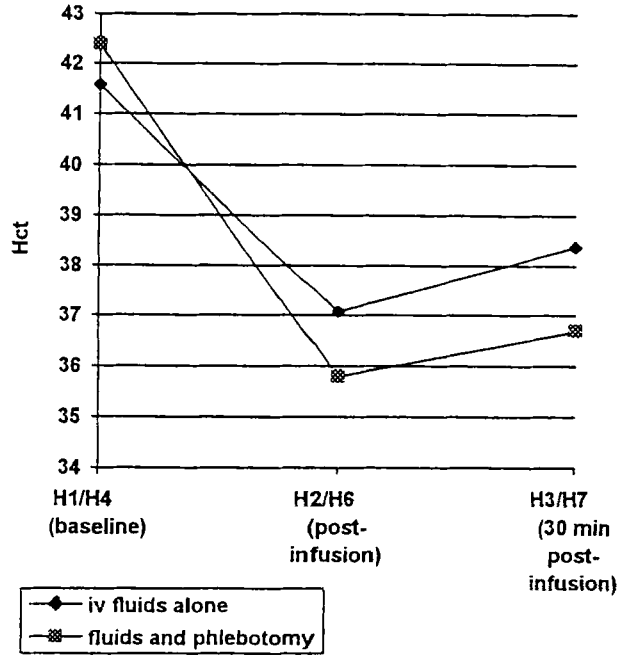

FIGURE 2. Hematocrit (Hct) values throughout the study.

rose to 38.4 ± 2.5 by 30 minutes later (H3). This indicates an average immediate drop (Diff1) of 4.5 ± 1.3 and an average delayed drop (Diff2) of 3.2 ± 1.3.

On the second visit, the average immediate Hct after phlebotomy followed by volume infusion (H6) was 35.8 ± 3.0, and the average delayed Hct (H7) was 36.7 ± 3.4. This indicates an average immediate drop (Diff3) of 6.6 ± 1.5, and an average delayed drop (Diff4) of 5.7 ± 1.1.

The immediate Hct drops with (Diff3) and without (Diff1) phlebotomy were significantly different ($p < 0.01$). Similarly, the delayed Hct drops with (Diff4) and without (Diff2) phlebotomy were also significantly different ($p < 0.01$).

A ROC curve was drawn to determine the threshold values that maximized the sensitivity and specificity of the Hct drop in detecting the 500-mL phlebotomy. This analysis yielded cutoff values for Hct drops of 5.4 when measured immediately after IV crystalloid and 4.3 when measured at 30 minutes. At these thresholds, the testing of serial Hcts had sensitivities/specificities of 95%/75%, and 90%/75%, respectively, for the detection of 500 mL of blood loss.

DISCUSSION

Traumatic deaths contribute a significant percentage of total deaths annually. These include "immediate" deaths, which are usually untreatable, "early" deaths, which occur within a few hours after injury, and "late" deaths, which occur days to weeks after injury. The emergency physician's goal is to prevent "early" deaths which often occur due to major internal hemorrhage.[6]

The ED evaluation of trauma patients with suspected acute blood loss includes, among other modalities, the use of serial Hcts.[1-3] While quantification of the change in Hct produced by IV crystalloid infusion has been studied,[4,5] the effect of blood loss in addition to IV crystalloid has received little attention.[7]

Greenfield et al.[4] reported a 4.5–6.3-point mean Hct drop in healthy subjects after 10-, 20-, and 30-mL/kg IV NS bolus. They further noted that despite continued infusion at 1–5 mL/kg/hr after the bolus, volunteers' Hcts subsequently rose an average of 1.5–2.3 points over 20 minutes, after which the Hcts remained stable. The authors concluded that it is possible that a failure of Hct to increase or the continued decline in Hct values after the termination of an infusion of crystalloid may signify acute blood loss.

Stamler[5] measured the Hct change after infusing 20 mL/kg of crystalloid over approximately 45 minutes. He noted a decrease in Hct of 1.6–6.4 (mean 4.1 and 4.8 in the 2 groups analyzed). He also noted that following the crystalloid bolus, a maintenance infusion of 15 mL/kg over 1 hour or 3 hours resulted in a mean increase of 0.6. He concluded that hemodilution following 20 mL/kg of IV crystalloid in nonbleeding volunteers should decrease Hct by 3–6 points.

