HHS Public Access

Author manuscript

J Clin Apher. Author manuscript; available in PMC 2017 October 01.

Published in final edited form as:

J Clin Apher. 2016 October ; 31(5): 459–463. doi:10.1002/jca.21438.

Citrate Anticoagulation: Are Blood Donors Donating Bone?

Walter Bialkowski[a], **Roberta Bruhn**[b], **Gustaf Edgren**[c], and **Paula Papanek**[a]

[a]Department of Physical Therapy, Marquette University, Milwaukee, WI

[b]Epidemiology Core, Blood Systems Research Institute, San Francisco, CA

[c]Department of Medical Epidemiology and Biostatistics, Karolinska Institutet, Stockholm, Sweden; and Department of Medicine, Division of Hematology, Karolinska University Hospital, Stockholm, Sweden

Abstract

An estimated 2.4 million volunteer apheresis blood donation procedures were performed in the United States in 2010 and increases in the proportion of transfused blood products derived from apheresis blood collections have been consistently reported. Anticoagulation is required during apheresis and is achieved with citrate. Donor exposure to citrate causes an acute physiological response in the donor maintaining serum mineral homeostasis. Some data are available on the sequelae of this acute response in the days and weeks following exposure, raising questions about bone mineral density in regular apheresis donors. New research is emerging that addresses the potential long term health outcomes of repeated citrate exposure. This article reviews the acute physiological response to citrate anticoagulation in volunteer blood donors, presents contrasting perspectives on the potential effects of citrate exposure on bone density, and identifies key knowledge gaps in our understanding of long term health outcomes in apheresis donors.

Keywords

citrate; donor; bone; anticoagulation

(1) Introduction

Apheresis blood collections produce blood components that save lives and represent an increasing fraction of all blood derived components in clinical use today. In the United States, approximately four apheresis derived platelet products were transfused in 2004 for every whole blood (WB) derived product [1]. This proportion increased to 5:1 in 2006 [2], 6:1 in 2008 [3], and 10:1 in 2010 [4] despite the widespread implementation of blood management programs. Similar trends of increased apheresis collections and transfusions are apparent in Australia [5, 6], European nations including Belgium, Ireland, Norway, Sweden, and the United Kingdom [7, 8], as well as China [9]. In some countries, such as Canada, the number of annual apheresis collections has marginally declined as a result of increases in

Correspondence to: Walter Bialkowski, Clinical and Translational Rehabilitation Health, Marquette University, P.O. Box 1881, Milwaukee, WI 53201-1881, walter.bialkowski@marquette.edu, Ph: (414) 937-3851, Fax: (414) 937-6284.

Conflicts of interest: none.

large volume apheresis platelet collections [10]. These observations indicate that there are global increases in the number of donors undergoing apheresis, increases in the number of procedures an individual donor experiences, and/or increases in the duration of individual apheresis procedures.

Apheresis donation frequencies are determined by national governments [11]. Platelet apheresis donation guidelines in the United States are founded on studies showing that frequent apheresis platelet donors are able to maintain platelet counts within the normal reference range [12–14]. These platelet apheresis studies supported an FDA policy increase in the number of platelet apheresis donations an individual volunteer donor can make from 12 [15] to 24 [16] per rolling 12 month period with no lifetime maximum. Many countries allow for 24 voluntary plasma or platelet apheresis procedures, with a minimum of 48 hours between collections, and a maximum of 15 liters of plasma removed per year [17]. U.S. Federal regulations on paid source plasma donors allow 110 apheresis donations in a rolling 12 month period [21CFR640.65(8)]. Operationally, apheresis donor recruitment strategies focus efforts on retaining donors willing to donate often and who are capable of giving multi-product donations in part because the number of donations in the previous year has a positive association with donor return [18]. These patterns of apheresis blood collection and transfusion emphasize the importance of understanding any long-term health impacts of apheresis on blood donors.

