Use of hemoglobin raffimer for postoperative life-threatening anemia in a Jehovah's Witness

[L'usage d'un raffimère d'hémoglobine en cas d'anémie postopératoire grave chez un témoin de Jéhovah]

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Purpose: To describe the successful treatment of acute, life-threatening anemia with the oxygen therapeutic agent, hemoglobin (Hb) raffimer.

Clinical features: A 53-yr-old female Jehovah's Witness developed severe anemia following total hip replacement. Due to prior patient directive, red blood cells were not transfused. Tachycardia, hypotension, electrocardiographic abnormalities and mental status changes developed with a nadir Hb concentration of 3.2 $g \cdot dL^{-1}$. Hb raffimer is a purified, cross-linked, human Hb solution developed as a substitute for red blood cell Hb. After obtaining informed consent as well as Food and Drug Administration and Institutional Review Board approval for compassionate use, 2 L of Hb raffimer (Hemolink™, Hemosol, Inc., Toronto, ON, Canada) were administered along with ferrous sulfate and epoetin alfa therapy. The patient's Hb level rose to 5.5 $g \cdot dL^{-1}$ with resolution of symptoms. To allow recovery of red blood cell mass while maintaining Hb level > 4.5 $g \cdot dL^{-1}$, additional 1000 mL doses of Hb raffimer were administered on postoperative days three, five and seven (total dose = 500 g Hb). The patient developed no serious adverse events related to treatment with Hb raffimer. By postoperative day 14, the patient's Hb level increased to 6.5 $g \cdot dL^{-1}$ with a hematocrit of 23%. The patient was discharged.

Conclusions: Use of Hb raffimer as a bridge to recovery of this patient's red blood cell mass may have prevented adverse clinical outcome. Because this product is a purified Hb solution devoid of other cellular components, it may be accepted as therapy by patients who, due to religious conviction, refuse allogeneic red blood cell transfusion.

Objectif : *Décrire le traitement réussi d'une grave anémie aiguë avec un agent d'oxygénothérapie, un raffimère d'hémoglobine (Hb).*

Éléments cliniques : *Une femme de 53 ans, témoin de Jéhovah, souffrait d'anémie sévère post-arthroplastie totale de la hanche. Elle refusait toute transfusion sanguine. De la tachycardie, de l'hypotension, des anomalies électrocardiographiques et des changements de l'état mental sont apparus avec une concentration d'Hb minimale de 3,2 $g \cdot dL^{-1}$. Le raffimère d'Hb est une solution d'Hb humaine purifiée, polymérisée, développée comme substitut de l'Hb des globules rouges. Avec l'approbation, pour usage humanitaire, de la patiente, de la Food and Drug Administration et du Comité d'examen de l'institution, 2 L de raffimère d'Hb (Hemolink™, Hemosol, Inc., Toronto, ON, Canada) ont été administrés avec du sulfate ferreux et de l'érythropoïétine humaine. L'Hb s'est élevée à 5,5 $g \cdot dL^{-1}$ avec la résolution des symptômes. Pour assurer la récupération de la masse sanguine tout en maintenant le niveau d'Hb > 4,5 $g \cdot dL^{-1}$, des doses additionnelles de 1000 mL de raffimère d'Hb ont été donnés aux jours trois, cinq et sept postopératoires (dose totale = 500 g d'Hb). Aucune complication sérieuse liée au raffimère d'Hb ne s'est développée. Au jour postopératoire 14, le niveau d'Hb est monté à 6,5 $g \cdot dL^{-1}$ avec un hématocrite de 23 %. La patiente a reçu son congé.*

Conclusion : *Le raffimère d'Hb, utilisé pour favoriser la récupération de la masse sanguine peut avoir évité des complications cliniques. Cette solution d'Hb purifiée dépourvue d'autres composantes cellulaires peut constituer un traitement acceptable aux patients qui, par conviction religieuse, refusent une transfusion allogénique.*

