Effect of perfluorocarbons on brain oxygenation and ischemic damage in an acute subdural hematoma model in rats

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Object. This study was conducted to determine whether perfluorocarbons (PFCs) improve brain oxygenation and reduce ischemic brain damage in an acute subdural hematoma (SDH) model in rats.

Methods. Forty adult male Sprague–Dawley rats were allocated to four groups: 1) controls, acute SDH treated with saline and 30% O2; 2) 30-PFC group, acute SDH treated with PFC infusion in 30% O2; 3) 100-O2 group, acute SDH treated with 100% O2; and 4) 100-PFC group, acute SDH treated with PFC plus 100% O2. Ten minutes after the induction of acute SDH, a single dose of PFC was infused and 30% or 100% O2 was administered simultaneously. Four hours later, half of the rats were killed by perfusion for histological study to assess the extent of ischemic brain damage. The other half were used to measure brain tissue oxygen tension (PO2). The volume of ischemic brain damage was 162.4 ± 7.6 mm3 in controls, 165.3 ± 11.3 mm3 in the 30-PFC group, 153.4 ± 17.3 mm3 in the 100-O2 group, and 95.9 ± 12.8 mm3 in the 100-PFC group (41% reduction compared with controls, p = 0.002). Baseline brain tissue PO2 values were approximately 20 mm Hg, and after induction of acute SDH, PO2 rapidly decreased and remained at 1 to 2 mm Hg. Treatment with either PFC or 100% O2 improved brain tissue PO2, with final values of 5.14 and 7.02 mm Hg, respectively. Infusion of PFC with 100% O2 improved brain tissue PO2 the most, with a final value of 15.16 mm Hg.

Conclusions. Data from the current study demonstrated that PFC infusion along with 100% O2 can significantly improve brain oxygenation and reduce ischemic brain damage in acute SDH.

Key Words • brain tissue oxygenation • acute subdural hematoma • cerebral ischemia • perfluorocarbon • rat

Recent advances in brain monitoring techniques have provided new data on the time-dependent changes in brain tissue oxygenation after severe head injury in humans and revealed that low brain tissue PO2 levels are a very common finding.14,15,34,35,54-56 Moreover, the lowest brain tissue PO2 levels are reported to be associated with increased morbidity and mortality rates35,55 and correlate closely with reduced regional blood flow.4,6 It therefore seems logical to search for ways to improve brain tissue oxygenation in these circumstances.5

Acute SDH is the most common mass lesion occurring after severe head injury.19,44 Despite major advances in the treatment of head-injured patients during the past two decades, acute SDH continues to be one of the most lethal of all intracranial injuries.59 Although a number of mechanisms have been proposed to account for brain damage that occurs following SDH, data from neuropathological studies have shown that cerebral ischemia is the most central mechanism integral to the pathogenesis of secondary brain damage in those who die after acute SDH.1,21,33 Given that the prevention of secondary brain damage in severely head injured patients is the most fundamental goal, a method of overcoming this ischemia, such as the enhancement of O2 delivery to cells of the endangered tissue, would be of great benefit.

Perfluorocarbons are fluorinated compounds originally developed during the Manhattan project to handle reactive uranium isotopes.3 These compounds are biologically inert, fluorine-substituted hydrocarbon chains that have the ability to dissolve large amounts of gases including O2 and CO2.20,40,41,48 Two major mechanisms underlie the efficacy of PFCs for O2 transport and delivery. First, they have a high solubility coefficient for O2 and CO2, with a negligible O2-binding constant. Thus, they do not bind O2 as a ligand (as in Hb) but rather enhance the solubility of O2 in plasma. When equilibrated with 1 atm of O2, pure PFCs can dissolve approximately 50 ml/dl of O2 or almost 2.5 times that of whole blood. Second, once emulsified, the PFC emulsion droplets are extremely small, with a median size of approximately 0.2 μm, compared with the RBC diameter of 7.0 μm. This small size allows PFC emulsion particles to fill the RBC gaps, thereby expanding O2 transport to the entire surface area of the capillary, and facilitates their passage through capillaries and partially blocked or constricted arterioles.
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microcirculatory vessels. Therefore, these compounds could have potential benefits in a wide variety of pathological conditions during which O₂ delivery to the tissue is compromised.

