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Use of Perftoran Emulsion to Decrease
Allogeneic Blood Transfusion in Cardiac Surgery:
Clinical Trial

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Abstract: Perfluorocarbon emulsions (PFC) have the capacity of transporting oxygen through the bloodstream and may be safe and effective alternatives to allogeneic blood transfusions during surgical procedures. Perftoran was the PFC used in a randomized clinical trial conducted at Hospital de Especialidades Centro Medico La Raza, Mexico City. The clinical trial took a sample group, n = 30, of patients that were scheduled for elective cardiac valvuloplasty surgery in combination with preoperative acute normovolemic hemodilution and an inspiratory oxygen fraction (FiO2) of 1.0. The participants were randomly divided into a Control group (n = 15) and a Perftoran (PFC) group (n = 15). The PFC group had significantly higher intraoperative PaO2 levels and needed less allogeneic red blood cell packs than the Control group. There were no complications or deaths in either group. These results suggest that Perftoran is safe, efficacious, and reduces the need for allogeneic blood and blood derivatives in patients undergoing cardiac surgery.

Keywords: Acute normovolemic hemodilution; Blood substitute; Hyperoxia; Perfluorocarbons; Perftoran

More than 800 cardiac surgeries are performed annually in the Cardiothoracic Surgery Department at Hospital de Especialidades Centro Medico La Raza in Mexico City. These surgeries are major consumers of blood bank resources. Although judicious blood and blood-byproduct

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administration is practiced to maintain adequate tissue oxygenation and homeostasis in cardiac surgery, transfusion-related complications remain a major problem. Cost [1], allogeneic blood transfusion reactions [2], alloimmunization [3], transmission of infectious agents [4], transmission of proinflammatory mediators [5–7], and immunosuppression [8] are constant concerns in elective surgeries where large volumes of blood are routinely required.

Patients undergoing elective cardiac surgery with cardiopulmonary bypass (CPB) generally require a significant amount of blood and blood derivatives. Acute normovolemic hemodilution (ANH) is a blood-saving technique used preoperatively to reduce the need for allogeneic blood transfusion. The standard ANH technique is performed by harvesting blood from patients immediately before surgery and simultaneously replacing the blood volume removed with crystalloid or colloid solutions to maintain normovolemia until hemoglobin (Hb) concentration is lowered to a target level [9–11]. The harvested autologous blood is then reinfused during the intraoperative period or at the end of surgery depending on blood-loss requirements. In order for ANH to effectively reduce allogeneic blood transfusion requirements during surgery, a significant amount of autologous blood must be removed [12]. An effective volume replacement agent with oxygen carrying capacities would enhance the safety of a large volume blood harvest before initiating cardiac surgery to significantly reduce the need for allogeneic transfusion. For this purpose, and a host of other conditions, the search for transfusion alternatives continues in the area of “artificial blood” or, more aptly, “artificial oxygen carriers.” Artificial oxygen carriers can be divided into two classes: 1) modified hemoglobin solutions, and 2) Perftorocarbon emulsions (PFC) [13].

PFCs are eight to ten carbon molecules whose hydrogen atoms have been replaced by fluorine. This results in chemically inert liquids that must be emulsified because they are immiscible in water and lipids. Beneficial qualities of PFC for clinical use are that they can be sterilized, they are universally accepted by all blood types, and large volumes can be cost-effectively manufactured [13,14].

An important characteristic of PFCs is that gases, such as O₂ and CO₂, are highly soluble in them. For this reason, PFC can carry a substantial amount of O₂. At room temperature, O₂ is nearly 20 times more soluble in PFC than in water [14]. The solubility of O₂ is 2.5% in water, 2.5% in plasma, 20% in whole blood, and 40% or more in PFC. The oxygen carrying capacity of PFC is directly related to the partial pressure of O₂ [14].

The efficiency of PFC in providing additional oxygen delivery and enhancing oxygen distribution capacity has been observed in experimental models of hemorrhagic shock [15], ischemia-reperfusion [16], hemodilution [17], during surgical blood loss [18], and CPB [1].

Perftoran (Perflec) is a PFC-based artificial oxygen carrier that was developed by the Russian National Academy of Science between 1979 and 1995. The Russian Federation Ministry of Health approved Perftoran for clinical application. The manufacture, distribution, and use of Perftoran in Russia began in 1997 [13].

Perftoran is a 20% wt/vol PFC emulsion based on a 2:1 ratio of perfluorodecalin and perfluoromethylcyclohexyloxepin. It is emulsified with 4% wt/vol propanol-268. The perfluorochemical emulsion particles have an average size of 0.87 μm, with a range of 0.03–0.15 μm [13,14].

