Effects of anemia and blood transfusion in acute myocardial infarction in rats

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BACKGROUND: The optimal hemoglobin (Hb) level in acute myocardial infarction (MI) is unknown. The goal of this study was to determine the optimal Hb concentration in acute MI and whether transfusion of fresh blood to correct anemia reduces myocardial injury and improves outcome.

STUDY DESIGN AND METHODS: Anemia was induced in rats by an iron-deficient diet and phlebotomy. MI was induced by left coronary artery ligation. Some rats received transfusion of fresh blood. Survival, hemodynamic measurements, and infarct size were determined 24 hours after MI.

RESULTS: Reduction of Hb to 80 to 90 and 70 to 80 g/L decreased 24-hour survival after MI to 42 and 47%, respectively ($p < 0.05$). Infarct size was increased in both 70 to 80 and 80 to 90 g/L anemic groups compared to the normal Hb group ($p < 0.05$). Cardiac function was decreased in anemic groups after MI ($p < 0.01$). Transfusion of fresh blood to increase Hb from 80 to 90 g/L to 100 g/L decreased infarct size ($p < 0.05$) and improved cardiac function ($p < 0.05$), and a trend toward better survival (73%) was observed. Transfusion from 80 to 90 g/L Hb to 120 g/L Hb was associated with larger infarct size ($p < 0.05$), decreased cardiac function ($p < 0.05$), and no improvement in survival (47%, p = NS).

CONCLUSION: Anemia increases infarct size and decreases cardiac function and survival in acute MI. Transfusion of anemic animals up to 100 g/L Hb with fresh blood reduces infarct size and improves cardiac function. However, transfusion to 120 g/L Hb did not demonstrate any additional benefit and was associated with larger infarcts.

The optimal hemoglobin (Hb) level in the setting of acute myocardial infarction (MI) is unknown. Anemia reduces the oxygen carrying capacity of blood and may theoretically exacerbate ischemia and increase myocardial injury after MI. The healthy host can tolerate a remarkable degree of normovolemic anemia to Hb levels as low as 50 g/L without significant organ dysfunction.[1] Since the myocardium has a basal oxygen extraction ratio of 55% to 70%, it has little capacity to increase oxygen extraction in response to anemia.[2] Thus, anemic patients compensate by increasing coronary blood flow, which may be limited in patients with coronary artery disease. Rheologically, increased Hb concentration can increase blood viscosity, which results in increased myocardial work and decreased O_2 delivery. Transfusion is the only available therapeutic intervention

ABBREVIATIONS: LV = left ventricle; LVEDP = left ventricular end-diastolic pressure; MAP = mean arterial pressure; MI = myocardial infarction.

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that can rapidly correct anemia in the setting of acute myocardial ischemia. Not surprisingly, without a known therapeutic target concentration for Hb, the benefit of transfusion to correct anemia in acute coronary syndromes is also controversial. Two large retrospective studies yielded conflicting results. Wu and colleagues[3] found that blood transfusion in patients over the age of 65 with a hematocrit (Hct) level of less than 30% (estimated Hb, 100 g/L) on admission reduced 30-day mortality. In contrast, a retrospective post hoc analysis of 24,112 patients with acute coronary syndrome from three large clinical trials (the GUSTO IIb, PURSUIT, and PARAGON B trials) showed that transfusion was associated with a higher mortality for Hct levels of greater than 25%, even after adjustment for other predictive factors and timing of events.[4]

The notion that a lower transfusion threshold may be beneficial is also supported by a multicenter randomized trial comparing two transfusion strategies in 838 critically ill patients.[5] Patients randomized to the restrictive transfusion strategy (transfusion threshold, <70 g/L; target Hb level, 70-90 g/L) demonstrated a trend toward a lower 30-day mortality rate than patients randomized to the liberal transfusion group (transfusion threshold, <100 g/L; target Hb level, 100-120 g/L; 23.3% vs. 18.7%, p = 0.11). In a retrospective follow-up analysis, a subpopulation of 357 patients with severe ischemic heart disease had a lower but nonsignificant absolute survival rate in the restrictive group compared to the liberal transfusion strategy group.[6] Based on the above observational studies[3] and the retrospective subgroup analysis,[6] it is not possible to make any definitive recommendation regarding the transfusion threshold in patients with ischemic heart disease. A recent prospective cohort study, examining the transfusion threshold in acute MI, demonstrated that transfusion in patients with a nadir Hb level of 80 g/L or less had better survival while transfusion with a nadir Hb level of greater than 80 g/L was associated with worse outcome,[7] suggesting that transfusion in patients with acute MI and Hb level of less than 80 g/L may be appropriate. However, differential bleeding events after thrombolytic treatment between the two groups of patients may have conferred adverse outcome in the transfused patients.[7,8] While further randomized clinical trials are needed to determine the threshold transfusion Hb for patients with acute MI, it is important to understand the effects of different levels of anemia and blood transfusion on cardiac function after acute MI.