In the current study, we investigated the ability of a PFC emulsion to improve brain oxygenation and reduce ischemic brain damage in the management of an acute SDH in which cerebral ischemia is the main pathophysiological finding responsible for poor outcome.

Materials and Methods

All experiments were performed under a protocol approved by the Institutional Animal Care and Use Committee at Virginia Commonwealth University. Animals were housed in the animal facility, with 12-hour light/dark cycles and were provided with food and water ad libitum. Forty adult male Sprague–Dawley rats (Harlan, Indianapolis, IN) weighing 310 to 380 g each were used.

Surgical Preparation

Animals were allocated to four groups: 1) control group, acute SDH treated with intravenous saline infusion while breathing in 30% O₂; 2) 30-PFC group, acute SDH with intravenous PFC emulsion infusion with administration of 30% O₂; 3) 100-O group, acute SDH and administration of 100% O₂ without PFC infusion; and 4) 100-PFC group, acute SDH induction and intravenous PFC emulsion infusion with simultaneous administration of 100% O₂. Each group comprised 10 rats; five animals were used for volumetric study to measure the extent of ischemic damage and another five rats were continuously monitored for brain tissue PO₂.

The model of acute SDH in rats, which is characterized by focal ischemic brain damage, has been extensively studied by our group and others. Animals were prepared for the induction of acute SDH as previously described. Briefly, rats were placed in a Plexiglas chamber and anesthetized using a mixture of N/O₂/H₁₇₀₂₀₂₂₉% isoflurane. Animals were then intubated and re-breathed with a mixture of N/O₂/H₁₇₀₂₀₂₂₉% isoflurane. When the measurements were completed, the probes were tested again for zero drift by using a zero solution (REF VM5; GMS, Kiel-Mielendorf, Germany) was inserted 6 mm past the dura into the subdural space over 5 minutes. Following injection, the subdural space was sealed when the probability value was less than or equal to 0.05.

Drug and O₂ Administration

The PFC emulsion used in this study was Oxygent (Alliance Pharmaceutical Corp., San Diego, CA), a second-generation sterile and concentrated 60% w/v PFC emulsion based on perfluoron (perfluorocetyl bromide, CF₂Br). Oxygent is intended for intravenous infusion and contains 0.6 g PFC/ml emulsion. The PFC is emulsified with egg yolk phospholipids, and the emulsion is suspended in phosphate-buffered saline. Ten minutes after induction of acute SDH, animals received a single intravenously infused dose of perfluorobutane emulsion (4.5 ml/kg, which is equivalent to 2.7 g PFC/kg body weight) or the same volume of sterile saline. Solutions administered in all groups were infused over 5 minutes at a rate of 250 μl/minute. For groups with 100% O₂ administration, the FlO₂ was increased to 1 with the start of PFC or saline infusion and was maintained throughout the 4 hours of the experiment.

Histological Study

Animals targeted for measurement of ischemic brain damage were killed by perfusion fixation while in a state of deep anesthesia 4 hours after the induction of acute SDH. Five rats, a number sufficient for statistical analysis, were allocated to each experimental group. Briefly, after increasing the isoflurane level to 4% for 2 minutes, a thoracotomy was performed and a catheter was passed through the left ventricle into the ascending aorta. The right atrium was incised, and the rat was perfused with 100 ml of phosphate-buffered saline followed by 100 ml 4% paraformaldehyde. Immediately after perfusion, the animal was decapitated. The brain was then removed from the skull; the hindbrain was detached and immediately frozen in liquid isopentane chilled with dry ice. Serial 12-μm-thick coronal tissue sections were cut using a cryostat and stained with H & E. Sections, which corresponded to eight predetermined stereotaxic planes distributed throughout the forebrain from 1 to 10.5 mm anterior according to König and Klippel, were selected and examined using light microscopy. The extent of ischemic damage at the eight predetermined planes was then measured using a commercially available program (Olympus Image System CAST; Olympus, Ballenup, Denmark). The volume of ischemic damage was calculated by integrating the areas over the known distance between each coronal level by using the method of Osborne and colleagues.