It has been observed that Perftoran enhances oxygen transportation from the lungs into tissue by its oxygen carrying capacity and rheological parameters, which include low viscosity, small particle size, large total surface area, modification of the red blood cell membrane, and the high mobility of PFC particles in the bloodstream during interaction with red blood cells (RBC) [14].

An optimal use of PFC emulsions is to reduce allogeneic blood transfusion requirements when used in conjunction with ANH [1,17,18]. Patients will undergo ANH to lower Hb concentrations preoperatively with an increase in blood-saving efficiency [19]; however, with PFC used simultaneously, improved safety during more profound anemia may result from the capacity of the PFC to carry O₂. This may allow a greater amount of a patient’s own blood to be drawn and set aside for later use, or delay the return of the autologous blood, thus minimizing the necessity of allogeneic transfusion. Similarly, use of PFC during surgery, where blood loss is greater than anticipated, may postpone or negate the need for allogeneic transfusion [1,17,18,20].

The objective of this clinical trial was to evaluate the safety and efficacy of Perftoran in reducing or avoiding allogeneic blood transfusion used in conjunction with preoperative ANH on patients undergoing cardiac surgery with CPB compared with standard of care. The primary efficacy endpoints measured were: 1) Perftoran’s oxygen-carrying capacity; 2) the mean number of allogeneic blood (and blood derivatives) units transfused; and 3) the percentage of patients that avoided allogeneic blood transfusion.

MATERIALS AND METHODS

Study Population

This was a randomized, controlled, parallel-group clinical trial conducted from August 2004 through March 2005 in the Cardiothoracic Surgery Unit of the Moscow City Hospital No. 54. The patients were randomized to receive either preoperative ANH or PFC in conjunction with CPB.

*Perflec is the commercial name for Perftoran approved by the Secretary of Health in Mexico.
Department of the Hospital de Especialidades Centro Medico La Raza, the flagship hospital of Instituto Mexicano del Seguro Social (IMSS). The protocol received approval from the IMSS' local and federal ethics committees. Thirty-one patients scheduled to undergo elective cardiac valvuloplasty surgery with CPB signed written informed consent forms. The participants were randomly divided into two groups: 1) control group with ANH alone; 2) PFC group with ANH in combination with the administration of Perforan. Other than the infusion of Perforan, the care, interventions, and surgery were similar for all patients. One participant was excluded from the study due to an allergic reaction to Perforan. This left a sample size of 30.

Inclusion criteria were: patients of either sex undergoing elective valvuloplasty surgery between the ages of 18 years and 75 years, with a preoperative Hb concentration between 12 g/dL and 18 g/dL. Exclusion criteria were pregnancy or lactation, emergency surgery, history of hypersensitivity to constituents of Perforan, recent heart infarct (<6 months), unstable angina pectoris, congestive heart failure greater than New York Heart Association class II, symptomatic cerebrovascular disease, severe renal disease, psychiatric illness, severe chronic obstructive pulmonary disease, significant hepatic disease (aspartate aminotransferase or alanine aminotransferase >2 upper limit of normal), preoperative blood transfusions, AIDS, obesity (BMI > 35), platelet count <80,000/µL, coagulation disorder, immunosuppression, local or systemic infection, alcohol or drug abuse within the past year. Elimination criteria were: patients who chose to withdraw from the study, fever, infection, Hb <12 g/dL just before surgery, a significant decrease in organic function in comparison with the state in the initial exam, considerable violation to the protocol, significant adverse effects during the administration of Perforan, development of any exclusion criteria during the treatment, and allergic reaction to Perforan during the preoperative biological test.

Trial Size

The 15 patients in the Control group and the 15 patients in the PFC group were selected from a population admitted to Hospital La Raza with a diagnosis of rheumatic or degenerative valvulopathy who were programmed for elective valvuloplasty surgery. The participants complied with the inclusion criteria and were willing to undergo the experimental protocol. As part of the informed consent process, the procedures, purposes, potential benefits, risks, and potential adverse actions were explained. Each participant signed an individual informed consent form.

Measurements

Safety parameters included vital signs, laboratory profiles (hematology including red cell, white cell and platelet counts, hemoglobin and hematocrit, coagulation parameters and blood chemistry), and the collection of adverse event data.

Efficacy parameters included characteristics of ANH (autologous blood volume harvested and volume of plasma expanders transfused during the hemodilution), characteristics of the intraoperative period (blood loss during surgery, duration of anesthesia, duration of CPB, duration of the aortic cross-clamp, and duration of surgery), transfusion parameters (subjects who avoid transfusion, mean units of RBC and other blood components transfused per patient), and blood-gas parameters.