In this study, we examined the effect of anemia and transfusion on myocardial infarct size and cardiac function using a rat model of MI. The goal of this study was to determine the optimal Hb concentration in acute MI and whether transfusion of fresh blood to correct anemia reduces myocardial injury and improves outcome. The results from this study may provide valuable information in the design of future clinical trials for the determination of transfusion threshold in patients with acute MI.

MATERIALS AND METHODS

Experimental animals and induction of anemia

The experiments were conducted using male Sprague-Dawley rats (170-190 g). All animals were provided water and food ad libitum and housed in a temperature- and humidity-controlled facility with 12-hour light and dark cycles. The investigation conforms to the *Guide for the Care and Use of Laboratory Animals* published by the US National Institutes of Health (NIH Publication No. 85-23, revised 1996). Experimental protocols were approved by the Animal Use Subcommittee at the University of Western Ontario.

To induce anemia, rats were fed with an iron-deficient diet (10-20 ppm; TestDiet 5859, Richmond, IN) previously described by Strube and coworkers[9] in combination with phlebotomy (2-3 mL) from the jugular vein twice weekly to reach target Hb levels of 70 to 80 or 80 to 90 g/L. Immediately after phlebotomy, rats were infused with an equal volume of 10% pentastarch (Bristol-Myers Squibb Canada, Montreal, Quebec, Canada).

Experimental design and transfusion

Rats were randomly assigned to eight experimental groups:

- Group 1: normal 140 to 150 g/L Hb, sham operation (n = 5);
- Group 2: normal 140 to 150 g/L Hb, MI (n = 10);
- Group 3: 80 to 90 g/L Hb, sham operation (n = 7);
- Group 4: 80 to 90 g/L Hb, MI (n = 19);
- Group 5: 70 to 80 g/L Hb, sham operation (n = 9);
- Group 6: 70 to 80 g/L Hb, MI (n = 15);
- Group 7: 80 to 90 g/L Hb, MI with transfusion to Hb level of 100 g/L (n = 11);
- Group 8: 80 to 90 g/L Hb, MI with transfusion to Hb level of 120 g/L (n = 17).

In the subgroup of anemic animals receiving blood, transfusion was performed immediately after coronary artery ligation surgery to increase their Hb levels to a target of 100 or 120 g/L using fresh blood (<4 hr stored) obtained from a donor rat. Donor blood was collected and stored as previously described.[10] Briefly, rats were anesthetized with intramuscular injection of ketamine (60 mg/kg) and xylazine (10 mg/kg), and the carotid artery was cannulated to retrieve the maximum amount of blood. Blood was collected into a 20-mL syringe with 3 mL of citrate-phosphate-dextrose-adenine (Baxter, Toronto, Ontario, Canada) and injected into a sterile blood collection bag (Fenwal, Baxter, Toronto, Ontario, Canada). The

blood was kept in a refrigerator at 4°C for less than 4 hours. Right before transfusion, the stored nonleukoreduced blood was drawn from the collection bag and centrifuged at $500 \times g$ for 5 minutes at 4°C. The supernatant was removed to achieve a Hct of 75%. Total volume infused in approximately 30 to 45 minutes to achieve 100 and 120 g/L Hb was 3 and 5 mL, respectively. Thirty minutes after transfusion, Hb levels in the recipient rats were measured from 10 μL of blood obtained through a saphenous vein by a spectrophotometric technique using a Hb assay kit (Pointe Scientific, Canton, MI).

Induction of MI

MI was induced by ligation of the left descending coronary artery as described in our previous studies.[11] Briefly, rats were anesthetized with intramuscular injection of ketamine (60 mg/kg) and xylazine (10 mg/kg). Animals were then intubated and artificially ventilated. A left thoracotomy was performed to expose the heart and left anterior descending coronary artery was ligated by positioning a 6-0 silk suture between the pulmonary artery outflow tract and the left atrium. The lungs were thereafter hyperinflated using positive end-expiratory pressure and the thorax was immediately closed. Sham-operated rats underwent the same surgical procedure without coronary artery ligation. Animals were caged individually after each surgical operation.

