

Original Article

The Relationship Between Membrane Oxygenator Blood Path Pressure Drop and Hemolysis: An In-vitro Evaluation

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ABSTRACT

Plasma hemoglobin (pl-Hgb) production was measured for the Bard HF-5000 (n=4), Sarns SMO (n=4) and Sarns Turbo (n=4) membrane oxygenators (MO), which were tested on separate closed-loop roller pump circuits to compare differences in hemolysis production as a function of pressure drop (P). The three oxygenators were selected because of their varied P characteristics. Circuits were prepared by clamping off the MO and then priming with freshly drawn, heparinized, filtered bovine blood that had an adjusted hemoglobin (Hgb) of 8.0 g/dL. Roller pump occlusions were set using a pressure transducer. To correct for hemolysis resulting from the roller pump in each oxygenator circuit, the blood was pumped through a bypass line around each MO for the first 90 minutes of each study. Blood flows were maintained at 4.0 L/min at a pressure of 200 mmHg and tests for pl-Hgb conducted at 30-minute intervals. Blood was then circulated through each MO for 180 minutes at outlet pressures of 200 mmHg. Total pl-Hgb values were corrected for flow and time in order to calculate a hemolysis index (HI) in grams of pl-Hgb generated per 100 L of blood pumped.

A weak correlation was demonstrated between P and HI ($r = 0.69$). This study suggests that variances in MO hemolysis are a function of design variables, such as membrane surface area, prime volume, and blood path length, rather than of P. The pl-Hgb generated from each MO was negligible when compared with typical pl-Hgb levels generated during routine cardiopulmonary bypass.

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INTRODUCTION

Blood circulating through an extracorporeal circuit encounters numerous mechanical and chemical forces that, to varying degrees, destroy or alter the cellular components. Concern for reducing hemolysis, decreasing complement activation, and preserving clotting mechanisms has had an impact on material and design requirements for perfusion products. Total blood damage results from the cumulative effects of all the components in the extracorporeal circuit and from the manner in which they are used. Oxygenators deserve particular scrutiny because of their complex, diverse designs and their demanding performance requirements. Recent membrane oxygenator (MO) designs have sought to enhance gas and heat transfer, while at the same time reducing blood contact with foreign surfaces. As a consequence, companies have introduced smaller devices, less traumatic material surfaces, less gas exchange surface area, lower prime volumes and, more recently, biocompatible coatings.

Most MOs are designed to be placed in the arterial line between the pump and the patient. This position results in varying degrees of resistance to arterial flow, which can be quantified by measuring the pressure drop (P) between the venous inlet and the arterial outlet. The intent of this investigation was to examine the relationship between oxygenator blood path P and hemolysis generation, and to determine the clinical relevance, if any, of this hemolysis.

MATERIALS AND METHODS

The Bard HF-5000^a (n = 4), Sarns SMO^b (n = 4), and Sarns Turbo^b (n = 4) MOs were chosen specifically for their differences in P characteristics, which were measured in preliminary in-vitro tests.

Four identical closed-loop test circuits were set up on a pump console. Three of the circuits incorporated one of the MOs chosen for the investigation (Figure 1). The circuits consisted of a 2 L flexible venous reservoir, a roller pump^b, and 3/8" I.D. x 3/32" tubing with a bypass line placed around the MO. Hemolysis production in these circuits represents the sum of hemolysis generated by the roller pump and the MO. To determine the hemolysis generated by the MO alone, the typical hemolysis generated by the roller pump was measured utilizing a fourth control circuit without an oxygenator.

Freshly drawn, heparinized, bovine blood from fasted animals (1) was adjusted, using lactated Ringer's solution, to a target hemoglobin of 8.0 g/dL, and the pH normalized using sodium bicarbonate. Clamps were placed between the pressure monitoring sites and the oxygenator ports, and the circuits were primed with 2 L of blood through the bypass line. Using blood prime is not normal perfusion practice but is the standard technique for most in-vitro test procedures (2). In this model, the blood prime served to ensure more consistent hematocrit levels among the circuits.