Study Protocol

Experimental Protocol

The clinical trial took a sample group, n = 30, of patients that were undergoing elective cardiac valvuloplasty surgery in combination with preoperative ANH and an inspiratory oxygen fraction (FIO₂) of 1.0. The participants were randomly divided into a Control group (n = 15) and a Perforan group (n = 15). The experimental protocol was divided into three periods: 1) preoperative ANH; 2) intraoperative; 3) postoperative.

Perforan Conservation and Handling

Perforan, a trademark from Perfloran Corp, Puschino, Moscu Region, Russia, is a white emulsion with a slightly blue tint. Its presentations are glass vials containing 100 mL, 200 mL, and 400 mL of Perforan emulsion. The emulsion was provided by Laboratorios Kem, SA de CV, Tijuana, Baja California, Mexico, who sponsored the study.

Perforan was stored frozen at −18°C. Twelve hours before Perforan was used it was removed from the freezer and kept refrigerated at +4°C. Later, the emulsion was left to thaw at room temperature. After thawing, the emulsion was carefully stirred until the complete homogeneity of the emulsion was obtained. As recommended by the manufacturer, the emulsion was inspected for color, stratification, or sedimentation formation to verify consistency and quality before use. Subsequently, 100% oxygen was introduced to the Perforan emulsion using a 15 cm sterile stainless steel needle and a 0.02 µm micro pore filter bubbling for 15 minutes. Once the Perforan oxygenation procedure was finished, a Perforan
dose calculation based on 5 mL/Kg of body weight was made to determine the volume of Perfloran to be infused. This dose was divided into two equal portions for administration at two specific times during the intraoperative period.

Before the surgery, a biological test was done on patients to detect hypersensitivity to constituents of Perfloran, which would exclude a patient from the study. A test dose of 5-20 drops of Perfloran was infused and the patient was observed for 5-10 minutes. If there were no allergic reactions or other symptoms of hypersensitivity, the infusion was continued at a rate of 60 drops/minute. In cases where an allergic reaction occurred, the infusion of Perfloran was stopped and the patient was excluded from the study.

Preoperative ANH

A peripheral venous cannula was inserted and anesthesia was induced. After tracheal intubations, patients were ventilated at an FIO2 of 1.0. Prior to surgical incision, patients underwent ANH to a hematocrit (Hct) value of 30% or Hb concentration of 9 g/dL, whichever came first. As blood was collected during ANH, crystalloid and colloid solutions were used to replace the volume lost. The target Hct was used to calculate the final volume of autologous blood to be harvested according to the formula:

\[ V = \frac{EBV \times (Hi - Hf)}{Hi} \]

where \( V \) = volume of blood to be harvested; \( EBV \) = patient's estimated blood volume = body weight (Kg) \( \times 65 \text{ mL} \) per kg; \( Hi \) = patient's initial Hct; \( Hf \) = patient's target Hct after hemodilution. The ANH period had an average duration of 20 minutes.

Measurements of vital signs, red blood cell count, white blood cell count, platelet count, coagulation, and blood chemistry were taken before and after surgery. After ANH, baseline measurements of vital signs, including Hb, Hct, hemodynamic and blood-gas, were taken.

Intraoperative Period

During the intraoperative period, measurements of Hct, hemodynamic and blood-gas variables were taken every 15-20 minutes. The following transfusion triggers were established: Hct levels <21% or Hb <7 g/dL; tachycardia (heart rate >125% of post-ANH rate or 110 bpm); and hypotension (mean arterial pressure <75% of post-ANH level or ≤60 mm Hg). In the event that any trigger occurred, a RBC package was transfused into the patient.

The intraoperative bleeding was evaluated by measuring the blood volume collected in the suction reservoir and by calculating the weight change in gauzes and surgical towels. The measurement of blood loss was made to determine the volume to be replaced with crystalloid and colloid solutions.

Patients in the PFC group received a first infusion of Perfloran at a dose of 2.5 mL/kg body weight immediately after ANH. A second infusion of Perfloran at the same dose was given when the aortic cross-clamp was removed. Patients in the Control group received a placebo infusion of crystalloid solution at a dose of 2.5 mL/kg body weight after ANH and another after aortic cross-clamp removal. After initiation of wound closure, the Hb concentration was adjusted to ≥9 g/dL by gradual transfusion of all remaining autologous blood. Then a determination was made of how much allogeneic blood was required.

Postoperative Period

During the first 24 hours after surgery, patients were ventilated at FIO2 of 0.21. The decision to transfuse packed RBC was guided by clinical judgment taking into account, not only the Hb concentration, but also the physical status of the patient (age, estimated blood volume, cardiovascular and respiratory functions), and the extent of postoperative bleeding. When transfused patients had a Hb between 7 g/dL and 9 g/dL, blood should not be transfused to patients with a Hb concentration >9 g/dL, while blood should be transfused to patients with Hb concentration ≤7 g/dL. Oral iron therapy was administered to patients whose Hb was ≤9 g/dL.