(2) Citrate Anticoagulation

Citrate is the standard anticoagulant (AC) used during apheresis donation procedures [19, 20]. The two most common citrate anticoagulant solutions used in platelet apheresis are acid citrate dextrose (ACD)-A which is 3% citrate and ACD-B which is 2% citrate. Trisodium citrate (4%) is predominantly used in plasmapheresis and citrate phosphate dextrose (CPD) is commonly used in red-cell only apheresis [11, 21]. Citrate in these solutions is comprised of different quantities of the active ingredients sodium citrate (dihydrate) and citric acid [22]. Three procedural settings determine the amount of citrate introduced to WB in the extracorporeal circuit: the inlet pump rate, the AC flow rate, and the WB to AC ratio [20]. Some apheresis systems derive these values using donor blood volume estimates [23], whereas others are controlled by the operator. Plasma concentration of citrate in the extracorporeal circuit is maintained at 15 – 24 mmol/L to facilitate anticoagulation [24].

The amount of citrate returned to an apheresis blood donor depends on a number of factors. Because citrate anticoagulation in the extracorporeal circuit is based on concentration, the volume of the blood component(s) returned, the concentration of citrate within the blood component(s), and the return speed determine citrate exposure to the donor. Citrate concentration among blood components has been quantified [25, 26] and the efficiency by which blood components are separated (primarily duration of procedure) also determines donor exposure to citrate [27]. Additionally, the intermittent nature of discontinuous apheresis systems creates variability with respect to the frequency and volume of citrate returned. Therefore, total citrate exposure to a donor is highly varied but generally correlated with the volume of plasma returned and the concentration of citrate in the component(s) that is returned. The amount of citrate used in apheresis platelet collections positively correlates

with serum citrate concentration in the donor during apheresis [28–30] illustrating the impact of longer procedure times. The half-life of citrate in the circulation is approximately 36 minutes [31] and donors are able to fully metabolize exogenous citrate from apheresis collections within 24 hours of exposure [28, 29]. Nevertheless, there is increasing evidence that even modest citrate exposure in apheresis blood donors serves as a biological stimulus with cascading implications on bone remodeling.

(3) Citrate Physiology

Citrate's role as a metal chelating agent that binds divalent cations, such as calcium, has been thoroughly characterized [32]. Like endogenous citrate, citrate AC solutions chelate calcium ions in the blood by forming calcium-citrate complexes that disrupt coagulation [33, 34]. Studies show reductions in circulating calcium concentrations in apheresis blood donors of 22–35% when comparing pre- and post-apheresis concentrations [28, 30, 35]. Decreases in iCa of 15% have been seen 10 minutes after initiation of apheresis and 31% after 90 minutes [19]. Donors generally tolerate decreases in concentrations of iCa up to 20% before experiencing side effects [36] with women having a greater sensitivity to declining concentrations than men [37]. Total calcium declines during apheresis to a lesser extent [28, 29, 38] or not at all [35].

G-protein coupled receptors on the surface of the parathyroid glands and kidneys directly sense declines in iCa concentration in the blood and stimulate secretory cells to release parathyroid hormone (PTH) [39]. Serum concentrations of PTH reach maximum levels within 5–15 minutes of the start of apheresis [40]. This has been corroborated in a number of platelet apheresis donor studies [28, 29, 38, 41]. Initial PTH surges in apheresis donors are short-lived and PTH concentration returns to near-baseline as early as 30 minutes after the infusion of citrate is terminated [28, 41]. One study has shown that PTH may remain elevated up to one day after the procedure despite a termination in exposure [38].

The release of PTH into circulation simultaneously triggers all three of the body's main mechanisms to restore normal iCa: increased calcium reabsorption in the distal tubules of kidney nephrons, increased intestinal calcium absorption, and increased bone resorption. Calcium reabsorption in the kidney is 98% efficient under normal conditions and thus, a mechanism to replenish iCa losses during apheresis that involves reabsorption will likely have minimal impact on iCa concentrations. Although increases in serum PTH increase calcium reabsorption in the distal convoluted tubule, citrate exposure through apheresis increases urinary loss of calcium [42] in a dose-dependent manner [28] and occurs during the 24 hour period after exposure [29, 35].