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LARGE-VOLUME blood loss is a known complication of hip arthroplasty. Any adult patient with the capacity to decide has the right to exercise autonomy in refusing transfusion once fully informed of risks and benefits of the proposed treatment.[1] Clinical data derived from the Jehovah's Witness population suggest that adverse clinical sequelae develop once the hemoglobin (Hb) concentration falls below 4.5 $g \cdot dL^{-1}$ and that mortality exceeds 95% when the Hb falls below 3.0 $g \cdot dL^{-1}$.[2] Although not yet approved for human use in North America, Hb-based oxygen carriers have been utilized on a compassionate-release basis in order to provide temporary support for tissue oxygen delivery in states of severe anemia.[3–5] We report a case of acute, life-threatening anemia successfully managed with the oxygen therapeutic agent, Hb raffimer (Hemolink™, Hemosol, Inc., Toronto, ON, Canada).

Case report

A 53-yr-old post-menopausal female with a history of osteoarthritis, unipolar depression and hypertension controlled with hydrochlorothiazide underwent right total hip replacement in 2002 for avascular necrosis. As a practicing Jehovah's Witness, she had explicitly refused transfusion of cellular blood products for the procedure. Preoperative Hb concentration was 12.6 $g \cdot dL^{-1}$ while receiving iron supplementation. Preoperative coagulation tests were within normal limits. Excessive perioperative bleeding was not anticipated by the surgical team. The procedure was complicated by intraoperative blood loss of 2100 mL, followed by an dditional blood loss of 600 mL in the first 20 hr. Supplemental oxygen was maintained at 6 $L \cdot min^{-1}$ per nasal cannula and epoetin alfa (Epogen®, Amgen Inc., Thousand Oaks, CA, USA) therapy was instituted. Epoetin alfa was administered subcutaneously at a dose of 10,000 units (100 $U \cdot kg^{-1}$) three times per week in combination with oral iron supplementation with ferrous sulfate at a dose of 324 mg three times per day. Although clinical evidence of postoperative hemorrhage resolved, the Hb level progressively declined to a nadir of 3.2 $g \cdot dL^{-1}$ on postoperative day one. Due to severity of anemia, deep venous thrombosis prophylaxis consisted of sequential compression devices only without postoperative anticoagulation. The patient was managed initially in a surgical intensive care unit setting but did not require postoperative mechanical ventilation.

Due to profound anemia, the patient developed sinus tachycardia (150 $beats \cdot min^{-1}$), tachypnea, somnolence and hypotension (65/36 mmHg). Hypotension was treated with 3.5 L of crystalloid solution and 1.25 L of 6% hetastarch solution (Hextend®, Abbott Laboratories, North Chicago, IL, USA) over the first 24 hr postoperatively as well as a phenylephrine infusion titrated from 0.5 to 1.5 $\mu g \cdot kg^{-1} \cdot min^{-1}$ to maintain mean arterial pressure above 60 mmHg. An electrocardiogram revealed sinus tachycardia with ST-segment depression in lateral leads suggestive of myocardial ischemia. The patient's mental status deteriorated, with worsening confusion and somnolence. In accordance with patient wishes, no transfusion of allogeneic human blood was administered. In view of potentially life-threatening cardiac and central nervous system ischemia, emergent compassionate release of the oxygen therapeutic agent, Hb raffimer, was requested and approved by the United States Food and Drug Administration. Informed consent for administration of Hb raffimer was obtained from the next of kin, in consultation with the patient's clergy.

