

| pISSN 2586-6052 | eISSN 2586-6060

A comparative study of stored arterial versus venous blood collected using the acute normovolemic hemodilution method in coronary artery bypass grafting patients in Iran

Mojtaba Mansouri¹, Alireza Yazdani¹, Gholamreza Masoumi¹, Mohsen Mirmohammadsadeghi², Amir Mirmohammadsadeghi²

¹Department of Anesthesiology, Anesthesiology Research Center, School of Medicine, Isfahan University of Medical Sciences, Isfahan, Iran ²Department of Surgery, School of Medicine, Isfahan University of Medical Sciences, Isfahan, Iran

Background: In the present study, arterial and venous blood was collected from patients who were candidates for elective coronary artery bypass grafting (CABG); the blood was stored for 28 days and cellular, biomechanical, and hematological changes in blood were compared to determine whether stored arterial blood is superior to stored venous blood.

Methods: The present follow-up comparative study included 60 patients >18 years of age, with hemoglobin >14 mg/dl and ejection fraction >40% who were candidates for CABG. After induction of anesthesia, 250 ml of arterial or venous blood was drawn from patients (arterial blood group and venous blood group). Laboratory blood samples were taken at specified times from the collected blood and re-injected into the patients after CABG.

Results: Significant differences were observed in pH, partial pressure of carbon dioxide (PCO_2), partial pressure of oxygen (PO_2), bicarbonate (HCO_3), and glucose values at several time points between the groups. Other parameters such as urea and creatinine did not show any significant differences between the groups.

Conclusions: Twenty-eight days of storage can have a negative effect on some of the cellular, biochemical, and hematological components of arterial and venous blood; however, the quality of stored arterial blood and venous blood does not differ significantly.

Key Words: coronary artery bypass graft; hemodilution; stored blood

Original Article

Received: November 1, 2022 Revised: March 16, 2023 Accepted: March 19, 2023

Corresponding author

Alireza Yazdani Department of Anesthesiology, School of Medicine, Isfahan University of Medical Sciences, Hezarjerib Ave, Isfahan 8174673461, Iran Tel: +98-31-3792-3243 Fax: +98-31-3668-8628 Email: dr_alirezayazdani@yahoo.com

INTRODUCTION

Transfusion of stored blood is a common medical intervention intended to improve oxygen delivery to body tissues and reduce the complications of anemia or related medical procedures [1]. Blood is routinely stored at 2 °C-6 °C for up to 35 days in preservatives such as citrate phosphate dextrose adenine (CPDA). During storage, several physical, biochemical, and biomechanical changes occur in blood products (storage lesion) that reduce their survival and function [2]. The changes include prominent alterations in red blood cell (RBC) morphology known as red cell storage lesion and blood stored for a long period of time is

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This is an Open Access article distributed under the terms of Creative Attributions Non-Commercial License (https://creativecommons.org/ li-censes/by-nc/4.0/) which permits unrestricted noncommercial use, distribution, and reproduction in any medium, provided the original work is properly cited. more affected than when stored for a short period [3]. Storage lesion leads to reduced adenosine triphosphate (ATP) and hemoglobin (Hb) level, increased lactate level with resulting lower pH, increased lysis, minimized blood movement and oxygen transport, decreased intracellular potassium (K), and increased sodium (Na) in the cytoplasm [3-5]. Consequently, red cell storage lesion reduces efficacy of the transfused blood products which increases the chances of transfusion-related morbidities [5].

In a previous study, the extent of correlation between arterial and peripheral venous samples for blood gases and acid base status in critically ill and emergency department patients were investigated. The results showed that venous blood may be a useful alternative to arterial blood during blood gas analysis, obviating the need for arterial puncture in difficult clinical situations, especially in trauma patients, for initial emergency department assessment and early stages of resuscitation [6].

In another investigation, significant changes were observed in some biochemical parameters such as serum aspartate aminotransferase, total protein, albumin, urea, chloride, and K levels assessed at periodic intervals. These changes might be caused by spontaneous hemolysis or non-viable RBCs due to storage, thus, blood should be transfused as early as possible to prevent a negative effect on the biochemical composition of RBCs [7]. However, less evidence exists regarding changes in donated arterial and venous blood after storage. Therefore, assessing the difference in donated arterial and venous blood after storage is beneficial for managing patients in emergency conditions.