Efficacy Evaluation

Primary efficacy endpoints for this protocol were: 1) the number of allogeneic RBC packs transfused during and after surgery; 2) the percentage of patients avoiding transfusion; 3) the change in percentage of blood-gas parameters before, during, and after the surgical procedure. The blood-gas parameters included were PaO2, PaCO2, pH, bicarbonate (HCO3), lactate, and base excess.

Secondary parameters included: 1) patient demographics and baseline characteristics; 2) data before and after ANH; 3) data about clinical parameters such as anesthesia, surgery. CPB and aortic cross-clamp duration; 4) volume of blood loss summarized for each group.
Safety Evaluation

Safety parameters included: 1) preoperative and postoperative physical examination with vital signs; 2) laboratory profiles including red cell count, white cell count, platelet count, Hb, Hct, coagulation parameters, and blood chemistry; 3) adverse effects.

Statistical Analysis

Efficacy and safety parameters were presented as the mean ± standard deviation. Differences between groups were assessed with Student's t-test. All statistical tests were conducted at a 0.05 level of significance and were two tailed.

RESULTS

General Characteristics

The patients, n = 30, selected to participate in the study had a diagnosis of rheumatic or degenerative valvulopathy. They were undergoing elective cardiac valvuloplasty surgery in combination with preoperative ANH and an FIO₂ of 1.0. The participants were randomly divided into a Control group (n = 15) and a PFC group (n = 15). Demographic characteristics were similar for participants in both groups (Table 1).

During the ANH period, patients in the PFC group had 1,189 mL ± 365 mL of blood harvested and the Control group had 1,280 mL ± 402 mL harvested. The PFC group received a replacement volume of colloid solution of 1,252 mL ± 501 mL and the Control group received 1,430 mL ± 416 mL. The PFC group received 1,371 mL ± 319 mL of crystalloid solution and the Control group received 1,179 mL ± 417 mL. No significant differences were observed between the groups (Table 2).

Post ANH arterial blood pressure, heart rate, respiratory frequency, and venous blood pressure were similar in both groups (Table 2). The mean post-ANH Hb concentration achieved was 9.38 g/dL ± 0.5 g/dL. All patients in the PFC group received two infusions of Perflutan at a dose of 2.5 mL/kg body weight each, resulting in a total dose of 5 mL/kg (equivalent to 1 g/kg of PFC). The patients in the Control group received two infusions of crystalloid solution, at the same dose of 2.5 mL/Kg body weight for each infusion.

The intraoperative bleeding for the PFC group was 565 mL ± 302 mL and 440 mL ± 205 mL for the Control group (Table 3). No significant differences were observed between the groups.

Efficacy

Transfusion Requirements

The autologous blood volume harvested during ANH, the estimated intraoperative blood loss and the replacement volume of colloid and crystalloid solutions, were similar in both groups, yet differences in transfusion requirements of alloegenic blood and blood components were

<table>
<thead>
<tr>
<th>Table 2. Post-acute normovolemic hemodilution characteristics</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clinical parameter</td>
</tr>
<tr>
<td>Autologous blood harvested during ANH (mL)</td>
</tr>
<tr>
<td>Volume of colloidal solution used for replacement during ANH (mL)</td>
</tr>
<tr>
<td>Volume of crystalloid solution used for replacement during ANH (mL)</td>
</tr>
<tr>
<td>Systolic Pressure (mm Hg)</td>
</tr>
<tr>
<td>Diastolic Pressure (mm Hg)</td>
</tr>
<tr>
<td>MAP (mmHg)</td>
</tr>
<tr>
<td>CF (beats/min)</td>
</tr>
<tr>
<td>RF (respirations/min)</td>
</tr>
<tr>
<td>CVP (mm Hg)</td>
</tr>
</tbody>
</table>

ANH = Acute normovolemic hemodilution; MAP = Mean arterial pressure; CF = Cardiac frequency; RF = Respiratory frequency; CVP = Central venous pressure. Values are mean ± SD.
Table 3. Allogeneic blood and blood components transfusion data