To set roller pump occlusion, a clamp was placed at the

Figure 1

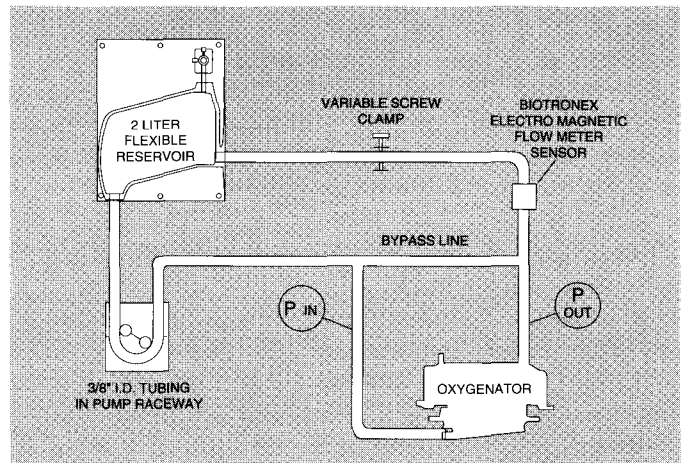
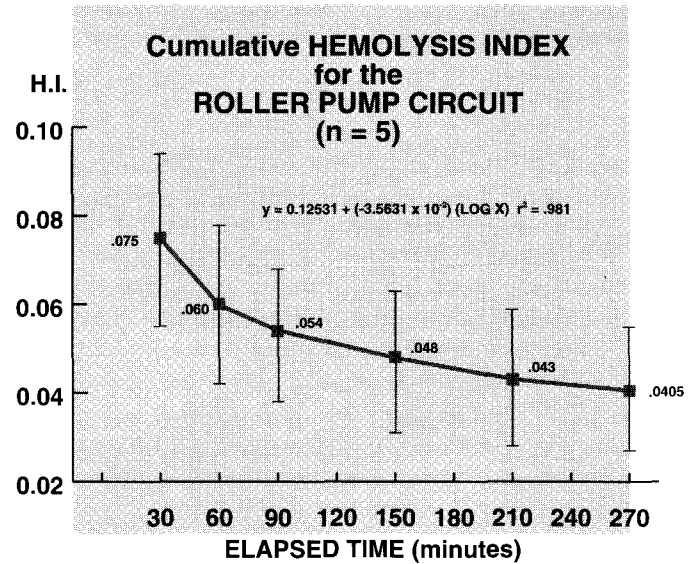


Figure 2



inlet to the 2 L reservoir and the pump slowly turned to achieve a static pressure of 500 mmHg in the circuit. Occlusion was then backed off to allow a gradual pressure drop to 250 mmHg during a 30-second period. Blood flows were regulated at 4.0 L/min with an in-line Biotronex BL 610 electromagnetic flow meter^c. Oxygenator inlet and outlet pressures were continuously monitored with a Gould transducer/amplifier system^d. Each circuit ran at room temperature. To eliminate occlusion setting variables associated with discrete differences in roller-to-raceway tolerance, four roller pumps were used. Each MO and control circuit was run on each of the four pumps.

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- b Sarns, 3M Health Care, Ann Arbor, MI 48103
- c Biotronex Laboratory, Chester, MD 21619
- d Gould Instrument Systems Division, Cleveland, OH