Monitoring Brain Tissue PO₂

Brain tissue PO₂ was measured using a Licox CMP tissue monitoring system and a Licox CC1.r tissue O₂ catheter (both Integra Neuroscience). The Licox CC1.r probe is a commercially available Clark microelectrode that has a diameter of 0.5 mm, a length of 200 mm, and an O₂-sensitive area of 1 mm². The brain tissue PO₂ data were collected using a MacLab data recording system (AD Instruments Pty Ltd., Castle Hill, NSW, Australia). After its placement in the rat brain, the Licox catheter was allowed to stabilize for 60 minutes while the animals were breathing in 30% O₂. Proper function of the probe in situ was confirmed by its response (increase in brain tissue PO₂, > 25 mm Hg within 90 seconds) after an O₂ challenge (transiently increasing FlO₂ to 100%). Given that this study was designed to measure the effect of O₂ treatment, any rat that showed a poor response after the O₂ challenge was excluded and the final number of animals included in each group was five. At the end of the 4-hour monitoring period, the rats were killed, the brains were removed and chilled on dry ice, and 2-mm coronal sections were cut using a brain matrix. If there were microhemorrhages in either the probe tract or ventricles, the animal was excluded. When the measurements were completed, the probes were tested for zero drift by using a zero solution (REF VMS; GMS, Kiel-Mielendorf, Germany) and for sensitivity drift at room air pressure. Actual room air PO₂ was calculated from local barometric pressures.

Statistical Analysis

Data analysis was performed using an ANOVA, a t-test, and a Mann–Whitney rank-sum test. All data were expressed as the means ± SEM for each group. A significant difference was assumed when the probability value was less than or equal to 0.05.

Results

Physiological Variables

Mean arterial blood pressure and arterial blood gas val-
ues are shown in Table 1. After induction of the SDH, there was early, transient but insignificant elevation of the MABP in all groups. Infusion of PFC alone did not affect any hemodynamic variable. Administration of 100% O₂ with or without PFC elevated PO₂ over 400 mm Hg (3.5- to four-fold increase; paired t-test, p < 0.05), and this increase was greatest in the group treated with PFC and 100% O₂. Although the difference was not statistically significant (unpaired t-test, p > 0.05). Compared with the baseline blood pressure, hyperoxic treatment seemed to elevate blood pressure, but the elevation was not statistically significant (paired t-test, p > 0.05).

**Histological Study**

In all 20 rats (five in each group) designated for histological studies, large SDHs were visible overlying the left hemisphere when the brains were removed. Boundaries between the areas of infarction and normal adjacent areas were sharply demarcated by a zone of pale staining on tissue samples with H & E. This result was consistent with what we observed previously.¹³

The volumes of ischemic damage in all groups are shown in Fig. 1. The extent of ischemic damage, expressed as the mean infarct volume, was 162.4 ± 7.6 mm³ in the control group, 165.3 ± 11.3 mm³ in the 30-PFC group, 153.4 ± 17.3 mm³ in the 100-O₂ group, and 95.9 ± 12.8 mm³ in the 100-PFC group. There was a highly significant 41% reduction in the infarct volume in the 100-PFC group when compared with that in the control group (p = 0.002). Administration of PFC combined with 100% O₂ reduced the extent of ischemic damage in all eight predetermined stereotactic planes and was most significant in the frontal regions of the forebrain (Fig. 2).

**Monitoring of Brain Tissue PO₂**

After 60 minutes of stabilization, baseline brain tissue PO₂ values in all four groups were measured at approximately 20 mm Hg. In the control group, after the SDH was induced, brain tissue PO₂ values rapidly decreased to 1 to 2 mm Hg within 10 minutes and remained less than 2 mm Hg, with a final mean value of 1.86 mm Hg at the end of the experiment. In the other groups, however, following PFC infusion and/or administration of 100% O₂, the reduced PO₂ values began to increase, with final values of 5.14 mm Hg in the 30-PFC group, 7.02 mm Hg in the 100-O₂ group, and 15.16 mm Hg in the 100-PFC group. The differences in the final PO₂ values among the four groups were statistically significant over time (ANOVA, p < 0.05). When compared with brain tissue PO₂ values in the control group, however, the small PO₂ increases observed in the 30-PFC group were not statistically significant at all measured time points (t-test, p > 0.05), whereas the moderate increases in the 100-O₂ group were statistically significant at six of 10 time points. Nevertheless, the marked increases in the 100-PFC group were statistically significant at all measured time points (t-test, p < 0.05; Fig. 3).