An initial dose of 2000 mL of Hb raffimer (200 g Hb) was administered, with subsequent reversal of signs and symptoms of inadequate tissue oxygen delivery. The total Hb level increased from 3.2 $g \cdot dL^{-1}$ to 5.1 $g \cdot dL^{-1}$ after the initial dose. The patient's tachycardia, ST-segment depression and mental status changes resolved. Blood pressure rose to 150/55 mmHg and phenylephrine therapy was discontinued. Side effects noted during therapy included scleral icterus and a transient elevation of systolic blood pressure following administration of the initial 200 g dose of Hb raffimer. Baseline blood pressure on admission was 143/92 mmHg. Maximum blood pressure was 180/54 mmHg, measured two hours following completion of the first Hb raffimer infusion. The systolic pressure elevation persisted for six hours and resolved with pain control and diuresis of 2700 mL of urine in response to a 20-mg dose of furosemide. Diastolic blood pressure remained below the patient's baseline and the systolic elevation did not recur with subsequent doses. Three supplemental doses of 1000 mL Hb raffimer were required over the next five days (total dose = 500 g Hb) to maintain the patient's total Hb above a target concentration of 4.5 $g \cdot dL^{-1}$ (Figure). The patient denied nausea or abdominal pain related to treatment. Reticulocyte count increased gradually from $30 \times 10^9 \cdot L^{-1}$ on postoperative day two, to $411 \times 10^9 \cdot L^{-1}$ by postoperative day 14, at which time epoetin alfa therapy was discontinued.

On postoperative day five, symptomatic arterial desaturation with disorientation and agitation developed when the patient removed her nasal cannula, which was carrying oxygen at a rate of 4 $L \cdot min^{-1}$. Arterial saturation measured by pulse oximetry fell to 68% while on room air with recovery to 93% once oxy-

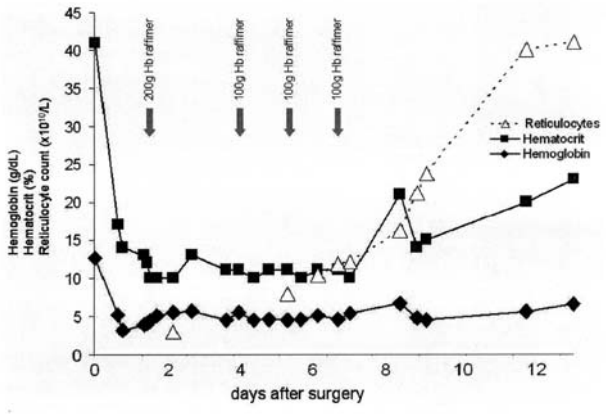

FIGURE Trend of hemoglobin level hematocrit, and reticulocyte count during hospitalization with relation to administration of hemoglobin raffimer. Reticulocyte count is graphed in units of cells × 10^{10}·L^{-1}. Multiplication of graph values by a factor of ten will result in standard units of cells x 10^{9}·L^{-1}. Hb = hemoglobin

gen therapy was replaced at 6 L·min^{-1} via nasal cannula. Symptoms resolved upon resumption of oxygen therapy. Serial arterial blood gases revealed an elevation in methemoglobin (MetHb) concentration during the period of Hb raffimer administration peaking at 7.9% on the third postoperative day with a maximum carboxyhemoglobin level of 3.8%. Serum bilirubin concentration peaked at 8.2 mg·dL^{-1} on the seventh postoperative day and returned to normal by the 12th postoperative day. Transient, asymptomatic elevation of amylase and lipase was noted following Hb raffimer administration with peak levels never exceeding twice the upper limit of normal. No elevation of aspartate aminotransferase, alanine aminotransferase, alkaline phosphatase, blood urea nitrogen, or creatinine was noted with serial measurements. On postoperative day 14, the patient's Hb level was 6.5 g·dL^{-1} with a hematocrit of 23%. She was discharged to an orthopedic rehabilitation facility with a continued regimen of oral iron supplementation. Follow-up evaluation three months after discharge revealed normal neurologic and cardiac function, a stable hip prosthesis, and a well-healed incision. Hemoglobin level measured at a clinic visit eight months after hospital discharge was 13.0 g·dL^{-1}.