Hemodynamic and infarct size measurements

Hemodynamic measurements to assess in vivo cardiac function were performed as previously described 24 hours after coronary artery ligation.[11] Rats were reanesthetized with ketamine and xylazine, and a polyethylene catheter (PE-50) was inserted into the right carotid artery to record the arterial pressure. The catheter was then advanced retrograde into the left ventricle (LV) to record LV pressures for assessment of cardiac function. Pressure signals were fed to an analog digital converter and collected by a computer. Heart rate, LV systolic pressure, LV end-diastolic pressure (LVEDP), and maximal positive and minimal negative first derivatives of LV pressure ($+dP/dt_{max}$ and $-dP/dt_{min}$) were analyzed by computer software (PowerLab Chart, ADInstruments, Colorado Springs, CO). LVEDP is the LV filling pressure during diastole. LV $+dP/dt_{max}$ and $-dP/dt_{min}$ indicate contractile and relaxation function of the LV, respectively.

Infarct size was determined according to our previous report.[12] Briefly, after hemodynamic measurements, 3 mL of Evans blue dye (2% in saline) was injected into the aortic root to stain the area of the myocardium perfused by the patent coronary arteries, thereby delineating the area at risk by negative staining. The heart was excised and the LV was carefully isolated and sliced transversely into sections 3 to 4 mm thick. Sections were weighed and incubated in 5% triphenyltetrazolium chloride in phosphate-buffered saline at 37°C for 15 minutes to stain the viable myocardium. Photographs of heart slices were taken with a digital camera. The nonrisk area (stained by Evans blue), risk area (unstained by Evans blue), and infarct area (unstained by triphenyltetrazolium chloride) were quantitated using an image analysis system (Sigma ScanPro, Ashburn, VA). Percent weight of nonrisk, risk, and infarct area was calculated. Infarct size was expressed as a percentage of the area at risk.

Statistical analysis

Data are expressed as the means ± standard error of the mean (SEM). Two-way analysis of variance (ANOVA) was performed followed by Bonferroni test. p Values less than 0.05 were considered significant.

RESULTS

Body weight and Hb

All animals had a similar body weight and Hb at baseline (Table 1). Animals fed an iron-deficient diet in combination with phlebotomy had significantly decreased Hb

TABLE 1. Body weight and Hb at baseline and after 2 weeks of treatment with an iron-deficient diet in combination with bleeding before surgery in the eight experimental groups*

	Body weight (g)		Hb (g/L)	
Group	Baseline	Before surgery	Baseline	Before surgery
140-150 g/L Hb, sham (n = 5)	176.8 ± 4.5	271.6 ± 3.8	146.8 ± 6.7	159.2 ± 4.1
140-150 g/L Hb, MI (n = 10)	169.6 ± 8.8	268.2 ± 5.9	152.3 ± 3.2	149.0 ± 2.6
80-90 g/L Hb, sham (n = 7)	177.0 ± 2.8	256.1 ± 6.1	146.1 ± 5.0	87.3 ± 2.6
80-90 g/L Hb, MI (n = 19)	175.0 ± 6.5	267.1 ± 7.5	151.4 ± 3.3	87.7 ± 1.2
70-80 g/L Hb, sham (n = 9)	187.3 ± 0.9	258.4 ± 1.4	150.6 ± 2.2	69.0 ± 1.3
70-80 g/L Hb, MI (n = 15)	189.5 ± 0.5	259.2 ± 1.7	148.7 ± 1.9	70.0 ± 1.2
80-90 g/L Hb, MI + T100 (n = 11)	184.0 ± 3.6	271.3 ± 3.9	153.1 ± 3.7	82.7 ± 1.6
80-90 g/L Hb, MI + T120 (n = 17)	189.8 ± 1.0	266.1 ± 1.1	148.4 ± 2.2	83.5 ± 1.1

* Data are mean ± SEM. Body weight and Hb are not statistically different between sham and MI at the same Hb levels.
T100 = transfusion to reach 100 g/L Hb; T120 = transfusion to reach 120 g/L Hb.

levels compared to the standard diet group by the end of a 2-week period before surgery, with mean Hb levels reaching the targets levels of 70 to 80 and 80 to 90 g/L, respectively. After transfusion of anemic animals, the Hb reached 101.9 ± 1.2 and 120.3 ± 6.3 g/L in the 100 and 120 g/L transfusion groups, respectively.

Effects of anemia and transfusion on survival after MI

After MI, 24-hour survival was monitored. In the normal Hb group (140-150 g/L), survival after sham or MI surgery was 100% (Fig. 1A). In the 80 to 90 g/L Hb group, survival was significantly decreased after MI (8/19, 42%) compared to sham 100% ($p < 0.05$). In the 70 to 80 g/L Hb group, one sham rat (1/9) died after surgery while MI was associated with eight deaths in 15 rats (survival, MI 47% vs. sham 89%, $p < 0.05$). Survival after MI in both 80 to 90 and 70 to 80 g/L groups was significantly decreased compared to the normal Hb group ($p < 0.05$, Fig. 1A).