PTH also stimulates the activation of Vitamin D, which in turn increases intestinal absorption of calcium. Activated Vitamin D exceeds baseline concentrations by an average of 26% [29] one day after apheresis. Through a Vitamin D mediated pathway, some calcium can be replenished in apheresis donors through small intestine absorption [25]. But despite being provided with large amounts of calcium as supplements, donors are not able to recover 100% of baseline iCa concentrations through this mechanism. Furthermore, calcium supplementation is, in practice, symptom dependent and not routinely employed during all

procedures. Thus, the apheresis donor's body may rely on resorption of metabolically active trabecular bone that comprises approximately 15 – 20% of the human skeleton to recoup lost calcium.

(4) Evidence of Bone Resorption in Donors

There are several markers of bone resorption; however only a few have been measured in apheresis donors. C-terminal telopeptides, such as β-CTX, are both sensitive and specific measures that quantify the breakdown of type 1 collagen [43]. In a randomized, placebo-controlled study of blood donors, citrate infusion increased serum concentrations of β-CTX in apheresis donors whereas controls not receiving citrate had no change in their serum β-CTX ($p < 0.0001$) [44]. This finding held true for donors in another study where both serum and urine concentrations of β-CTX were elevated by as much as 26% and 17%, respectively, and remained elevated for up to 24 hours post-donation [38]. The largest measured increases in β-CTX have been observed at the completion of citrate infusion [44] suggesting that bone resorption begins during exposure to citrate. When concentrations of β-CTX are compared to concentrations of osteocalcin (OC), a protein secreted by bone-forming osteoblasts, the proportion of these two markers throughout the procedure increases suggesting that bone metabolism may shift toward resorption during apheresis.

Results from a bone density study of 102 apheresis platelet donors with a lifetime average of 85 apheresis procedures (range 16 – 633) as compared to non-blood donor controls demonstrated significantly lower bone density at the lumbar spine (Z-score P=0.038) for apheresis donors as compared to controls [38]. The lumbar spine is rich in metabolically active trabecular bone that requires 14 days or longer to replenish serum calcium, a period over which some have shown evidence of bone remodeling [45]. The opportunity exists to fully catalog apheresis blood donor physiology in the weeks following IV citrate exposure. Making use of the available data in predicting long term effects on bone health in this donor population is challenging, though a prospective study at the National Institutes of Health (NCT00073060) is incorporating a longitudinal assessment of bone density to address this.

(5) Evidence of Bone Deposition in Donors

In contrast to the lumbar spine data, no significant differences in bone density were seen at the hip or femoral neck for apheresis platelet donors compared to controls (Z-score P=0.36 and P=0.72, respectively) [38]. There is some evidence that exposure to citrate from apheresis actually favors bone deposition, not resorption. OC has been shown to remain slightly elevated at 24 hours post-apheresis donation [29]. Furthermore, concentrations of osteoprotegerin (OPG), a receptor that regulates the maturation of bone degrading osteoclasts, decreased following 120 minutes of citrate exposure and recovered to baseline 24 hours post-exposure [38]. Tartrate-resistant acid phosphatase (TRAP), an enzyme expressed by osteoclasts, has been shown to be a useful marker of bone resorption because of its limited variability *in vivo* [46, 47]. In apheresis platelet donors TRAP was observed to be lower than baseline at both 120 minutes and 24 hours post-exposure suggesting that apheresis acutely suppresses bone resorption. The authors of this study do not address the paradoxical nature of this finding, especially considering their claim that a finding of lower

bone density in apheresis donors relative to controls is a “true finding”. It should be mentioned that a limitation of using TRAP to assess bone resorption in healthy people may be the inability to make meaningful interpretations when threshold concentrations below that of pathological conditions are not met [48].