<table>
<thead>
<tr>
<th></th>
<th>PFC group (n = 15)</th>
<th>Control group (n = 15)</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Estimated intraoperative bleeding (mL ± SD)</td>
<td>565 ± 302</td>
<td>440 ± 205</td>
<td>0.18</td>
</tr>
<tr>
<td>Patients transfused with pRBC (%)</td>
<td>37.5</td>
<td>60</td>
<td></td>
</tr>
<tr>
<td>Mean no. of pRBC units transfused per patient (range)</td>
<td>0.8 (0–4)</td>
<td>1.4 (0–10)</td>
<td>0.16</td>
</tr>
<tr>
<td>Patients transfused with CP (%)</td>
<td>6.3</td>
<td>67</td>
<td></td>
</tr>
<tr>
<td>Mean no. of CP units transfused per patient (range)</td>
<td>0.4 (0–6)</td>
<td>5.8 (0–10)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Patients transfused with Cryo (%)</td>
<td>6.3</td>
<td>47</td>
<td></td>
</tr>
<tr>
<td>Mean no. of Cryo units transfused per patient (range)</td>
<td>0.6 (0–10)</td>
<td>5.6 (0–10)</td>
<td>&lt;0.02</td>
</tr>
<tr>
<td>Patients transfused with FFP (%)</td>
<td>12.5</td>
<td>87</td>
<td></td>
</tr>
<tr>
<td>Mean no. of FFP units transfused per patient (range)</td>
<td>0.3 (0–3)</td>
<td>1.6 (0–5)</td>
<td>&lt;0.002</td>
</tr>
</tbody>
</table>

pRBC = Packed red blood cells (300 mL per unit); CP = Concentrated platelets (70 mL per unit); Cryo = Cryoprecipitate (70 mL per unit); FFP = Fresh frozen plasma (200 mL per unit).

Observe the difference observed between the groups (Table 3). With respect to reduction in the number of allogeneic packed RBC units transfused, patients in the PFC group required fewer transfusions than patients in the Control group. The PFC group received 0.8 units ± 1.15 units and the Control group received 1.4 units ± 2.4 units. The difference is not statistically significant but it is noted that the reduction of required RBC units was 43% for the PFC group (Table 3). The total units of allogeneic packed RBC for the PFC group were 12 compared with 22 for the total of the Control group. A total of 62.5% of PFC patients avoided transfusions compared with 40% of the patients in the Control group (Table 3).

A number of patients in each group received other allogeneic blood components during surgery, including cryoprecipitate: PFC group 6.3%, Control group 4.7%; fresh frozen plasma: PFC group 12.5%, Control group 8.7%; and platelets: PFC group 6.3%, Control group 6.7% (Table 3). The differences between the groups in the mean number of cryoprecipitate, fresh frozen plasma, and platelets units transfused per patient were statistically significant (Table 3).

The number of patients avoiding all types of transfusions of allogeneic blood and blood components was significantly higher in the PFC group than the Control group (10/15 versus 0/15, respectively) (p < 0.05).

Transoperative Hemoglobin Concentrations

Hemoglobin concentrations before surgery and after ANH were similar among groups. However, during the transoperative period Hb concentrations were lower in the PFC group than in the Control group (Fig. 1). These differences were statistically significant. The Hb concentration for the PFC group was 7.1 g/dL ± 1.4 g/dL at 110 minutes and 6.83 g/dL ± 1.1 g/dL at 140 minutes. For the Control group it was 8.8 g/dL ± 1.3 g/dL at 110 minutes and 8.8 g/dL ± 1.5 g/dL at 140 minutes (Fig. 1). At the end of surgery, and during the postoperative period, no significant differences in Hb concentrations were observed between groups.

Blood-Gas Parameters

Preoperative blood-gas parameters (baseline values) were similar between groups (Figs. 2–7). PFC group showed PaO₂ values higher than the
Control group during the majority of the intraoperative period (Fig. 2). The differences were statistically significant at 50 minutes, which was 20 minutes to 30 minutes after the first Perfloran infusion. The same was true at 110 minutes, 20 minutes to 30 minutes after the second Perfloran infusion. It remained the same until minute 140 (p < 0.05) (Fig. 2). Levels of PaCO2 were similar in both groups during the entire experimental protocol, including ANH and intraoperative period (Fig. 3). At postoperative, the Control group showed PaCO2 levels significantly higher than the PFC group (p < 0.05) (Fig. 3). Blood pH values were slightly higher in the PFC group than the Control group. These differences were only statistically significant at 100 minutes (p < 0.05) (Fig. 4). Lactate blood levels in the PFC group were higher than those in the Control group during most of the intraoperative period, but these differences were not statistically significant (Fig. 5). In contrast, a constant increment in lactate blood levels was observed in both groups. This trend persisted during the entire experimental protocol including ANH and intraoperative period (Fig. 5), to the extent that from 50 minutes until the procedure was done, lactate blood levels increased to levels that were significantly higher than the preoperative baseline values in both groups (p < 0.005). Bicarbonate blood levels (HCO3) behaved similarly in both groups until 200 minutes when HCO3 blood levels in the Control group rose significantly higher than the PFC group (p < 0.05) (Fig. 6). It was observed that the base excess in the PFC group tended to rise slightly and in the Control group it tended to decrease. The base excess between both groups remained without statistically significant differences during most of the experimental protocol, with the exception of the statistically significant difference at 200 minutes (p < 0.05) (Fig. 7).