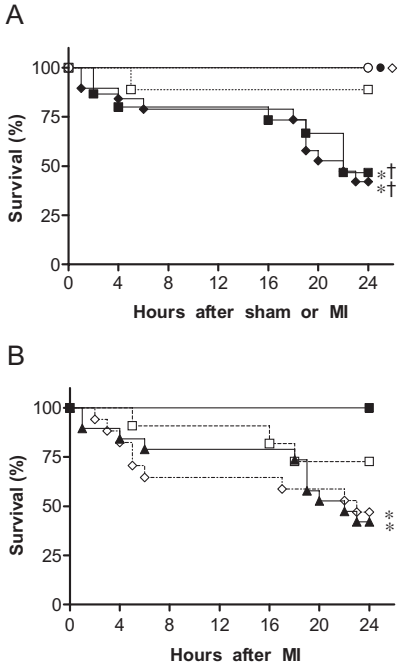

Fig. 1. Effects of Hb on survival in rats after MI. Rats were fed with an iron-deficient diet in combination with phlebotomy from the jugular vein twice weekly to reach target Hb levels of 70 to 80, 80 to 90, or 140 to 150 g/L. MI was induced by ligation of the left descending coronary artery. (A) Survival was monitored for 24 hours after surgery. n = 5 to 19, *p < 0.05 versus corresponding sham, †p < 0.05 versus 140 to 150 g/L Hb, MI. (○) 140 to 150 g/L Hb, sham; (●) 140 to 150 g/L Hb, MI; (◇) 80 to 90 g/L Hb, sham; (□) 70 to 80 g/L Hb, sham; (■) 70 to 80 g/L Hb, MI; (◆) 80 to 90 g/L Hb, MI. (B) Transfusion was performed in rats with 80 to 90 g/L Hb immediately after MI to increase Hb levels to 100 or 120 g/L using fresh blood. Survival was monitored in the subsequent 24 hours. n = 10 to 19, *p < 0.05 versus 140 to 150 g/L Hb, MI. (■) 140 to 150 g/L Hb; (▲) 80 to 90 g/L Hb; (□) transfusion to 100 g/L Hb; (◇) transfusion to 120 g/L Hb.

To study whether blood transfusion had any effect on the outcome, fresh blood was immediately transfused after MI. There was a trend of better survival after transfusion to increase the Hb from 80 to 90 g/L (42%, 8/19) to 100 g/L (73%, 8/11), but no statistical difference was observed between the two groups ($p = 0.16$, Fig. 1B). Furthermore, survival in transfusion to 100 g/L group was not significantly decreased from the normal Hb group (73% vs. 100%, p = NS). Transfusion from 80 to 90 g/L Hb to 120 g/L Hb showed no significant improvement in survival (47%, 8/17) compared to untransfused anemic (80-90 g/L Hb) animals (42%, p = NS, Fig. 1B). Additionally, transfusion to 120 g/L was associated with significantly higher mortality compared to the normal Hb group ($p < 0.05$).

Effects of anemia and transfusion on infarct size after MI

Twenty-four hours after MI, areas at risk to LV weight ratios were not significantly different between any groups indicating similar area of myocardial ischemia among all groups (p = NS, Fig. 2A). However, infarct-to-risk weight ratios were significantly increased in both 70 to 80 and 80 to 90 g/L groups compared to the normal Hb group ($p < 0.05$, Fig. 2B).

Area at risk-to-LV weight ratios were similar among all groups in Fig. 3A. Transfusion from 80 to 90 g/L Hb to 100 g/L Hb significantly decreased infarct to risk weight ratios compared to the 80 to 90 g/L Hb group ($p < 0.05$). However, transfusion to 120 g/L Hb resulted in significantly larger infarct size compared to the 100 g/L Hb group ($p < 0.05$, Fig. 3B).

Effects of anemia and transfusion on cardiac function after MI

Cardiac function was determined at 24 hours after MI. Heart rate, mean arterial pressure (MAP), left ventricular systolic pressure (LVSP), and LVEDP were not significantly different among all groups (Table 2). Reduction in Hb from 140 to 150 g/L to 80 to 90 g/L and 70 to 80 g/L showed a significant decrease in LV $+dP/dt_{max}$ and $-dP/dt_{min}$ in both sham and MI rats ($p < 0.01$, Fig. 4). However, a further significant decrease in LV $+dP/dt_{max}$ and $-dP/dt_{min}$ was seen in MI rats compared to sham-operated rats ($p < 0.05$, Fig. 4).