In the present study, the cellular, biochemical, and hematological changes in donated arterial and venous blood after 28 days of storage were compared in patients who were candidates for elective coronary artery bypass grafting (CABG).

MATERIALS AND METHODS

The present follow-up comparative study was conducted in Chamran Heart Hospital in 2021 in Iran. Sixty patients >18 years of age with Hb >14 mg/dl and ejection fraction (EF) >40% scheduled for elective CABG were enrolled in the study. Subjects were selected using convenience sampling method. Sampling was continued until the required number of samples was obtained. Patients with emergency surgery, severe renal and hepatic dysfunction, hematological disorders, EF <40%, or undergoing a reoperation were excluded.

The Ethics Committee of Isfahan University of Medical

KEY MESSAGES

- Significant changes were not found in urea and creatinine at 28 days of whole blood storage.
- Blood should be transfused as early as possible to prevent a negative effect on the cellular, biochemical, and hematological components of blood.
- After candidates for elective coronary artery bypass grafting, significant differences in pH and bicarbonate (HCO₃) were observed between the arterial and venous blood groups.

Sciences approved this study (ethics code: IR.MUI.MED. REC.1401.114). In addition, written informed consent was obtained from all participants.

First, patients were monitored using a five-lead electrocardiogram, pulse oximetry, noninvasive blood pressure monitoring, and capnography, and their radial artery was cannulated for blood sampling. All patients were anesthetized in the same manner; general anesthesia was administered with fentanyl (5 μ g/kg) as a narcotic drug, cisatracurium (0.2 mg/kg) as a muscle relaxant, and Na thiopental (5 mg/kg) as a hypnotic drug. Then, tracheal intubation was performed and the lungs were ventilated using a ventilator adjusted to produce an arterial partial pressure of carbon dioxide (PCO₂) of 35-45 mm Hg. Anesthesia was maintained with propofol (100 µg/kg/min) and morphine (0.2 mg/kg). A central venous catheter was inserted into the right internal jugular vein in all patients after anesthesia induction. Patients were randomly assigned to two groups based on arterial and venous blood donation. Donated arterial blood was collected from the radial artery in patients in the arterial group and donated venous blood was obtained from the peripheral vein in patients in the venous group. Blood was slowly drawn into bags containing 50 ml CPDA-1 (Figure 1).

Acute normovolemic hemodilution, which is essential to minimize surgical blood loss, was performed immediately after induction of anesthesia and before heparin injection and starting the cardiopulmonary pump. The patient had 250 ml of arterial or venous blood drawn and simultaneously replaced with a triple volume of crystalloid (750 ml intravenous saline or Ringer's solution) through the central or peripheral venous cannula to maintain normovolemia.

The first blood sample was then sent to the laboratory for venous blood gas and arterial blood gas analyses, complete blood count, and biochemistry (creatinine [Cr], lactate de-



Figure 1. Consolidated Standards of Reporting Trials (CONSORT) flow diagram.

hydrogenase [LDH], albumin, Na, K, platelet, glucose, mean corpuscular hemoglobin [MCH], mean corpuscular volume [MCV], and MCH concentration [MCHC]) analysis.

The collected blood was re-injected into the patients through peripheral venous cannula after CABG and hemostasis established and protamine injected. During the blood transfusion, the second blood sample was collected from the patients and immediately sent to the hospital laboratory for analysis.

After the blood infusion, 10 ml of the blood remained in the bag, which was immediately transferred to the hospital laboratory. This sample was stored in the blood bank refrigerator at a temperature between 1 °C–6 °C for 28 days and then analyzed.

Statistical Analysis

The results are expressed as the mean± standard deviation. The Student t-test was used to compare parametric variables and the chi-square test to compare categorical variables between groups. Time course experiments of blood were analyzed using repeated measures analysis of variance with posthoc Bonferroni for multiple comparisons at each time point. A P-value <0.05 was considered to indicate statistical significance. All the statistical analyses were performed using IBM SPSS ver. 21 (IBM Corp.).