Steddon and Cunningham [49] noted in their review of calcium receptor manipulation therapies that short periods of elevated PTH favor bone formation by means of expediting the maturation of osteoblasts. Further, it has been conceptualized that large and rapid increases in PTH followed by normalization, such as that stimulated by calcilytic drug therapies, may translate into anabolic effects on bone [50]. Finally, we should not ignore that oral potassium citrate is a common treatment for low bone density with documented efficacy [51]. Thus, intermittent exposure to citrate through apheresis blood donation could theoretically have beneficial effects on bone. The conclusions of many of these studies have been derived from clinical trials of postmenopausal women only, all of whom have declining estrogen. Because of estrogen’s central role in bone metabolism, the generalization of these findings to apheresis blood donors warrants very careful attention and additional research. Should there be a positive association between apheresis blood donation and bone density, then a large-scale, retrospective cohort study currently being conducted on the Scandinavian Donations and Transfusions (SCANDAT) database [52, 53] may lend insight into this association.

(6) Conclusion

Apheresis blood products are increasingly represented in the national blood supply. These products are derived from volunteer donors who undergo anticoagulation via citrate. Intravenous citrate exposure causes fluctuations in blood donor mineral homeostasis, especially ionized calcium. Acute physiological data on fluctuations in ionized calcium are robust; however, data on complete recovery following intravenous citrate exposure are limited. Subsequent and prolonged effects on important markers of bone health are inconclusive. The blood collection community should be highly interested in reported declines in bone density among high frequency apheresis blood donors as a measure of donor safety and studies are underway to help improve our understanding of long term health outcomes in this donor population.

Acknowledgments

The authors would like to thank members of the NHLBI Recipient Epidemiology and Donor Evaluation Study-III (REDS-III) Domestic Steering Committee (Dr. D.J. Brambilla, Dr. M.P. Busch, Dr. R.G. Cable, A.M. Cristman, Dr. R.Y. Dodd, Dr. S.A. Glynn, Dr. J.L. Gottschall, Dr. J.E. Kiss, Dr. S.H. Kleinman, Dr. A.E. Mast, Dr. E.L. Murphy, Dr. P.J. Norris, Dr. E.L. Snyder, Dr. B. St. Lezin, M.T. Sullivan, and Dr. D. J. Triulzi), Dr. Robert D. Blank, Dr. Hershel Raff, and Linda Gruber for thoughtful consideration of the ideas presented. This review was partially funded by the NHLBI Recipient Epidemiology and Donor Evaluation Study-III (REDS-III).

References

1. Whitaker, BISM. Services UDoHaH. The 2005 Nationwide Blood Collection and Utilization Survey Report. Washington D.C: 2006.
2. Whitaker, BIGJ., et al. Services UDoHaH. The 2007 Nationwide Blood Collection and Utilization Survey Report. Washington D.C: 2008.