Transfusion of fresh blood after MI from 80 to 90 g/L Hb to 100 g/L Hb significantly increased LV $+dP/dt_{max}$ ($p < 0.05$) with no significant changes in LV $-dP/dt_{min}$ (p = NS, Fig. 5). Blood transfusion to 120 g/L Hb did not

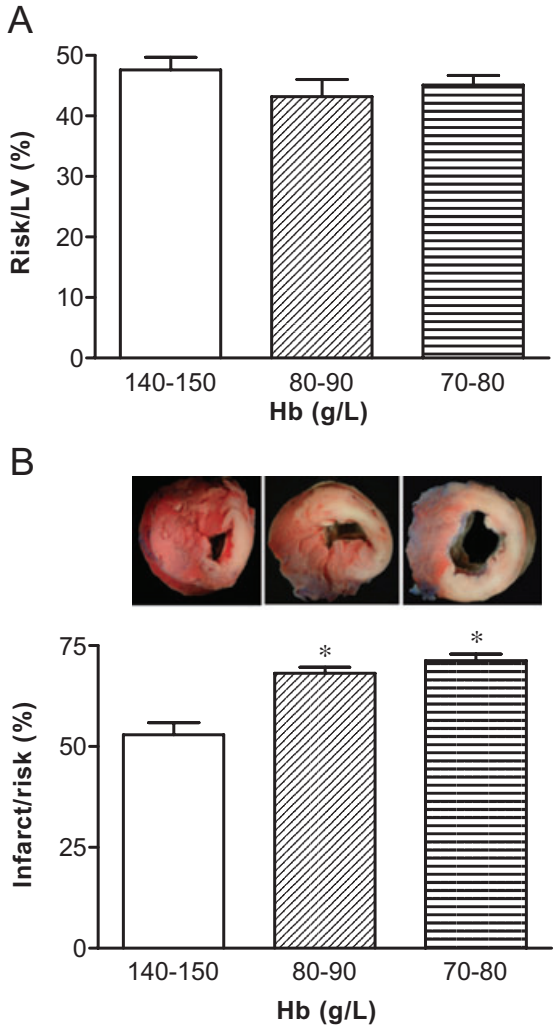

Fig. 2. Effects of anemia on infarct size after MI. (A) Percent weight of risk area to LV weight. (B) Infarct size expressed as percentage of infarct to risk weight. Inserted images are representative sections of triphenyltetrazolium chloride–stained hearts corresponding to each group. Data are mean ± SEM from 7 to 10 rats per group. *p < 0.01 versus 140 to 150 g/L Hb.

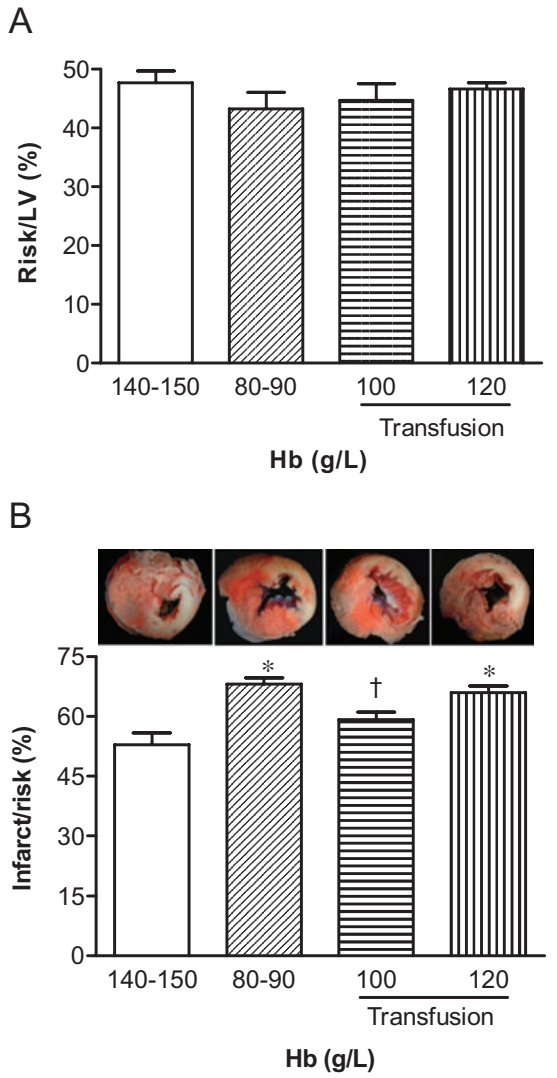

Fig. 3. Effects of transfusion on infarct size after MI in anemic rats. Transfusion was performed in the rats with 80 to 90 g/L Hb immediately after MI to reach Hb levels to 100 or 120 g/L. Infarct size was determined 24 hours after MI. (A) Percent weight of risk area to LV weight. (B) Infarct size expressed as a percentage of infarct to risk weight. Inserted images are representative sections of triphenyltetrazolium chloride–stained hearts corresponding to each group. Data are mean ± SEM from 8 to 10 rats per group. *p < 0.01 versus 140 to 150 g/L Hb, †p < 0.05 versus 80-90 g/L Hb.

result in any further improvement in cardiac function. In fact, LV $+dP/dt_{max}$ and $-dP/dt_{min}$ tended to decrease from transfusion to 100 g/L Hb group but was not significantly different (p = NS, Fig. 5).