Figure 3

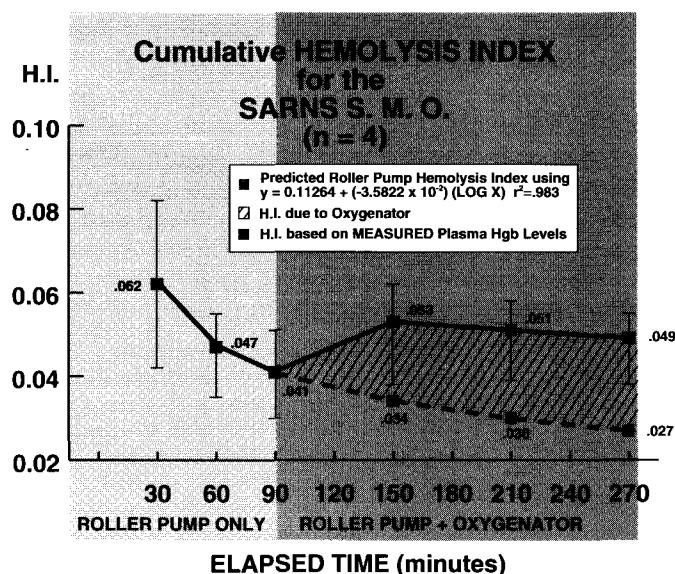


Figure 5

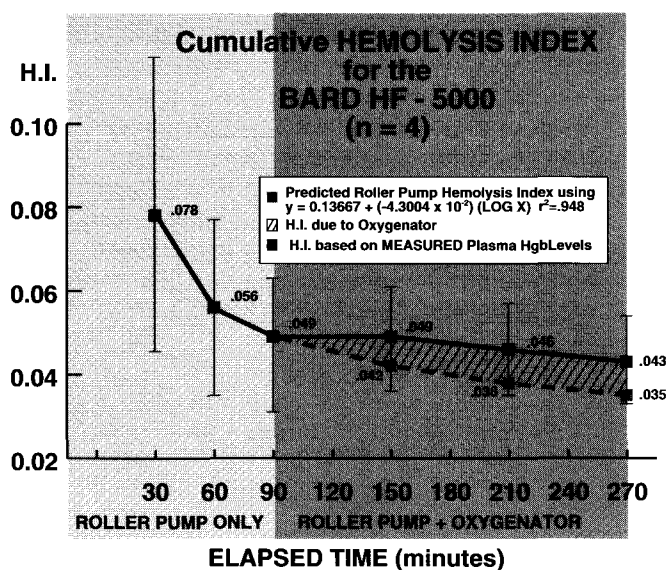
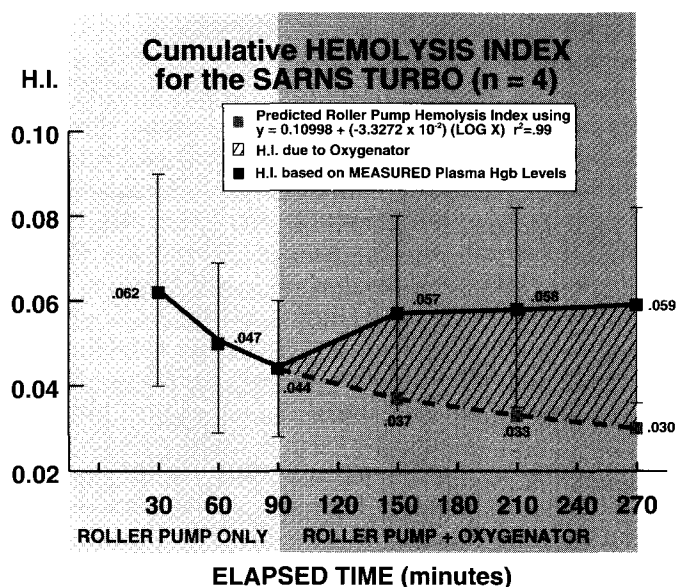


Figure 4



For the first 90 minutes, blood flow was maintained through the bypass loop only. Pump outlet pressures were adjusted to 200 mmHg with a variable screw clamp. Samples were collected every 30 minutes and analyzed for pl-Hgb with a Sargent-Welch UV/VIS spectrophotometer^e. The oxygenators were then filled with blood from their respective reservoirs, the bypass lines clamped, and the flow resumed for 180 minutes. Oxygenator outlet pressures were maintained at 200 mmHg. Samples were collected every hour and analyzed for pl-Hgb. The control circuit

e Sargent-Welch Instruments, Pye Unicam Ltd., Cambridge, England

was regulated and monitored in the same way.

Pl-Hgb values were used to calculate the hemolysis index (HI), a formula that compensates for differences in circuit volume, hematocrit and elapsed time on pump. The HI represents the grams of pl-Hgb generated per 100 L of pumped blood, as demonstrated here:

1. Determination of total pl-Hgb (mg) generated since time = 0

$$\text{Pl-Hgb (mg/dL)} \times \text{blood volume (mL)} \times (1 - \text{av Hct \%}) \times 0.01$$

[0.01 = correction factor for plasma volume]

2. Determination of the cumulative HI

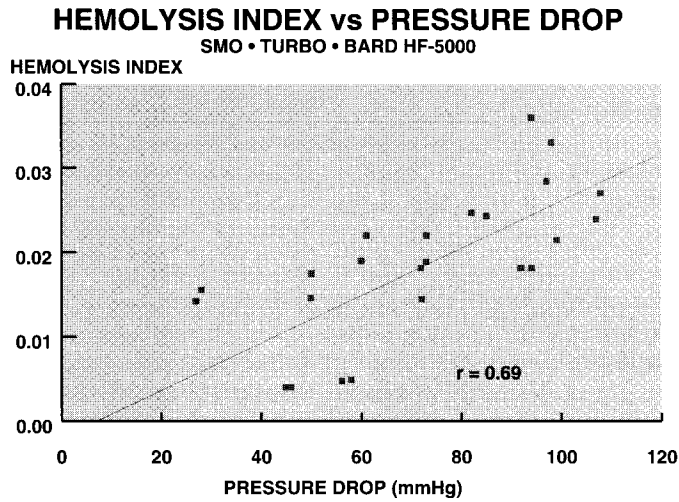
$$\frac{\text{Total pl-Hgb (mg)} \times 0.1}{\text{Blood flow (L/min)} \times \text{elapsed time (min)}}$$