Postmeasurement calibration of the Licox probe revealed a mean zero drift of 0.18 ± 0.07 mm Hg. Sensitivity drift, calibrated at a mean room air PO₂ of 155.3 ± 0.9 mm Hg, was 1.6 ± 0.3% and was not significantly different among groups.

**Discussion**

The prevention of secondary brain damage, thereby improving outcome, is the most important goal in the management of head injuries. Evidence of ischemic neuropathological damage can be found in 80 to 90% of severely head-injured patients on autopsy.¹²¹ Tissue hypoxia leading to cerebral “ischemic neuronal damage” is the final pathway common to many different types of secondary brain damage.²⁴⁻⁶⁰ Therefore, improving brain tissue oxygenation is a logical and important strategy in the treatment of head injuries.

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**TABLE 1**

<table>
<thead>
<tr>
<th>Treatment Group &amp; Variable (mm Hg)</th>
<th>Before Induction</th>
<th>10 Mins</th>
<th>1 Hour</th>
<th>2 Hours</th>
<th>3 Hours</th>
<th>4 Hours</th>
</tr>
</thead>
<tbody>
<tr>
<td>control</td>
<td>MABP 90.5 ± 4.6</td>
<td>97.0 ± 9.7</td>
<td>91.5 ± 6.2</td>
<td>90.0 ± 4.4</td>
<td>90.3 ± 4.0</td>
<td>91.3 ± 4.1</td>
</tr>
<tr>
<td></td>
<td>PaO₂ 121.3 ± 4.1</td>
<td>123.5 ± 1.5</td>
<td>121.0 ± 4.4</td>
<td>112.0 ± 3.0</td>
<td>114.3 ± 6.3</td>
<td>123.7 ± 9.1</td>
</tr>
<tr>
<td></td>
<td>PaCO₂ 39.5 ± 1.0</td>
<td>38.8 ± 1.1</td>
<td>37.5 ± 2.5</td>
<td>39.8 ± 2.0</td>
<td>40.2 ± 0.5</td>
<td>38.5 ± 2.1</td>
</tr>
<tr>
<td>PFC + 100% O₂</td>
<td>MABP 89.8 ± 1.4</td>
<td>94.0 ± 3.8</td>
<td>95.2 ± 2.4</td>
<td>96.0 ± 1.1</td>
<td>93.4 ± 3.2</td>
<td>92.8 ± 3.2</td>
</tr>
<tr>
<td></td>
<td>PaO₂ 110.2 ± 2.3</td>
<td>107.4 ± 4.7</td>
<td>477.6 ± 28.6</td>
<td>478.6 ± 20.1</td>
<td>455.8 ± 26.3</td>
<td>476.2 ± 23.5</td>
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<tr>
<td></td>
<td>PaCO₂ 38.6 ± 1.2</td>
<td>36.6 ± 1.3</td>
<td>38.0 ± 1.3</td>
<td>35.0 ± 1.2</td>
<td>37.0 ± 1.1</td>
<td>36.2 ± 1.5</td>
</tr>
<tr>
<td>PFC + 30% O₂</td>
<td>MABP 92.2 ± 2.9</td>
<td>92.2 ± 2.2</td>
<td>93.6 ± 2.8</td>
<td>90.6 ± 2.4</td>
<td>90.0 ± 2.5</td>
<td>88.5 ± 2.3</td>
</tr>
<tr>
<td></td>
<td>PaO₂ 115.8 ± 8.8</td>
<td>119.0 ± 7.5</td>
<td>118.4 ± 8.7</td>
<td>127.4 ± 7.2</td>
<td>118.2 ± 7.7</td>
<td>120.7 ± 12.2</td>
</tr>
<tr>
<td></td>
<td>PaCO₂ 39.4 ± 1.7</td>
<td>40.2 ± 1.6</td>
<td>37.8 ± 2.5</td>
<td>39.2 ± 1.6</td>
<td>37.0 ± 0.9</td>
<td>40.2 ± 0.3</td>
</tr>
<tr>
<td>100% O₂</td>
<td>MABP 89.4 ± 0.8</td>
<td>96.4 ± 4.1</td>
<td>94.2 ± 1.1</td>
<td>95.8 ± 1.0</td>
<td>92.4 ± 0.8</td>
<td>92.2 ± 1.9</td>
</tr>
<tr>
<td></td>
<td>PaO₂ 119.6 ± 4.1</td>
<td>124.2 ± 4.8</td>
<td>441.4 ± 40.6</td>
<td>412.6 ± 43.1</td>
<td>426.5 ± 35.2</td>
<td>444.0 ± 33.8</td>
</tr>
<tr>
<td></td>
<td>PaCO₂ 38.2 ± 1.0</td>
<td>38.0 ± 1.7</td>
<td>40.0 ± 1.1</td>
<td>38.8 ± 1.0</td>
<td>39.6 ± 1.3</td>
<td>38.6 ± 1.0</td>
</tr>
</tbody>
</table>