3. Services RotUDoHaH. US Department of Health and Human Services OotASoH. The 2009 National Blood Collection and Utilization Survey Report. Washington D.C: 2011.
4. Services DoHaH. DHHS U. The 2011 National Blood Collection and Utilization Survey Report. Washington D.C: 2013.
5. Committee NBAHA. Australian Haemovigilance Report. Canberra, ACT; Australia: 2015.
6. Committee NBAHA. Australian Haemovigilance Report. Canberra, ACT; Australia: 2013. (Last accessed
7. Janssen MP, B-GM-E. The collection, testing and use of blood and blood components in Europe. 2010
8. van Hoeven LRJMP, Rautmann G. The collection, testing and use of blood and blood components in Europe. 2012
9. Shi L, Wang J, Liu Z, et al. Blood donor management in china. Transfus Med Hemother. 2014; 41:273–82. [PubMed: 25254023]
10. Services CB. Canadian Blood Services Annual Report. Ottawa, Ontario K1G 4J5: 2014.
11. Organization WH. WHO Recommendations for the Production, Control and Regulation of Human Plasma for Fractionation. 2005.
12. Glowitz RJ, Slichter SJ. Frequent multiunit plateletpheresis from single donors: effects on donors' blood and the platelet yield. Transfusion. 1980; 20:199–205. [PubMed: 7368268]
13. Slichter SJ. Relationship between platelet count and bleeding risk in thrombocytopenic patients. Transfus Med Rev. 2004; 18:153–67. [PubMed: 15248165]
14. Lazarus EF, Browning J, Norman J, et al. Sustained decreases in platelet count associated with multiple, regular plateletpheresis donations. Transfusion. 2001; 41:756–61. [PubMed: 11399815]
15. Administration FaD. Division of Blood and Blood Products H-. Guideline for the Collection of Platelets, Pheresis. Bethesda, MD: Food and Drug Administration; 1981.
16. Administration USFaD. Research DoHaHSCfBEa. Guidance for Industry and FDA Review Staff - Collection of Platelets by Automated Methods. 2007.
17. Service NB. Guidelines for the Blood Transfusion Services in the United Kingdom. London, UK: The Stationery Office; 2013.
18. Custer B, Rios JA, Schlumpf K, et al. Adverse reactions and other factors that impact subsequent blood donation visits. Transfusion. 2012; 52:118–26. [PubMed: 21682732]
19. Hester JP, McCullough J, Mishler JM, et al. Dosage regimens for citrate anticoagulants. J Clin Apher. 1983; 1:149–57. [PubMed: 6546053]
20. Lee G, Arepally GM. Anticoagulation techniques in apheresis: from heparin to citrate and beyond. J Clin Apher. 2012; 27:117–25. [PubMed: 22532037]
21. Burnouf T. Modern plasma fractionation. Transfus Med Rev. 2007; 21:101–17. [PubMed: 17397761]
22. Kishimoto M, Ohto H, Shikama Y, et al. Treatment for the decline of ionized calcium levels during peripheral blood progenitor cell harvesting. Transfusion. 2002; 42:1340–7. [PubMed: 12423519]
23. Despotis GJ, Goodnough LT, Dynis M, et al. Adverse events in platelet apheresis donors: A multivariate analysis in a hospital-based program. Vox Sang. 1999; 77:24–32. [PubMed: 10474087]
24. Strauss RG. Mechanisms of adverse effects during hemapheresis. J Clin Apher. 1996; 11:160–4. [PubMed: 8915821]
25. Hester JP, Ayyar R. Anticoagulation and electrolytes. J Clin Apher. 1984; 2:41–51. [PubMed: 6536658]
26. Penny A. Plasmapheresis procedure, design, and operation: a consideration of citrate anticoagulant usage. Transfus Sci. 1989; 10:51–6.
27. Burgstaler EA. Blood component collection by apheresis. J Clin Apher. 2006; 21:142–51. [PubMed: 15880369]
28. Bolan CD, Greer SE, Cecco SA, et al. Comprehensive analysis of citrate effects during plateletpheresis in normal donors. Transfusion. 2001; 41:1165–71. [PubMed: 11552076]