Intraoperative Parameters

There were no significant differences in anesthesia, surgery, CPB and aortic cross-clamp duration between the groups (Table 4).
Safety

Hematology

The preoperative levels of platelets were significantly higher in the PFC group than the Control group (p < 0.05) (Table 5). All other preoperative values of hematologic parameters were similar among the groups. Postoperative levels of Hb, Hct, red blood cells, mononuclear cells, and platelets were significantly lower than preoperative levels in both groups (p < 0.05) (Table 5). In contrast, the number of polymorphonuclear (PMN) cells showed a significant increase at the end of the surgery when compared with preoperative levels in both groups (p < 0.0001) (Table 5).

No significant differences were observed between the groups in the coagulation parameters before and after surgery (Table 6). Blood

\[
\text{\textbf{Figure 4.} Graph showing blood pH during the preoperative (t = 0), post-ANH (t = 20) and throughout the intraoperative (t = 20 to t = 200). Control group (n = 15) represents patients with standard of care. PFC group (n = 15) represents patients treated with Perfluran. Each point represents mean ± SD. *p < 0.05 PFC group vs. Control group.}
\]

\[
\text{\textbf{Figure 5.} Time course of lactate blood values during the preoperative (t = 0), post-ANH (t = 20), and throughout the intraoperative (t = 20 to t = 200) in patients treated with Perfluran (PFC group) and in patients with standard of care (Control group). Although lactate values were higher most of the time in the PFC group than the Control group, the differences were not statistically significant. Each point represents mean ± SD.}
\]

chemistry parameters for both groups, in the preoperative and in the postoperative, are listed in Table 7.

Biological Test

During the induction of anesthesia, patients in the PFC group received a test dose infusion of 30 drops of Perfluran at a rate of 10 drops/minute and were observed for 5 to 10 minutes to test for hypersensitivity. One patient presented allergic reaction (urticaria) during the observation period and as a result was excluded from the study.

Adverse Events

During the present study, Perfluran was well tolerated with no adverse effects attributable to it. There were no major complications or deaths in either group.
DISCUSSION

In this study, ANH was used in combination with PFC to enhance the patient's capacity to tolerate low Hb concentrations during the intraoperative period while maintaining high systemic oxygen levels. The hypothesized outcomes were reduced need of allogeneic blood and fewer complications during and after surgery. The purpose of this study was to evaluate whether Perforan, a PFC, had the necessary characteristics to achieve the expected goals, namely its oxygen carrying capacity and its potential to reduce the need, or avoid the use of transfused allogeneic blood and blood derivatives.

Because of the nature of PFC emulsions, hypersensitivity was of concern. Of the 16 patients subjected to the biological test, one showed an allergic reaction and was excluded from the study. The patient was treated satisfactorily with an intravenous antihistaminic. This case represents a hypersensitivity frequency ratio of 6.25% (1/16).

The entire PFC group responded well to the Perforan infusion and, the overall consumption of allogeneic blood and blood derivatives decreased in comparison with the Control group. There was not a significant reduction in the amount of allogeneic packed RBC units transfused.

Table 4. Intraoperative characteristics

<table>
<thead>
<tr>
<th>Parameter</th>
<th>PFC group (n = 15)</th>
<th>Control group (n = 15)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Anesthesia duration (min)</td>
<td>280 ± 24</td>
<td>246 ± 66</td>
</tr>
<tr>
<td>Surgery duration (min)</td>
<td>198 ± 30</td>
<td>180 ± 46</td>
</tr>
<tr>
<td>CPB duration (min)</td>
<td>86 ± 23</td>
<td>102 ± 46</td>
</tr>
<tr>
<td>ACC duration (min)</td>
<td>60 ± 18</td>
<td>72 ± 30</td>
</tr>
</tbody>
</table>

CPB = Cardiopulmonary bypass, ACC = Aortic cross-clamp. Values are mean ± SD.
Table 5. Hematological parameters in the preoperative and the postoperative

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Preoperative</th>
<th>Postoperative</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>PFC group</td>
<td>Control group</td>
</tr>
<tr>
<td>RBC (10⁶/µL)</td>
<td>4.8 ± 0.7</td>
<td>4.9 ± 0.5</td>
</tr>
<tr>
<td>Hemoglobin (g/dL)</td>
<td>14.6 ± 1.5</td>
<td>14.8 ± 2.0</td>
</tr>
<tr>
<td>Hematocrit (%)</td>
<td>44 ± 5</td>
<td>45 ± 6</td>
</tr>
<tr>
<td>WBC (10⁹/µL)</td>
<td>6.9 ± 2.3</td>
<td>5.7 ± 1.9</td>
</tr>
<tr>
<td>PMN cells (10⁹/µL)</td>
<td>4.2 ± 1.9</td>
<td>3.4 ± 1.2</td>
</tr>
<tr>
<td>Mononuclear cells (10⁹/µL)</td>
<td>2.8 ± 0.8</td>
<td>2.3 ± 0.9</td>
</tr>
<tr>
<td>Platelets (10⁹/µL)</td>
<td>210 ± 47*</td>
<td>172 ± 46</td>
</tr>
</tbody>
</table>