DISCUSSION

This study demonstrated that anemia increases infarct size and decreases cardiac function and survival in acute MI induced by coronary artery ligation in rats. Transfusion of anemic animals up to 100 g/L Hb with fresh blood decreases infarct size and improves cardiac function, with a trend toward better survival. However, transfusion to 120 g/L Hb did not demonstrate any additional benefit and was associated with significantly larger infarcts. Our study demonstrates that anemia strongly correlates with survival, infarct size, and LV function in the setting of acute MI. Myocardial tissue oxygenation is dependent on three factors: coronary blood flow, the oxygen-carrying capacity or microcirculatory Hct, and the density of perfused capillaries, which affects the diffusion distance in the tissue.[13-15] As coronary ligation immediately reduces blood flow to a similar degree in all groups of animals, placing a similar volume of myocardial tissue at risk, the

TABLE 2. Changes in heart rate, MAP, LVSP, and LVEDP in eight experimental groups 24 hours after MI*

Group	Heart rate (bpm)	MAP (mmHg)	LVSP (mmHg)	LVEDP (mmHg)
140-150 g/L Hb, sham (n = 5)	354.8 ± 9.0	89.8 ± 3.9	125.2 ± 6.5	7.6 ± 2.2
140-150 g/L Hb, MI (n = 7)	358.2 ± 21.3	82.9 ± 4.7	101.8 ± 5.9	7.5 ± 2.1
80-90 g/L Hb, sham (n = 7)	349.1 ± 11.4	78.1 ± 2.9	112.4 ± 4.5	8.7 ± 1.5
80-90 g/L Hb, MI (n = 8)	341.1 ± 12.5	76.7 ± 6.7	93.4 ± 6.6	9.8 ± 1.8
70-80 g/L Hb, sham (n = 7)	307.4 ± 6.1	79.8 ± 2.9	109.6 ± 2.8	9.9 ± 1.9
70-80 g/L Hb, MI (n = 7)	318.4 ± 12.3	74.0 ± 4.9	108.9 ± 4.7	13.1 ± 1.6
80-90 g/L Hb, MI + T100 (n = 7)	360.7 ± 13.8	91.0 ± 2.5	112.7 ± 3.4	11.2 ± 1.2
80-90 g/L Hb, MI + T120 (n = 8)	354.8 ± 12.2	85.8 ± 4.5	123.0 ± 4.4	11.6 ± 1.2

* Data are mean ± SEM. Two-way ANOVA showed no significance among any groups. Measurements were made in surviving animals. T100 = transfusion to reach 100 g/L Hb; T120 = transfusion to reach 120 g/L Hb.

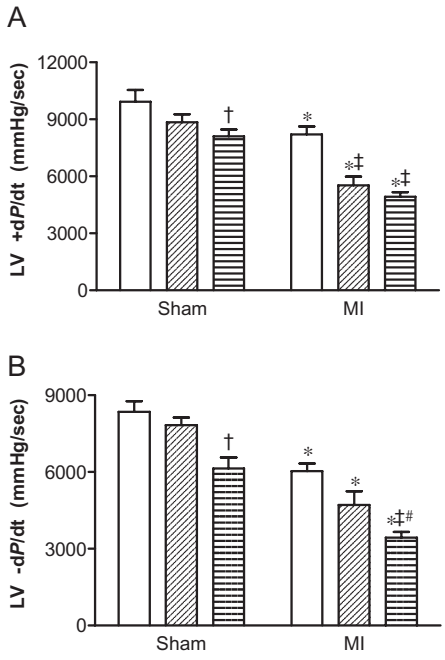

Fig. 4. Effects of anemia on LV function after MI. Hb levels in rats were reduced from 140 to 150 g/L to 70 to 80 g/L or 80 to 90 g/L. LV ±d*P*/dt were measured 24 hours after MI. (A) LV +d*P*/dt. (B) LV –d*P*/dt. Data are mean ± SEM from five to eight rats per group. *p < 0.01 versus corresponding sham; †p < 0.05 versus 140 to 150 g/L Hb, sham; ‡p < 0.01 versus 140 to 150 g/L Hb, MI; #p < 0.05 versus 80-90 g/L Hb, MI. (□) Hb 140-150 g/L; (▨) Hb 80-90 g/L; (▤) Hb 70-80 g/L.