[0.1 = conversion factor for mg to g and L to 100 L]

RESULTS

Relationship Between Hemolysis and P: The average cumulative HI for control circuits is depicted in Figure 2. The curve represents the cumulative HI attributable to the roller pump during 270 minutes of operation. Pump-generated hemolysis was the greatest single variable influencing hemolysis in this investigation. The wide variation in HI at each time interval likely was attributable to inconsistent occlusion settings despite attempts to be precise. The definition of the roller pump curve [$y = 0.12531 + (-3.5631 \times 10^{-2}) (\text{LOG } X)$] was used to correct for the cumulative HI contributed by the roller pumps in each of the

Figure 6



oxygenator circuits during the 180 minutes when the pumps were generating flow through the MOs. The initial rise in pl-Hgb was attributed to weaker cells being destroyed during the start of roller pumping. Pl-Hgb would typically level off after 90 minutes which is when the oxygenators were included into the circulation.

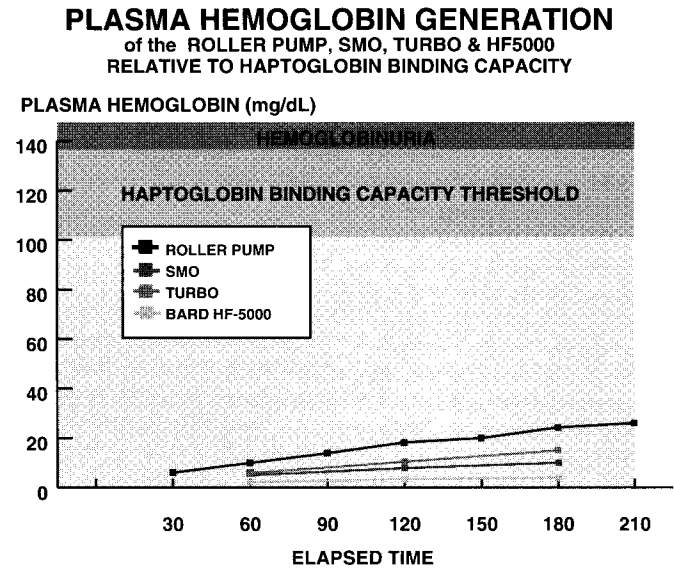
Adjusted cumulative hemolysis values for the SMO, Turbo, and Bard oxygenators are graphed in Figures 3, 4, and 5 respectively. The average HI was .021 for the SMO unit, .025 for the Turbo, and .008 for the Bard. The average P was 78 mmHg for the SMO device, 94 mmHg for the Turbo, and 45 mmHg for the Bard. A scattergram of all HI and P data produced a correlation coefficient (r) of 0.69 (Figure 6). Because the intent of this investigation was to determine the influence of P on hemolysis, no statistical comparison of HI for the different oxygenators was necessary.

Clinical Relevance of Oxygenator Hemolysis Generation: The rate of pl-Hgb generation was 3.6 mg/dL per hour for the SMO device, 5.3 mg/dL per hour for the Turbo, and 1.1 mg/dL per hour for the Bard. Figure 7 plots pl-Hgb generation for each of the oxygenators relative to roller pump hemolysis and haptoglobin (Hp) binding capacity.

DISCUSSION

Extensive research suggests that when all the sources of hemolysis in the extracorporeal circuit are considered, the hemolysis imparted by the MO is relatively benign. The primary physical forces applied to blood cells in a cardiopulmonary bypass system are shear stress, pressure, wall impact, and surface phenomena (3-6). Red blood cells (RBCs) can withstand considerable shear stress, which alone does not induce hemolysis (7). In-vitro studies of the effects of shear on human donor blood demonstrated that little hemolysis occurs when shear stress is $<3,000$ dynes/cm². At higher shear stress levels, hemolysis increased greatly (8). The typical shear stress calculated for an MO under physiologic conditions of flow and blood viscosity is one-tenth