Discussion

Hb raffimer is a purified, cross-linked, human Hb solution developed for use as an alternative to red blood cell (RBC) transfusion. It is produced from outdated units of packed RBCs that have been previously screened and approved for clinical use. Hb is extracted, heat-treated, ultra-filtered, and then cross-linked between beta-subunits with o-raffinose. This yields a solution of 10 g·dL^{-1} HbAo, (human adult Hb tetramer containing two alpha chains and two beta chains) with > 99% purity and a molecular size of 64 kDa or larger (> 95%). It is stored in the deoxygenated state. The high molecular weight of the cross-linked raffimer results in minimal renal toxicity, prolonged intravascular persistence (half-life of 15–18 hr), and a more favourable side-effect profile compared to previously-tested Hb-based oxygen carriers.[6,7]

Preliminary data from phase 2 trials in the United States suggested that doses up to 750 mL of Hb raffimer as an adjunct to acute normovolemic hemodilution during coronary artery bypass grafting surgery were well tolerated.[8] Prior to March 2003, further phase 2 studies in the United States and phase 3 clinical trials in Canada and the United Kingdom using larger doses of Hb raffimer were ongoing in cardiac and orthopedic surgical patients. Due to an increased incidence of myocardial infarction noted in Hb raffimer-treated cardiac surgical patients, enrollment in all clinical trials was suspended in March 2003 and has not been restarted. The mechanism and clinical significance of the myocardial infarctions observed in the cardiac surgical patients is under investigation and it is currently unknown whether this effect would be relevant to patient populations not undergoing myocardial revascularization surgery.

In this 53-yr-old female without known coronary artery disease, signs and symptoms of severe anemia including electrocardiographic evidence of myocardial ischemia resolved following the administration of Hb raffimer. Measurements of the MB isoenzyme of creatine kinase (CK-MB) on postoperative day one as well as from postoperative day five through hospital discharge remained within normal limits (≤ 2 ng·mL^{-1}, normal range 0–9 ng·mL^{-1}). No measurements of CK-MB were performed from postoperative day two through four in an effort to minimize phlebotomy in extreme anemia. While limited, these data show no evidence of myocardial infarction in this patient.

A known side effect of Hb raffimer administration is the binding of nitric oxide. Arterial vasoconstriction and resultant hypertension can occur, as in this patient. The wide pulse pressure likely reflects the role of nitric oxide in regulating arteriolar vasodilation. In this scenario, the increased blood pressure was a favourable side effect in a previously hypotensive patient and did not result in clinically-apparent adverse events such as myocardial ischemia. The

TABLE Oxygen extraction of Hb raffimer,* fresh RBCs† and aged RBCs‡ with varying PaO_2§

	PaO_2 = 100 mmHg			PaO_2 = 300 mmHg			PaO_2 = 600 mmHg		
	Hb rafffimer	*Fresh RBCs*	*Aged RBCs*	*Hb rafffimer*	*Fresh RBCs*	*Aged RBCs*	*Hb rafffimer*	*Fresh RBCs*	*Aged RBCs*
SaO_2 (%)	61	97	99	82	100	100	90	100	100
SvO_2 (%)	41	74	93	41	74	93	41	74	93
% O_2 Extraction	20	23	6	41	26	7	49	26	7

Hb = hemoglogin; RBCs = red blood cells. Comparison of oxygen delivery at various levels of inspired oxygen for fresh red blood cells, hemoglobin raffimer and red blood cells stored for 40 days. (O_2 = oxygen, FiO_2 = fraction of inspired oxygen, PaO_2 = partial pressure of oxygen in arterial blood, PvO_2 = partial pressure of oxygen in venous blood, mmHg = millimeters of mercury. *D. Mazer, et al.[15] †Derived from C. Bauer.[16] ‡Derived from R. McConn and J.B. Derrick.[17] §Assuming constant PvO_2 of 40 mmHg.

patient's diastolic blood pressure remained lower than her admission diastolic pressure of 92 mmHg.