RBC = Red blood cells; WBC = White blood cells; PMN = Polymorphonuclear; Mononuclear cells = Lymphocytes and Monocytes. Values are mean ± SD. *p < 0.05 PFC group vs. Control group. **p < 0.05 for comparisons between preoperative vs. postoperative values in the same group. *p < 0.0001 for comparisons between preoperative vs. postoperative values in the same group.

in individual patients treated with Perforan (Table 3), nevertheless, as a group, the difference was substantial. The PFC group required 12 units of allogeneic packed RBC, while the Control group required 22 units, a 43% reduction of transfused allogeneic packed RBC. Furthermore, the study shows that the percentage of patients that avoided transfusion of allogeneic packed RBC was 62.5% in the PFC group and 40% in the Control group (Table 3). The PFC group also required fewer units of allogeneic blood derivatives such as cryoprecipitate, fresh frozen plasma, and platelets than patients in the Control group. This difference was statistically significant (Table 3).

Table 6. Preoperative and postoperative coagulation parameters

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Preoperative</th>
<th>Postoperative</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>PFC group</td>
<td>Control group</td>
</tr>
<tr>
<td>PT (s)</td>
<td>15.0 ± 3.5</td>
<td>16.0 ± 3.4</td>
</tr>
<tr>
<td>APTT (s)</td>
<td>47.2 ± 16.0</td>
<td>44.5 ± 16.0</td>
</tr>
<tr>
<td>Activity (%)</td>
<td>65 ± 21</td>
<td>59.2 ± 21.3</td>
</tr>
<tr>
<td>Fibrinogen (mg/dL)</td>
<td>258 ± 51</td>
<td>212 ± 41</td>
</tr>
</tbody>
</table>

PT = Prothrombin time; APTT = Activated partial thromboplastin time. Values are mean ± SD.

Table 7. Preoperative and postoperative blood chemistry parameters

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Preoperative</th>
<th>Postoperative</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>PFC group</td>
<td>Control group</td>
</tr>
<tr>
<td>Glucose (mg/dL)</td>
<td>107 ± 25*</td>
<td>144 ± 27</td>
</tr>
<tr>
<td>Urea nitrogen (mg/dL)</td>
<td>37.5 ± 10.1</td>
<td>37.3 ± 12.4</td>
</tr>
<tr>
<td>Creatinine (mg/dL)</td>
<td>1.0 ± 0.3</td>
<td>1.0 ± 0.3</td>
</tr>
<tr>
<td>Na⁺ (mEq/L)</td>
<td>141 ± 4.4</td>
<td>139 ± 4.5</td>
</tr>
<tr>
<td>K⁺ (mEq/L)</td>
<td>4.4 ± 0.2</td>
<td>4.0 ± 0.6</td>
</tr>
<tr>
<td>Ca²⁺ (mEq/L)</td>
<td>7.1 ± 1.7*</td>
<td>9.0 ± 1.5</td>
</tr>
<tr>
<td>Albumin (g/dL)</td>
<td>4.0 ± 0.6</td>
<td>4.4 ± 0.7</td>
</tr>
<tr>
<td>Globulin (g/dL)</td>
<td>2.8 ± 0.6*</td>
<td>2.1 ± 1.0</td>
</tr>
<tr>
<td>Ratio A/G</td>
<td>1.5 ± 0.2*</td>
<td>2.2 ± 0.8</td>
</tr>
<tr>
<td>Total proteins (g/dL)</td>
<td>6.8 ± 0.9</td>
<td>6.3 ± 1.6</td>
</tr>
<tr>
<td>AP (IU/L)</td>
<td>81 ± 24</td>
<td>72 ± 30</td>
</tr>
<tr>
<td>CPK (IU/L)</td>
<td>78 ± 40</td>
<td>93 ± 27</td>
</tr>
<tr>
<td>AST (IU/L)</td>
<td>27 ± 12</td>
<td>34 ± 22</td>
</tr>
<tr>
<td>ALT (IU/L)</td>
<td>26 ± 16</td>
<td>49 ± 57</td>
</tr>
<tr>
<td>T-3 (mg/dL)</td>
<td>1.6 ± 0.4</td>
<td>1.9 ± 0.5</td>
</tr>
<tr>
<td>T-4 (mg/dL)</td>
<td>1.0 ± 0.2</td>
<td>1.3 ± 0.8</td>
</tr>
<tr>
<td>Total cholesterol</td>
<td>175 ± 39</td>
<td>152 ± 56</td>
</tr>
<tr>
<td>Triglycerides (mg/dL)</td>
<td>136 ± 66</td>
<td>127 ± 60</td>
</tr>
<tr>
<td>LDL (mg/dL)</td>
<td>105 ± 40</td>
<td>121 ± 28</td>
</tr>
<tr>
<td>HDL (mg/dL)</td>
<td>37 ± 10</td>
<td>30 ± 8</td>
</tr>
</tbody>
</table>