Figure 7



the magnitude (approximately 200-300 dynes/cm²) necessary to cause hemolysis (9). RBC injury does become evident when high shear stress is combined with RBC-wall interaction or exposure to a foreign surface (10, 11), as would be expected with over-occluded roller pumps, high blood flows through small cannulae tips (12), cardiotomy suction, and materials interaction within the extracorporeal circuit (13). Studies examining the influence of pressure on erythrocyte destruction indicate that blood can tolerate frequent positive pressure compressions as high as 2,300 mmHg with relatively little hemolysis. Conversely, RBCs exposed to negative pressures as small as 300 mmHg resulted in occasional high hemolysis (11, 14). In-vitro studies with roller pumps demonstrated a direct relationship between hemolysis and Reynolds Number (14), occlusion settings, tubing materials, roller geometry, tubing diameter (15), and roller pump speed (16, 17).

When applying in-vitro hemolysis data to the clinical setting, one must consider the body's natural mechanisms for clearing pl-Hgb from the bloodstream. Ninety-seven percent (97%) of the solid content of an RBC is Hgb. Hgb released into the plasma can combine with haptoglobin (Hp), an alpha-2 globulin. Normal human plasma contains enough Hp to bind from 75-175 mg of Hgb per 100 ml of plasma (18, 19). Because of its large molecular weight, the Hgb-Hp complex is too big to be filtered by the normal renal glomeruli. Instead, Hgb-Hp is cleared through the reticuloendothelial system. The liver is the primary site for Hgb-Hp catabolism.

Clinical manifestations of RBC hemolysis (i.e., hemoglobinuria) are rare due to the body's mechanisms for clearing serum Hgb. The normal pl-Hgb production during routine cardiopulmonary bypass is in the range of 25-35 mg/dL per hour (6, 21). Therefore, it would be uncommon for pl-Hgb levels to exceed 60 mg/dL unless the time on bypass is extended or there is excessive cardiotomy suction volume (22, 23). In the

normal person, any pl-Hgb arising from hemolysis is disposed of either by filtration through the kidneys or by combining with Hp. Clinical studies by Laurell and Nyman (18) demonstrated that Hgb concentration in the plasma decreased at a constant rate of 13 mg/dL per hour as a result of Hgb-Hp clearance and reabsorption through the renal glomeruli. Hp concentration is the dominant method for clearance. In fact, a clearance rate of 13 mg/dL per hour exceeds the rate of pl-Hgb production noted for the MOs studied in this investigation. Hemoglobinuria does not occur until the pl-Hgb concentration exceeds the threshold level of the binding capacity for Hp (18, 20). Once hemoglobinuria occurs, it may continue until the pl-Hgb falls to 30-50 mg/dL.

Oxygen transfer with an MO is based on the filming properties of the blood over the membrane surface. Designs that induce blood-side convective mixing enhance oxygen transfer performance. Hollow fiber devices with blood flow inside the fibers cannot induce mixing. Such oxygenators rely solely on a large membrane surface area to achieve adequate gas transfer and, therefore, exhibit a low mass transfer coefficient. Conversely, hollow fiber devices with blood flow outside the fiber rely on bundle geometry to promote blood mixing. Such devices exhibit a high mass transfer coefficient and, therefore, are able to perform with reduced membrane surface area (approximately 2 m²). The mixing used to achieve a high mass transfer coefficient (or oxygen transfer) induces wall interaction between the membrane surface and the RBC, which in turn affects pl-Hgb production.

Blackshear, et al. (11) proposed that the rate of pl-Hgb generation can be expressed as a function of a system's mass transfer coefficient per unit of surface area. Their research suggests that an MO with high mass transfer (or oxygen transfer) and a small membrane surface area would generate more pl-Hgb than a similarly performing oxygenator that has a larger membrane surface area. In this investigation, a stronger correlation ($r = 0.89$) was found when comparing HI with device oxygen transfer per square meter of membrane surface area. This finding suggests that HI is more strongly related to the design variables that influence oxygen transfer than to P.

CONCLUSION

We found a weak correlation ($r = 0.69$) between oxygenator P and hemolysis generation. In the context of these findings and results of related studies, the P resulting from blood flow through an MO does not generate hemolysis. Furthermore, P under normal physiologic conditions is not a reliable indicator of device hemolysis generation. Regardless of the mechanisms responsible for hemolysis, the hemolysis produced by MOs is clinically negligible relative to roller pump hemolysis, Hp binding capacity, and normal physiologic clearance mechanisms. In the clinical setting, the hemocompatibility of an MO is often overshadowed by cardiotomy suction, which is still regarded as the primary source of hemolysis in the cardiopulmonary bypass circuit.

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