* Significant increases in PaO₂ between 1 and 4 hours after hematoma induction were observed in groups treated with PFC plus 100% O₂ or 100% O₂ alone. Values are expressed as the means ± SEM.
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**Oxygen Treatment**

In normal conditions, more than 90% of the O\(_2\) consumed by the brain is used to generate adenosine triphosphate; the mitochondria are responsible for this O\(_2\) consumption.\(^{43}\) Transport of O\(_2\) from ambient air to the mitochondria occurs by diffusion down a stepwise decrease in the O\(_2\) pressure gradient.\(^{60}\) The amount of O\(_2\) in the blood can be expressed as the CaO\(_2\), which can be calculated by adding the amount of O\(_2\) bound to Hb to the amount of O\(_2\) dissolved in the plasma. In normal physiological conditions, a PaO\(_2\) of 100 mm Hg gives a CaO\(_2\) of 20.4 ml/dl, of which 20.1 ml/dl is carried by Hb (99%) and 0.3 ml/dl is dissolved in the plasma, making the dissolved O\(_2\) portion trivial. By increasing the FiO\(_2\), it is possible to increase the amount of dissolved O\(_2\) in the plasma without altering the amount of O\(_2\) bound to Hb. By increasing the FiO\(_2\) to 100%, the PaO\(_2\) can theoretically increase more than 600 mm Hg and raise the dissolved O\(_2\) to 2 ml/dl and the portion of the O\(_2\) dissolved in the plasma to approximately 9%.

It has been argued that by simply increasing the PaO\(_2\), there would be no effect on increasing O\(_2\) delivery because the majority of O\(_2\) carried in the blood is bound to Hb. In normal circumstances, this small increase in O\(_2\) content may not be so important. Nevertheless, in areas of acellular capillary flow where there are no RBCs due to the small capillary lumen, the non-Hb–dissolved O\(_2\) transport may become an important source of tissue O\(_2\) delivery. Confocal microscopic study of the living rat brain cortex has revealed that 10 to 20% of cerebral capillaries may not contain RBCs at any given time.\(^{29,30}\) In addition, it has been demonstrated that even in experimental models with complete arrest of RBC flow such as middle cerebral artery occlusion, acellular plasma perfusion persists in most of the areas supplied by the middle cerebral artery.\(^{51}\) It has also been demonstrated that neuronal mitochondria require an intracellular PO\(_2\) of only 1.5 mm Hg to maintain oxidative glucose metabolism.\(^{3,45}\) These findings indicate that in certain conditions, non-Hb O\(_2\) transport may be more significant than previously thought and that even a small increase in tissue PO\(_2\) may be adequate to afford some degree of neuroprotection. This line of thinking is the rationale for any treatments capable of increasing the amount of O\(_2\) carried in the plasma.

Conversely, there are several concerns about the safety of therapeutic O\(_2\). If prolonged, O\(_2\) treatment can be harmful in patients with obstructive pulmonary disease.\(^{32,46}\) Oxygen toxicity is a function of both partial pressure and exposure time;\(^{39}\) thus, long-term administration of O\(_2\) with a high FiO\(_2\) level should be undertaken cautiously. There is also a theoretical concern that higher blood O\(_2\) levels increase the production of O\(_2\)-free radicals. Note, however, that data from several studies have revealed that free radical injury does not increase with enhanced O\(_2\) delivery.\(^{2,14,49}\) Another concern with O\(_2\) treatment is cerebral arterial vasocostriction, which theoretically could reduce blood flow to infarcted areas. Oxygen is known to cause vasoconstriction and decrease cerebral blood flow in the normal brain.\(^{39}\) Nevertheless, paradoxical elevations in blood flow have been documented in the ischemic brain after hypoxia, indicating that the effects of hyperoxia may differ in ischemic and nonischemic tissues,\(^{39}\) which may represent a teleological/physiological adaptive mechanism. Singhal, et al.\(^{46}\) reported that cortical regional cerebral blood volume tended to increase with hyperoxia because O\(_2\) might prevent microvascular collapse in ischemic tissues. This propensity may represent another potential neuroprotective mechanism of O\(_2\) treatment.