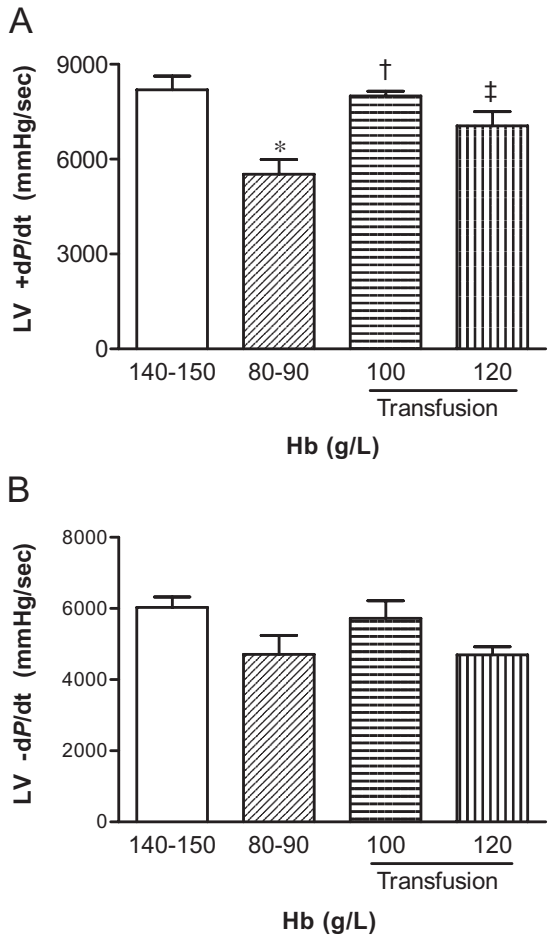

Fig. 5. Effects of transfusion on LV function. Hb levels in rats were reduced from 140 to 150 g/L to 80 to 90 g/L. Transfusion was performed in the rats with 80 to 90 g/L Hb immediately after MI to reach Hb levels to 100 or 120 g/L. LV ±d*P*/dt were measured 24 hours after MI. (A) LV +d*P*/dt. (B) LV –d*P*/dt. Data are mean ± SEM from seven to eight rats per group. *p < 0.01 versus 140-150 g/L Hb, †p < 0.01 versus 80 to 90 g/L Hb, ‡p < 0.05 versus 80 to 90 g/L Hb.

observed difference in infarct size in anemic groups can be attributed to a reduction in capillary Hct or density. Although we do not have data to show these changes in capillary density, improvement in cardiac function and reduction in infarct to risk ratio after transfusion support the direct effect of a relatively modest degree of anemia (80-90 g/L Hb) on myocardial ischemia.

The effect of modest anemia on survival and infarct size is likely a consequence of the interaction between coronary stenosis/ligation and anemia. In the healthy host, a number of clinical[1] and animal experiments[13-17]

suggest a remarkable physiologic tolerance for normovolemic anemia. To a large extent, the physiologic reserve in normovolemic anemia is maintained by the healthy host through increases in cardiac output and O_2 extraction and shunting from the noncritical sites such as splanchnic circulation to vital organs.[13-17] Levy and colleagues[17,18] demonstrated that aerobic metabolism and cardiac function can be maintained over a wide range of Hct levels, reaching as low as 10% (estimated Hb, 33 g/L) in normal dogs. However, in animals with experimentally induced coronary artery stenosis, cardiac dysfunction occurs at higher Hct levels (20%; estimated Hb, 66 g/L). Compensation for acute normovolemic anemia occurs at the expense of regional blood flow, with initial shunting from kidney and splanchnic vascular beds to maintain coronary and cervical cord blood flow. But in the setting of coronary stenosis, loss of vasodilatory reserve leads to reduction in myocardial blood flow, potentially leading to myocardial ischemia as Hct approaches 20%.[19] Our results indicate that mild anemia with Hb levels as high as 80 to 90 g/L places vulnerable myocardium at risk resulting in increased infarct size and reduced overall survival after MI.