Table 1. Comparison of demographic variables between groups

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Variable	Arterial blood	Venous blood	P-value
Age (yr)	64±9	55±13	0.003
Sex			0.125
Male	24 (80.0)	28 (93.3)	
Female	6 (20.0)	2 (6.7)	
Diabetes			0.781
Yes	10 (33.3)	9 (30.0)	
No	20 (66.7)	21 (70.0)	
Smoking			0.513
Yes	5 (16.7)	7 (23.3)	
No	25 (83.3)	23 (76.7)	

Values are presented as mean±standard deviation or number (%).

RESULTS

In the present study, 60 patients were divided into two groups using convenience sampling: 30 in the arterial blood group and 30 in the venous blood group (Figure 1). Sampling was continued until the required number of samples was obtained. Table 1 shows that all demographic data were similar between the groups with the exception of age (P=0.003).

The results showed the changes in donated blood at three time points were significantly different in almost all parameters in both groups except hematocrit (P=0.445 in arterial group and P=0.122 in venous group) and MCH (P=0.476 in



Variable	Time	Arterial blood	Venous blood	P-value
рН	Immediately after donation	7.15±0.11	6.09±0.10	0.05
	4 Hours after donation	7.07±0.06	7.04±0.06	0.02
	28 Days after donation	6.59±0.34	6.45±0.24	0.06
	P-value	<0.001	<0.001	
	P-value for interaction	0.804		
	P-value for intervention	0.288		
PCO ₂	Immediately after donation	49.60±8.33	57.25±10.63	0.003
	4 Hours after donation	54.81±7.79	62.23±7.61	0.001
	28 Days after donation	27.31±11.19	27.72±9.8	0.883
	P-value	<0.001	<0.001	
	P-value for interaction	0.594		
	P-value for intervention	0.239		
pO ₂	Immediately after donation	190.82±124.57	68.94±18.62	<0.001
	4 Hours after donation	131.60±61.84	71.38±20.31	<0.001
	28 Days after donation	237.82±36.90	243.09±31.10	0.568
	P-value	<0.001	<0.001	
	P-value for interaction	0.033		
	P-value for intervention	0.393		
HCO ³	Immediately after donation	16.83±2.84	17.01±2.21	0.785
	4 Hours after donation	15.40±1.66	16.34±1.38	0.021
	28 Days after donation	2.09±1.74	1.98±1.66	0.800
	P-value	<0.001	<0.001	
	P-value for interaction	0.500		
	P-value for intervention	0.878		
Na	Immediately after donation	146.75±5.16	148.51±3.66	0.135
	4 Hours after donation	149.52±4.03	150.84±4.00	0.210
	28 Days after donation	140.14±6.75	142.77±7.75	0.168
	P-value	<0.001	<0.001	
	P-value for interaction	0.153		
	P-value for intervention	0.102		
К	Immediately after donation	3.22±1.76	2.85±0.50	0.269
	4 Hours after donation	2.63±0.46	2.73±0.59	0.494
	28 Days after donation	17.23±2.39	18.06±2.31	0.178
	P-value	<0.001	<0.001	
	P-value for interaction	0.352		
	P-value for intervention	0.281		
Glucose	Immediately after donation	431.53±132.08	513.67±90.18	0.007
	4 Hours after donation	416.63±123.37	446.17±123.33	0.602
	28 Days after donation	223.63±68.40	235.43±60.97	0.538
	P-value	<0.001	<0.001	
	P-value for interaction	0.503		
	P-value for intervention	0.509		
Lactate	Immediately after donation	11.39±3.66	12.46±3.05	0.224
	4 Hours after donation	21.44±4.29	19.34±4.14	0.059
	28 Days after donation	175.03±15.26	173.40±13.73	0.665
	P-value	<0.001	<0.001	
	P-value for interaction	0.364		
	P-value for intervention	0.543		