AP = Alkaline phosphatase; CPK = Creatin phosphokinase; AST = Aspartate aminotransferase; ALT = Alanine aminotransferase; LDL = Low density lipoproteins; HDL = High density lipoproteins. Values are mean ± SD. *p < 0.05 PFC group vs. Control group. **p < 0.05 for comparisons between preoperative vs. postoperative values in the same group.

In spite of the fact that the PFC group reached Hb concentrations significantly lower than the Control group during the intraoperative period (Fig. 1), the study showed that the PFC group had PaO₂ values significantly higher than the Control group (Fig. 2). This attests to the efficacy of Perforan as an oxygen carrier that provided the patient, and surgical team, with ample safety margin to manage low levels of Hb during surgery. A similar behavior was observed during intraoperative bleeding, which was slightly higher in the PFC group, but required less volumes of allogeneic blood transfused. The postoperative Hb concentrations were similar in both groups.
Other authors have reported significant depletion in platelet counts and significant elevation in PMN cell counts after cardiac surgery with CPB [21,22]. This study was no exception. The postoperative platelet counts showed a significant reduction when compared with preoperative levels in both groups (p < 0.05) and the number of PMN cells showed a significant increment after surgery when compared with preoperative levels in both groups (p < 0.0001) (Table 5).

In similar studies using ANH with CPB [23,24], metabolic acidosis has been reported during surgery. Again, in the intraoperative period, patients in both groups experienced certain changes where pH, PaCO₂, HCO₃, lactate and base excess parameter anomalies were observed (Figs. 3–7) without apparent negative consequences. The cause of acidosis seems to be multifactorial. The contributing factors that promote the establishment and progression of this phenomenon are the organism's physiological response mechanisms to preoperative medication, the hemodilution pump, prime solutions, and colloidal and crystalloid solutions used for volume replacement [23,25,26]. Nevertheless, important measures have been developed to control acidosis and avoid its development into a severe acid-base decomposition [24,27,28].

In the present study, blood lactate levels increased persistently, in both groups, from the beginning of the surgical procedure. Significantly higher blood lactate levels were reached in both groups compared with preoperative levels, at 50 minutes of the protocol and onward (p < 0.005). Even though the levels of lactate were slightly higher in the PFC group, there is no reason to assume that Perforan is a major contributing factor since this lactate behavior has been reported in clinical studies where patients underwent cardiac surgery with CPB without PFC administration [29,30]. At present the mechanisms involved in the increase of lactate levels are not completely known. In spite of the lactate levels in the trial, an acceptable range of acidosis was maintained in the patients treated with Perforan infusion. Perforan was well tolerated and the patients in this group experienced no complications attributable to this PFC.

The efficiency of PFC emulsions carrying O₂ and enhancing O₂ unloading capacity has been shown in experimental models of hemorrhagic shock [15,31], ischemia-reperfusion [16], and hemodilution [17]. These emulsions have also been proven to be effective in clinical trials such as heart infarction [32], severe trauma [33], as well as orthopedic [20], cardiac [1,34], abdominal [18] and cancer [18] surgery. Thus, Perforan could be used to enhance the safety of surgery during prolonged periods of hypoxia. It can also serve to reduce the volume of transfused alloimmune blood and blood-derivatives. Perforan has the potential to be used as a safe bridge for alloimmune blood transfusion in cases where blood is not readily available, such as in remote areas or third world countries. Perforan is also a viable alternative when blood is not an option due to religious restrictions.

CONCLUSIONS

The results of this study suggest that infusions of Perforan, in combination with preoperative ANH, are safe and well tolerated by patients undergoing cardiac surgery with CPB. Perforan was effective in transporting O₂ when the patient received a FIO₂ of 1.0 and it enhanced patient safety during periods of intraoperative anemia. Perforan helped a considerable percentage of patients avoid transfusions and it reduced the units of alloimmune packed RBC transfused per patient. It also significantly reduced the use of blood-derivatives. Due to the adverse effects of alloimmune blood transfusions, further studies should be conducted utilizing Perforan in other surgical procedures as well as urgent care applications involving significant blood loss.

REFERENCES