**Perfluorocarbons and TBI**

Perfluorocarbons are inert organic substances produced by substituting fluorine for hydrogen in specific positions within highly stable carbon chains.\(^{40,41}\) They are not metabolized by the body, and after phagocytosis of PFC emulsion...
Because of their low water solubility, PFCs first content that may, these increases in the diffusion level rapidly decreased to 1 to 2 mm Hg and remained less than 2 mm Hg. In the treated groups, however, as PFC infusion and/or 100% O₂ administration was performed 10 minutes after hematoma induction, the decreased PO₂ values increased to the final values of 5.14 mm Hg for the PFC plus 30% O₂ group, 7.02 mm Hg for the 100% O₂ group, and 15.16 mm Hg for the PFC plus 100% O₂ group. The differences in the PO₂ values among the four groups were statistically significant (ANOVA, \( p < 0.05 \)). When compared with the control group, the marked increases in the PFC plus 100% O₂ group were statistically significant at all measured time points (t-test, \( p < 0.05 \)). #p < 0.05 (t-test) compared with controls. Tx = treatment.

**Fig. 3.** Graph illustrating the results of brain tissue PO₂. Baseline brain tissue PO₂ values in all four groups were approximately 20 mm Hg. After the induction of SDH, these values rapidly decreased to 1 to 2 mm Hg and remained less than 2 mm Hg. In the treated groups, however, as PFC infusion and/or 100% O₂ administration was performed 10 minutes after hematoma induction, the decreased PO₂ values increased to the final values of 5.14 mm Hg for the PFC plus 30% O₂ group, 7.02 mm Hg for the 100% O₂ group, and 15.16 mm Hg for the PFC plus 100% O₂ group. The differences in the PO₂ values among the four groups were statistically significant (ANOVA, \( p < 0.05 \)). When compared with the control group, the marked increases in the PFC plus 100% O₂ group were statistically significant at all measured time points (t-test, \( p < 0.05 \)). #p < 0.05 (t-test) compared with controls. Tx = treatment.

droplets by the reticuloendothelial system, the PFCs are excreted, predominantly via the lungs, although some small portion may be excreted in the bile and hence end up in the feces. Because of their low water solubility, PFCs first must be emulsified before injection, and problems caused by a synthetic emulsifier (Pluronic F-68) are one of the reasons for the failure of first-generation emulsions such as Fluosol. Since the late 1980s, improvements in both fluorocarbon and emulsion technology have led to the development of significantly more biocompatible, stable, and efficient second-generation injectable O₂ carriers. Oxygen is a second-generation sterile and concentrated 60% w/v PFC emulsion based on perfluorobutane (perfluorooctyl bromide). Minimal side effects, including mild flulike symptoms and transient thrombocytopenia insufficient to reduce bleeding, have been reported with Oxygen’s use, and these aspects require further study.

In normal conditions, the primary limiting factor for O₂ transport from Hb to tissue is the plasma, because of its low O₂ solubility. Most capillaries are perfused by a single column of RBCs (rouleaux), and the cell-free gaps probably contribute little to O₂ transport. By adding PFC droplets to the circulation, however, O₂ transport can take place over the entire capillary surface area, increasing overall O₂ delivery to the tissues. Padnick, et al., reported that even a single infusion of low-dose perfluorobutane emulsion (1 g PFC/kg body weight) while breathing room air increased brain tissue PO₂, 8.2 ± 18.5 mm Hg, and they posited that this increase reflected a PFC effect of lowering plasma resistance to O₂ transport. In our study, compared with the final brain tissue PO₂ of 1.86 mm Hg in the control group, even the group treated with PFC infusion alone showed an increase, with a final PO₂ of 5.14 mm Hg. This increase may have resulted from such a PFC effect of lowering plasma resistance.