Table 2. Comparison between and within blood groups

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Table 2. Continued



Variable	Time	Arterial blood	Venous blood	P-value
RBC	Immediately after donation	3.92±0.53	4.11±0.45	0.143
-	4 Hours after donation	3.95±0.75	3.95±0.95	0.986
	28 Days after donation	3.24±0.77	3.47±0.69	0.233
	P-value	< 0.001	<0.001	
	P-value for interaction	0.210		
	P-value for intervention	0.448		
WBC	Immediately after donation	5 45+1 39	531+106	0.643
nbc	4 Hours after donation	5 24+1 40	5 74+1 53	0.189
	28 Days after donation	4 69+1 48	4 84+1 24	0.665
	P_volue	0.001	0.014	0.005
	P-value for interaction	0.001	0.014	
		0.000		
Hemoglobin	Immediately after denation	12 12+1 66	12 E2+1 E0	0.227
Hemoglooin		12.12±1.00	12.52±1.50	0.327
	4 Hours after donation	11.99±2.13	11.83±2.11	0.762
	28 Days after donation	9.96±2.39	10.30±2.20	0.569
	P-value	<0.001	<0.001	
	P-value for interaction	0.703		
	P-value for intervention	0.867		
Hematocrit	Immediately after donation	35.82±4.50	37.84±3.31	0.055
	4 Hours after donation	35.89±6.17	35.67±5.47	0.886
	28 Days after donation	34.61±7.82	35.95±7.30	0.497
	P-value	0.445	0.122	
	P-value for interaction	0.413		
	P-value for intervention	0.394		
MCV	Before CABG	90.67±3.96	92.16±4.87	0.196
	End of CABG	90.12±6.53	91.58±4.44	0.313
	28 Days after CABG	103.14±10.21	104.15±11.66	0.722
	P-value	<0.001	<0.001	
	P-value for interaction	0.105		
	P-value for intervention	0.775		
MCH	Immediately after donation	30.79±1.79	30.75±1.88	0.926
	4 hours after donation	31.05±1.71	31.03±2.15	0.971
	28 Days after donation	30.88±1.84	30.67±1.71	0.651
	P-value	0.476	0.109	
	P-value for interaction	0.650		
	P-value for intervention	0.305		
MCHC	Immediately after donation	33.74±2.03	33.53±1.89	0.679
	4 Hours after donation	33.85±1.88	33.94±1.90	0.848
	28 Days after donation	29.85±2.27	29.74±2.67	0.864
	P-value	<0.001	<0.001	
	P-value for interaction	0.211		
	P-value for intervention	0.388		
Platelet	Immediately after donation	190.30±45.67	170.37±41.11	0.081
	4 Hours after donation	171.67±40.42	158.07±39.57	0.193
	28 Days after donation	154.17±89.87	139.17±53.04	0.434
	P-value	0.045	0.004	
	P-value for interaction	0.022		
	P-value for intervention	0.326		

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Variable	Time	Arterial blood	Venous blood	P-value
Urea	Immediately after donation	31.50±8.71	32.51±9.60	0.672
	4 Hours after donation	32.71±9.38	32.61±9.02	0.968
	28 Days after donation	36.73±11.22	37.48±9.92	0.786
	P-value		<0.001	
	P-value for interaction	0.523		
	P-value for intervention	0.192		
Creatinine	Immediately after donation	1.03±0.20	1.10±0.21	0.232
	4 Hours after donation	1.09±0.18	1.16±0.22	0.186
	28 Days after donation	1.18±0.27	1.22±0.28	0.623
	P-value	<0.001	<0.001	
	P-value for interaction	0.594		
	P-value for intervention	0.092		
LDH	Immediately after donation	228.20±70.37	245.60±68.50	0.336
	4 Hours after donation	240.23±95.93	259.07±92.10	0.441
	28 Days after donation	1,306.37±352.06	1,230.08±396.83	0.434
	P-value	<0.001	<0.001	
	P-value for interaction	0.133		
	P-value for intervention	0.818		
Albumin	Immediately after donation	3.33±0.56	3.51±0.46	0.170
	4 Hours after donation	3.23±0.52	3.36±0.56	0.350
	28 Days after donation	3.32±0.45	3.48±0.44	0.147
	P-value	0.363	0.010	
	P-value for interaction	0.579		
	P-value for intervention	0.443		

Table 2. Continued

 PCO_2 : partial pressure of carbon dioxide; PO_2 : partial pressure of oxygen; HCO_3 : bicarbonate; Na: sodium; K: potassium; RBC: red blood cell; WBC: white blood cell; MCV: mean corpuscular volume; MCH: mean corpuscular hemoglobin; MCHC: mean corpuscular hemoglobin; LDH: lactate dehydrogenase.

arterial group and P=0.109 in venous group). In addition, the changes in albumin were not significant in the arterial group (P=0.363) (Table 2). P-values for intervention showed the changes between the groups were not significantly different (Table 2).