Based on data that demonstrated an increase in adverse events at Hb levels lower than 4.5 $g \cdot dL^{-1}$ a target concentration of a Hb level of 4.5 $g \cdot dL^{-1}$ was chosen.[2] Hb levels of 5 $g \cdot dL^{-1}$ have been shown to be well tolerated in healthy patients.[9–11] The dosing of epoetin alpha was based on data on treating postoperative anemia.[12] Iron was supplemented orally, since in addition to the inconvenience and cost, parenteral iron therapy is associated with potential serious risks compared to oral iron therapy. No superior benefit is expected from parenteral iron administration unless oral supplementation therapy fails. Our patient responded well with an increase in reticulocytes.

Transient recurrence of mental status changes accompanied by arterial Hb desaturation on postoperative day five were temporally related to the patient's removal of her nasal cannula oxygen. Symptomatic hypoxemia while breathing room air was likely due to alteration of the patient's oxygen dissociation curve as well as the presence of carboxyhemoglobin and MetHb in the plasma. In a recent canine study of MetHb persistence, measured MetHb levels of Hb raffimer stored in the deoxygenated form were 8.6%.[13] Similar to endogenous plasma Hb, Hb raffimer is catabolized via the heme oxygenase pathway, resulting in elevated levels of unconjugated bilirubin and carboxyhemoglobin that is dose-dependent.[7] With a large-volume administration of Hb raffimer in this case, MetHb levels increased to a peak of 7.9% with a peak carboxyhemoglobin level of 3.8%, limiting maximum oxyhemoglobin saturation to 88%.

The second mechanism for the development of symptomatic hypoxemia while breathing room air is likely related to the binding characteristics of the Hb-based oxygen carrier. Hb raffimer binds oxygen in a relatively linear fashion as a function of PO_2 (P_{50} = 52 mmHg, Hill coefficient = 0.97).[14] Due to replacement of up to 40% of the circulating RBC Hb with Hb raffimer, this patient's overall Hb dissociation curve would display decreased oxygen affinity compared with fresh RBC Hb. Employing supplemental oxygen to maintain a PaO_2 > 150 mmHg increased arterial saturation of, and oxygen delivery from, Hb raffimer. Higher plasma oxygen tensions would not be expected to offer additional benefit once near-maximal saturation of Hb raffimer is achieved. The contribution of additional dissolved oxygen in the plasma becomes clinically insignificant once additional Hb in the form of Hb raffimer is present. While the dissociation curve of Hb raffimer requires an elevated alveolar oxygen concentration for maximal oxygen binding, off-loading of oxygen at the tissue level[14] while breathing room air[15] is comparable to that of fresh RBC Hb,[16] and would be substantially greater than that from stored RBCs[17] depleted of 2,3-diphosphoglycerate. The Table compares the previously published oxygen delivery characteristics of Hb raffimer with those of fresh red blood cells and red blood cells stored for 40 days. At elevated PO_2, Hb raffimer would likely demonstrate increased oxygen delivery compared with fresh RBCs and markedly increased oxygen delivery compared with stored RBCs, suggesting that Hb-based oxygen carriers may be effective resuscitation fluids for acute blood loss (Table).

Conclusions

Given the nadir Hb concentration of 3.2 $g \cdot dL^{-1}$ in this patient, the use of Hb raffimer as a bridge to recovery of RBC mass likely prevented an adverse clinical outcome. Because Hb raffimer is a purified Hb solution that is devoid of other cellular components, it may be acceptable to patients who, due to religious conviction, refuse allogeneic RBC transfusion. While the side-effect profile may preclude use for patients at risk

for myocardial ischemia, Hb-based oxygen carriers in combination with supplemental oxygen may provide an acceptable alternative to RBC transfusion in a patient population without coronary artery disease.

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