Knottenbelt[8] evaluated the relation between initial hemoglobin level and hemorrhage in trauma patients (mostly penetrating). It has been well established that acute hemorrhage results in a decline in Hct due to movement of fluid from the extravascular to the intravascular space, otherwise known as "autoinfusion."[1,9,10] The concept of autoinfusion has important implications regarding the clinical assessment of blood loss. Blood volume is reflected clinically by hemodynamic parameters such as blood pressure and pulse, mental status, skin perfusion, and urine output. The degree of true blood loss can, however, be clinically underrepresented if significant autoinfusion has taken place, restoring intravascular volume. Knottenbelt concluded that a low hemoglobin level (especially ≤8 g/dL) identified a group of trauma patients with higher mortality and more severe blood loss.

Snyder[11] corroborated the utility of initial spun Hcts in detecting acute blood loss in trauma patients. His retrospective study noted that patients with initial spun Hcts ≤30 had an increased incidence of hypotension (40% vs 8%) and need for operative control of hemorrhage (55% vs 10%). He concluded that a low initial spun Hct (≤35 and especially ≤30) correlates with an increased incidence of hypotension, operative intervention for hemorrhage control, and mortality in trauma patients.

This study addresses the issue of whether there is a statistically significant difference in Hct drop between phlebotomized and nonphlebotomized individuals receiving IV crystalloid infusions. Though there is clearly overlap between the 2 populations, they are statistically different, and an Hct drop of ≥5.4 points immediately or ≥4.3 after 30 minutes is highly sensitive and specific for the detection of 500 mL of blood loss in this study.

LIMITATIONS AND FUTURE QUESTIONS

Future studies should address validation of our results in evaluation of trauma patients in the ED and the influence of this information on management. In reality, blood loss and crystalloid infusion occur simultaneously, not sequentially. As this was a pilot study, healthy volunteers without underlying comorbid conditions or ongoing blood loss were studied. The effects of these complicating variables need to be addressed in a larger validation study.

An argument could be made for using H5, the Hct after phlebotomy, as the baseline for the experimental phase. In fact, if the data are analyzed in this fashion, it becomes clear that phlebotomy accounts for much of the difference in the Hcts, and the 2 groups are no longer significantly different. However, using H5 as a baseline would be most analogous to a trauma patient with out-of-hospital blood loss that ceased prior to IV crystalloid and Hct measurement. In recognition of the fact that it is on-

going blood loss that is clinically most significant, we elected to use the Hct difference that includes both the blood loss and the volume infusion (i.e., H4–H6). In our clinical practice, we make an attempt to use the out-of-hospital blood drawn with the initial field IV placement to determine our baseline Hct because it is unaffected by hemodilution from crystalloid infusion and hopefully represents the earliest time when significant blood loss has not yet occurred.

While mild changes in Hct have been noted with regard to posture,[12-14] logistical requirements necessitated the ambulation of volunteers from phlebotomy at the Red Cross Center to the ED for the remainder of the study. The effect of this ambulation, while presumably minor, cannot be fully estimated.

It also has been reported in a study using rats that the degree of compensation by a subject in response to acute blood loss is proportional to the percentage of blood volume lost.[10] While our subjects were phlebotomized a nearly constant blood volume, we recognize that the percentage of blood volume lost was not the same for all volunteers. The volunteers ranged 49–111 kg (mean 76.4 kg) of body weight, with phlebotomy of 500 mL of blood accounting for 4.3–9.8 mL/kg. Evaluation of the effect of larger volumes of blood loss in the presence of IV crystalloid infusion on Hct should be pursued in volunteer studies in addition to using a consistent volume/kg of blood loss, e.g., 10 mL/kg.

CONCLUSIONS

The practice of measuring serial Hcts may be justifiable. In this pilot study, it was demonstrated that even in the presence of 15-mL/kg crystalloid infusion, an immediate Hct drop of ≥5.5, or a drop at 30 minutes of ≥4.3, suggested concomitant blood loss. Lesser degrees of Hct change may be solely due to hemodilution. Validation of these findings in a larger, unselected population (especially the trauma population) is needed.

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