As shown in Table 2, PCO₂ in the venous blood group was significantly higher than in the arterial blood group immediately after donating blood (57.25 ± 10.63 vs. 49.60 ± 8.33) and 4 hours after donating blood (62.23 ± 7.61 vs. 54.81 ± 7.79). However, after 28 days of storage, significant difference was not observed in PCO₂ between the groups (P=0.883).

In addition, partial pressure of oxygen (PO₂) in the arterial blood group was significantly higher than in the venous blood group immediately after donating blood (190.82±124.57 vs. 68.94±18.62) and 4 hours after donating blood (131.60±61.84 vs. 71.38±20.31). After 28 days of storage, significant difference was not observed in PO₂ between the groups (P=0.568). Significant differences were observed in pH (P=0.02) and bicarbonate (HCO₃; P=0.021) between the groups 4 hours after donating

blood. Immediately after donating blood, significant difference was observed in glucose (P=0.007) which was significantly lower in the arterial blood group than in the venous blood group (431.53 \pm 132.08 vs. 513.67 \pm 90.18) before CABG (Table 2). P-values for interaction showed no interaction between the groups in all parameters except PO₂ (P=0.033) and platelets (P=0.022) (Table 2).

DISCUSSION

Anemia affects one-quarter of the global population, nearly 50% of hospitalized patients, and up to 75% of hospitalized elderly patients. A high percentage of patients (60%–66%) become anemic during intensive care unit stay. Anemia is associated with a range of postoperative consequences, major side effects, rehospitalization, duration of stay in the intensive care unit, hospital stay, and mortality [8]. Therefore, blood transfusion is a vital and life-saving measure in many emergency situations [9].

In the present study, parameters such as albumin, LDH, Cr, urea, platelets, MCHC, MCH, Hb, white blood cells (WBCs), RBCs, lactate, K, Na, MCV, and hematocrit did not show significant differences at any time point between the groups. However, arterial pH and HCO₃ were higher than venous pH and HCO₃ after CABG. Arterial PCO₂ was lower than venous PCO₂ before and after CABG. Arterial PO₂ was significantly higher than venous PO₂ before and after CABG. In addition, arterial glucose was lower than venous glucose before CABG. The venous blood will have a lower PO₂, lower pH, and higher PCO₂ than the arterial blood due to metabolism in the tissues.

The results of the present study showed the mean values of arterial Na and K were lower than venous Na and K but without statistical significance; however, these differences were significant in another study [10]. Those differences were attributed to the effect of heparin because arterial samples were collected in heparinized containers [10].

The present findings showed the changes in arterial and venous RBCs, WBCs, Hb, MCV, MCHC, and platelets were significantly different after 28 days of storage. The WBCs significantly decreased in both groups after 28 days of storage. Similarly, in previous studies, significant reduction in WBCs was observed after 35 days of storage [11,12]. In addition, WBCs were shown to constantly decrease during the 28-day storage period [13].

The results of the present study showed that MCV significantly increased in both groups, which is in agreement with previous studies in which MCV was significantly increased during storage [11,14]. The RBCs, Hb, MCHC, and platelets significantly decreased after 28 days of storage. However, albumin showed significant decrease only in the venous group after 28 days of storage.

The decreased levels might be due to hemolysis that occurs during storage. In a previous study, significant reduction was observed in RBCs at day 28 and further significantly decreased during 35 days of storage which was in agreement with our finding [11]. However, the results of the present study are in contrast to a previous study in which statistically significant changes were not observed in RBCs during the 28-day storage period [15]. During blood storage at 2 °C-6 °C, glycolysis is reduced and the amount of ATP is also reduced, thus, the structure and volume of the RBCs are changed [16].