It is well known that TBI can induce perivascular astrocytic foot process swelling, which in turn may reduce the cross-sectional area of capillaries, thus preventing normal RBC flow into these injured areas. In areas where RBCs cannot penetrate as a result of their size, PFCs may be able to deliver O₂ because of their small particle size (< 0.2 μm), thereby maintaining oxygenation through the cell-free plasma. The astrocytic foot process swelling may also increase the diffusion distance for O₂ from the microvasculature. Considering that the transport of O₂ to the tissues and finally to the mitochondria occurs by diffusion along the O₂ pressure gradient, these increases in the diffusion distance may therefore require higher tissue PO₂, as the driving force to deliver more O₂ to cells. In an experiment using a low-dose PFC emulsion, despite small increases in the O₂-carrying capacity of the blood—for example, O₂ content—after PFC infusion, the brain usually exhibited an increase in tissue oxygenation that was much greater than expected. Such enhancement of tissue PO₂ probably results from something more than simply a change in the O₂-carrying capacity of the blood. Therefore, it seems likely that it is the increased PO₂ rather than the increased O₂ content that may, in manner not yet understood, affect events at the mitochondrial or intracellular level, which may alter either oxidative phosphorylation activity or activity of enzymes within the Krebs cycle, inside mitochondria.

**Monitoring Brain Tissue PO₂**

In a clinical study by van den Brink, et al., early brain tissue hypoxia was observed in more than 50% of severely head injured patients, despite aggressive treatment for intracranial pressure and cerebral perfusion pressure, and the depth and duration of brain tissue hypoxia was related to outcome and proved to be an independent predictor of unfavorable outcome and death. Thus, continuous measurement of brain oxygenation by an intraparenchymally placed Clark-type catheter is a technically reliable, clinically applicable technique in patients at risk. For monitoring of global cerebral oxygenation status, it is recommended that the monitoring probe be inserted into undamaged brain tissue. If the probe is placed in strategic areas of the brain where regional ischemia may be present or likely, however, we can take advantage of the local nature of brain tissue PO₂ monitoring techniques instead. In our study, we placed the probe in endangered areas where focal ischemia may be present or likely, however, we can take advantage of the local nature of brain tissue PO₂ monitoring techniques instead. In our study, we placed the probe in endangered areas where focal ischemia may be present or likely, however, we can take advantage of the local nature of brain tissue PO₂ monitoring techniques instead.
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widely, with a range of 5 to 15 mm Hg; most authors propose 10 mm Hg as the hypoxic threshold.\(^\text{10,12,22,32,52}\) In our study, only treatment with both PFC emulsion and 100% O\(_2\) administration elevated the brain tissue PO\(_2\) level above this so-called critical threshold. In addition, the final infarct volume in the group treated with both PFC emulsion and 100% O\(_2\) (95.9 \(\pm\) 12.8 mm\(^3\)) compared with that in the control group (162.4 \(\pm\) 7.6 mm\(^3\)) was reduced 41% (p = 0.002). This reduction in ischemic damage is very likely to result from the improvement in brain oxygenation.

Conclusions

In summary, results of the current study have demonstrated that combined PFC and 100% O\(_2\) therapy has significant beneficial effects in improving brain tissue oxygenation and reducing ischemic damage in a model of acute SDH in rats. The PFC emulsion combined with 100% O\(_2\) treatment, which can be initiated even in the prehospital or emergency room setting, could be a potential therapeutic option for patients with severe TBIs.

Acknowledgments

We thank Drs. P. E. Keipert and S. Faithfull, Allianse Pharmaceutical Corporation, for comments on this manuscript.

References


33. Miller JD, Bullock R, Graham DI, Chen MH, Teasdale GM: Isch-

Manuscript received May 13, 2004. Accepted in final form May 26, 2005. These studies were funded by National Institutes of Health Grant No. NS12587. Oxygent was provided, free of charge, by Alliance Pharmaceutical Corp., but no financial support was given by the company for these studies. Address reprint requests to: M. Ross Bullock, M.D., Ph.D., Department of Neurosurgery, Virginia Commonwealth University Health System, P.O. Box 980631, Richmond, Virginia 23290-0631. email: robulloc@hsc.vcu.edu.