The effect of fresh rat blood transfusion to different target Hb levels of 100 and 120 g/L showed disparate results. Transfusion immediately after MI from Hb 80 to 90 g/L to target of 100 g/L significantly decreased infarct size and improved cardiac function compared to the 80 to 90 g/L Hb group. However, transfusion to a higher level of 120 g/L Hb resulted in significantly greater infarct size compared to the 100 g/L Hb group ($p < 0.05$, Fig. 3). The lack of beneficial effect beyond a Hb threshold of 100 g/L might be explained in terms of coronary capillary microhematocrit, which is a major determinant of myocardial oxygenation. Capillary Hct is approximately 75% of systemic Hct and is generally well maintained over a wide range in Hb levels from normal to mild anemia.[20] A change in Hb level from 100 to 120 g/L is therefore unlikely to have much influence on capillary microhematocrit and affect O_2 delivery significantly. Thus a transfusion target of 100 g/L in this population is supported; however, higher levels of Hb are unlikely to provide additional benefit. Our experimental studies support the clinical observations that modest degree of anemia can affect mortality in setting of coronary ischemia and that optimal Hb for this population might be 100 g/L as opposed to the restrictive threshold of 70 g/L.[3,7,21] The question remains why animals transfused to 120 g/L demonstrated increased infarct size and mortality. A number of studies have linked RBC transfusion and possibly the RBC storage lesion to poor outcome.[22,23] The RBC transfusion used in this experiment was all fresh nonleukoreduced blood (defined as <4 hr storage) and less likely to have undergone all of the changes associated with prolonged storage.[10,24] It does not preclude some of the early changes seen in storage of rat blood such as changes in RBC pH and S-nitrosylation of Hb.[25,26] Similarly the expected changes in blood viscosity with transfusion to higher targets may be a factor but unlikely to be a major factor at modest Hb increment to a level of 120 g/L.

Anemia and transfusion also influence cardiac function. At 24 hours after MI, heart rate, MAP, LVSP, and LVEDP were not significantly different among all groups. However, anemic groups 80 to 90 g/L Hb and 70 to 80 g/L Hb showed a decrease in LV systolic contractility and diastolic relaxation in both sham and MI rats with a greater effect in animals after MI. Transfusion after MI from 80 to 90 g/L Hb to 100 g/L Hb significantly improved LV function. Transfusion to 120 g/L Hb did not result in any further improvement in cardiac function. In fact, LV $+dP/dt_{max}$ and $-dP/dt_{min}$ tended to decrease from transfusion to the 100 g/L Hb group but were not significantly different ($p = NS$). Impaired cardiac function with anemic groups is likely a consequence of increased myocardial ischemia, which can contribute to both diastolic and systolic dysfunction.[18] It should be noted that transfusion volume was 3 and 5 mL in the 100 and 120 g/L groups, respectively. It is difficult to control transfusion volumes to the same level in all study groups because infusion of colloid solution in the anemic groups may further reduce Hb concentrations acutely due to hemodilution. Thus, we cannot exclude the possibility that the difference in transfusion volume may potentially have influenced our study results.

The results from our animal studies support the clinical observations that anemia may contribute to incidence of heart failure in the setting of coronary artery disease. In a retrospective study, patients with coronary artery disease and anemia had a greater incidence of heart failure (31% vs. 18%), arrhythmia (41% vs. 16%), and higher mortality (13% vs. 4%) than a matched cohort without anemia.[27] In 6635 patients in the Studies of Left Ventricular Dysfunction (SOLVD) database, a 1% decrease in Hct was associated with a 2.7% higher risk for all-cause mortality after adjusting for traditional cardiovascular risk factors.[28] Felker and coworkers[29] noted that in patients with heart failure, the risk of death or rehospitalization within 60 days increased by 13% for every 10 g/L decrease in Hb independent of other risk factors. In a retrospective cohort study of 300 Jehovah's Witnesses who declined transfusion and had Hb levels of 80 g/L or less, the 30-day postoperative mortality was 2.5 times higher (95% CI, 1.9-3.2) for each 10 g/L decrease in Hb.[30] In a case-control study using electrocardiographic monitoring in the postoperative period, Nelson and colleagues[31] observed a high frequency (10/13 patients) of myocardial ischemia and morbid cardiac events in patients with Hct levels below 28% (estimated Hb, 90 g/L).

In conclusion, this study demonstrated that anemia increases infarct size and decreases cardiac function, leading to increased mortality after MI in rats. Transfusion of fresh blood to 100 g/L Hb but not 120 g/L reduces these

effects. Our study suggests that 100 g/L Hb may be the optimal target for blood transfusion after MI in rats. Direct extrapolation of these results to patients should be cautioned due to several technical limitations of our animal model. First, MI was induced by coronary artery ligation without reperfusion while patients with acute coronary syndromes develop thrombosis as a result of atherosclerosis and eligible patients are routinely reperfused by thrombolysis or angioplasty. Second, the timing of the transfusion intervention in the experimental model, immediately after MI, would be unusual for patients with MI who typically receive transfusions later in their hospital course. Finally, our study is underpowered to detect a significant survival benefit of transfusion. Therefore, further clinical studies are required to determine transfusion threshold and optimal Hb concentrations in anemic patients with acute MI.

CONFLICT OF INTEREST

None for all authors.

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