In a previous study, Hb concentration gradually decreased during the 35-day storage period, which is in agreement with our findings and might be due to hemolysis that occurs during storage [14]. Significant reduction in MCHC was found in a previous study, similar to the results of the present study [11]. As reported in previous studies, the changes in MCH and MCHC can be the result of a deregulated mechanism of cell volume, which causes the increase in the volume of RBCs, increasing hypochromia and anisocytosis. Therefore, the decrease in MCHC apparently is the result of an increase in cell volume [17].

In the present study, platelets were significantly decreased after 28 days of storage. This finding was in agreement with Marabi et al. [11], and can be explained by the cells losing viability due to ATP depletion in addition to platelet utilization caused by micro-aggregate development [18].

Contradictory results have been reported for hematocrit. A slight decrease in hematocrit was found in both groups in the present study but without statistical significance. In a previous study, significant increase in hematocrit was observed at day 35 of storage [11], however, this finding was refuted in another study [14]. Decrease in hematocrit during storage can be attributed to depletion of RBC ATP or morphological alterations that occur during blood storage [14,19]. In addition, MCH changes were insignificant in both groups which is in agreement with a study in which the MCH changes were insignificant during 28 days of storage [15]. However, MCH reportedly decreased during the storage period in another study [20].

In the present study, significant increase in lactate was observed after 28 days of storage in both groups. Whole blood stored for more than 14 days was proposed to have reduced efficacy with increased markers of red cell storage lesion, such as increased K level, lactate, and decreased pH [1]. Based on previous studies, a decrease in pH is the direct result of the increase in lactate level due to anaerobic metabolism of glucose [21-23]. In this study, the changes in Na and K were significantly different in each group. The results showed significant decrease in Na and significant increase in K in both groups after 28 days of storage. In agreement with our results, Antwi-Baffour et al. [24] demonstrated that K concentration in refrigerated blood increases and plasma Na concentration decreases with storage time. The present findings indicated a significant reduction in glucose and HCO₃ after 28 days of storage. However, other parameters such as LDH, urea, and Cr significantly increased in both groups after 28 days of storage. A significant increase in K concentration and decrease in HCO₃ levels from 1 day to 21 days of storage were reported in another study which is in agreement with the results of the present study [25].

In other studies, decreased concentration of plasma glucose and HCO_3 and increase in K, lactate, and LDH concentration with storage were observed which confirms the results of the present study [26]. Contrary to the present study, Verma et al. [25] reported no significant changes in urea and Cr during 21 days of whole blood storage. The glucose concentration is limited and glucose is used during storage, thus, ATP depletion and decrease in red cell viability occur concomitantly. Therefore, the energy required for operating the ATPase pump in RBCs decreases over time. Inhibition of the Na pump results in hyperkalemia and hyponatremia as observed in the present study [25]. As a result of glycolysis, the concentration of lactic acid increases which may cause a decrease in pH, subsequently significantly increasing LDH. In addition, hemolysis results in the increased concentration of LDH and the decreased concentration of RBCs, Hb, MCHC, and platelets in plasma [1,25].

Based on the results, although significant biochemical changes occurred in the donated arterial and venous blood groups during the blood storage period, the process of biochemical changes between the two groups did not show any significant difference. Therefore, arterial blood donation for blood banking cannot be recommended over venous blood donation.

CONFLICT OF INTEREST

No potential conflict of interest relevant to this article was reported.

FUNDING

None.

ACKNOWLEDGMENTS

None.

ORCID

Mojtaba Mansourihttps://orcid.org/0000-0002-5725-3012Alireza Yazdanihttps://orcid.org/0000-0001-9941-0699Gholamreza Masoumihttps://orcid.org/0000-0003-1399-1782Mohsen Mirmohammadsadeghi

https://orcid.org/0000-0002-5996-407X Amir Mirmohammadsadeghi

https://orcid.org/0000-0001-9212-0138

AUTHOR CONTRIBUTIONS

Conceptualization: AY. Data curation: AY. Formal analysis: GM. Methodology: AY, MM. Project administration: MM. Visualization: AY, AM. Writing–original draft: AY, MM. Writing– review & editing: AY, GM, MM.

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