29. Bolan CD, Cecco SA, Yau YY, et al. Randomized placebo-controlled study of oral calcium carbonate supplementation in plateletpheresis: II. Metabolic effects. Transfusion. 2003; 43:1414–22. [PubMed: 14507273]
30. Szymanski IO. Ionized calcium during plateletpheresis. Transfusion. 1978; 18:701–8. [PubMed: 726016]
31. Kramer L, Bauer E, Joukhadar C, et al. Citrate pharmacokinetics and metabolism in cirrhotic and noncirrhotic critically ill patients. Crit Care Med. 2003; 31:2450–5. [PubMed: 14530750]
32. Dzik WH, Kirkley SA. Citrate toxicity during massive blood transfusion. Transfus Med Rev. 1988; 2:76–94. [PubMed: 2980082]
33. Mann KG, Krishnaswamy S, Lawson JH. Surface-dependent hemostasis. Semin Hematol. 1992; 29:213–26. [PubMed: 1641667]
34. Mann KG, Lawson JH. The role of the membrane in the expression of the vitamin K-dependent enzymes. Arch Pathol Lab Med. 1992; 116:1330–6. [PubMed: 1456880]
35. Bolan CD, Cecco SA, Wesley RA, et al. Controlled study of citrate effects and response to i.v. calcium administration during allogeneic peripheral blood progenitor cell donation. Transfusion. 2002; 42:935–46. [PubMed: 12375668]
36. Ladenson JH, Miller WV, Sherman LA. Relationship of physical symptoms, ECG, free calcium, and other blood chemistries in reinfusion with citrated blood. Transfusion. 1978; 18:670–9. [PubMed: 31718]
37. Bolan CD, Wesley RA, Yau YY, et al. Randomized placebo-controlled study of oral calcium carbonate administration in plateletpheresis: I. Associations with donor symptoms. Transfusion. 2003; 43:1403–13. [PubMed: 14507272]
38. Amrein K, Katschnig C, Sipurzynski S, et al. Apheresis affects bone and mineral metabolism. Bone. 2010; 46:789–95. [PubMed: 19922822]
39. Hebert SC, Brown EM, Harris HW. Role of the Ca(2+)-sensing receptor in divalent mineral ion homeostasis. J Exp Biol. 1997; 200:295–302. [PubMed: 9050237]
40. Toffaletti J, Nissenson R, Endres D, et al. Influence of continuous infusion of citrate on responses of immunoreactive parathyroid hormone, calcium and magnesium components, and other electrolytes in normal adults during plateletapheresis. J Clin Endocrinol Metab. 1985; 60:874–9. [PubMed: 3980672]
41. Mercan D, Bastin G, Lambermont M, et al. Importance of ionized magnesium measurement for monitoring of citrate-anticoagulated plateletpheresis. Transfusion. 1997; 37:418–22. [PubMed: 9111280]
42. Hamm LL. Renal handling of citrate. Kidney Int. 1990; 38:728–35. [PubMed: 2232510]
43. Rosen HN, Moses AC, Garber J, et al. Serum CTX: a new marker of bone resorption that shows treatment effect more often than other markers because of low coefficient of variability and large changes with bisphosphonate therapy. Calcif Tissue Int. 2000; 66:100–3. [PubMed: 10652955]
44. Chu XL, Hou JM, Lin H, et al. Short-term effects of citrate on markers of bone metabolism in Chinese blood donor volunteers. Zhongguo Shi Yan Xue Ye Xue Za Zhi. 2010; 18:785–9. [PubMed: 20561451]
45. Ronquillo JYY, Stevens W, Cecco S, Matthews C, Byrne P, Alvandi F, Collins M, Wesley R, Rehak N, Leitman SF, Bolan CD. Acute and Sub-acute Citrate Mediated Effects and Responses to IV Calcium in Healthy Apheresis Donors; AABB. Transfusion. 2007:15. [PubMed: 17207225]
46. Hannon RA, Clowes JA, Eagleton AC, et al. Clinical performance of immunoreactive tartrate-resistant acid phosphatase isoform 5b as a marker of bone resorption. Bone. 2004; 34:187–94. [PubMed: 14751577]
47. Capeller B, Caffier H, Sutterlin MW, et al. Evaluation of tartrate-resistant acid phosphatase (TRAP) 5b as serum marker of bone metastases in human breast cancer. Anticancer Res. 2003; 23:1011–5. [PubMed: 12820340]
48. Chao TY, Yu JC, Ku CH, et al. Tartrate-resistant acid phosphatase 5b is a useful serum marker for extensive bone metastasis in breast cancer patients. Clin Cancer Res. 2005; 11:544–50. [PubMed: 15701839]
49. Steddon SJ, Cunningham J. Calcimimetics and calcilytics--fooling the calcium receptor. Lancet. 2005; 365:2237–9. [PubMed: 15978932]

50. Rachner TD, Khosla S, Hofbauer LC. Osteoporosis: now and the future. Lancet. 2011; 377:1276–87. [PubMed: 21450337]

51. Jehle S, Hulter HN, Krapf R. Effect of potassium citrate on bone density, microarchitecture, and fracture risk in healthy older adults without osteoporosis: a randomized controlled trial. J Clin Endocrinol Metab. 2013; 98:207–17. [PubMed: 23162100]

52. Edgren G, Rostgaard K, Vasan SK, et al. The new Scandinavian Donations and Transfusions database (SCANDAT2): a blood safety resource with added versatility. Transfusion. 2015

53. Kleinman S, Busch MP, Murphy EL, et al. The National Heart, Lung, and Blood Institute Recipient Epidemiology and Donor Evaluation Study (REDS-III): a research program striving to improve blood donor and transfusion recipient outcomes. Transfusion. 2014; 54:942–55. [PubMed: 24188564]