Blood Flow After Retinal Ischemia in Cats

Steven Roth and Zbigniew Pietrzyk

Purpose. To determine the changes in blood flow in the cat retina after 1 hour of ischemia.

Methods. Blood flow in the retina and choroid of adult cats anesthetized with chloralose, acepromazine, and halothane was measured using sequential injections of radioactively labeled microspheres. Ischemia was induced by elevation of intraocular pressure above systolic arterial pressure. Measurements were carried out before (baseline) and during ischemia, and at 5, 10, 15, 60, 120, and 240 minutes after the return of ocular circulation. In another two series of cats, blood flow was measured at comparable time periods without ischemia (controls). Arterial blood gas tension, systemic arterial pressure, hematocrit, and anesthetic level were controlled in each experiment.

Results. Retinal blood flow was decreased to 6%, and choroidal blood flow to 0.6%, of baseline value during ischemia. Within 5 minutes of the return of ocular circulation, retinal blood flow was approximately 200% of baseline, and choroidal blood flow was 108% of baseline. Blood flow 1 hour after the return of ocular circulation was not significantly different from baseline. There was no late decrease in blood flow after the ischemic period.

Conclusion. As does cerebral ischemia, retinal ischemia results in a hyperemic response but no delayed hypoperfusion. The mechanism of this effect is unknown. Invest Ophthalmol Vis Sci. 1994;35:3209-3217.

Retinal ischemia commonly occurs as a result of primary ocular disease, such as retinal vascular occlusion, or as a consequence of systemic disease, such as diabetes mellitus.1 Blindness may result if prompt treatment is not initiated. Studies of ischemia in both the cerebral cortex and the retina suggest the involvement of excitatory amino acids,2-5 which produce secondary damage from the "ischemic cascade."6 In the cerebral cortex, this sequence of events is accompanied by increases in blood flow soon after cerebral ischemia and recirculation and decreases later.6,7 No previous studies have examined changes in ocular blood flow after ischemia and recirculation. The changes have potentially important consequences for visual outcome. Increases in blood flow may lead to edema, and hypoperfusion may result in further cell death. We hypothesized that retinal blood flow after ischemia would be hyperemic first and hypoperfused later, similar to the flow patterns after cerebral ischemia.

METHODS

Procedures were approved by our Animal Care Committee and conformed to the ARVO Statement for Use of Animals in Ophthalmic and Vision Research.

Surgical Preparation

Anesthesia was induced in adult cats with halothane and oxygen in a closed box, followed by ventilation by mask. Pancuronium (Gensia Laboratories, Irvine, CA) 0.3 mg/kg was administered intravenously after the animal was unconscious, and it was continued at a constant infusion rate of 0.2 mg/kg per hour. Halothane (Fluothane, Ayerst Laboratories, New York, NY) concentration was adjusted during surgical preparation in response to changes in heart rate and blood pressure. After intubation of the trachea, the lungs were mechanically ventilated using an anesthesia circle absorber system equipped with a pediatric descending bellows anesthesia ventilator (Air-Shields, Hatboro, PA). Arterial PO2 was maintained at 100 to 200 mm
Hg with a 25% to 30% oxygen–air mixture, whereas arterial PCO\(_2\) was maintained at normocarbia (28 to 30 mm Hg). Intraocular pressure (IOP) was measured using a 21-gauge, ¾-inch butterfly needle inserted under direct vision into the anterior chambers of both eyes. Before placement of the needles, corneal analgesia was achieved using 0.5% proparacaine HC1 (Alcon, Humacao, Puerto Rico). Pupils were dilated with 0.5% tropicamide (Alcon, Humacao) and cyclopleydril (0.2% cyclopentolate HCl and 1% phenylephrine HCl, Alcon, Fort Worth, TX).

Catheters were placed through cutdowns into bilateral femoral arteries and veins. One of the arterial catheter tips was advanced into the abdominal aorta. A left atrial catheter was placed through a left thoracotomy. The following were continually measured and recorded: arterial pressure (systolic and diastolic in the femoral artery), left atrial pressure, IOP, end-tidal carbon dioxide concentration, and end-tidal halothane concentration. Before determination of blood flow, arterial blood gas tension (pH, P\(_{CO2}\), P\(_{O2}\)) was measured using an ABL II blood gas analyzer (Radiometer, Copenhagen, Denmark). Pressures (referenced to the level of the right atrium with the animal supine, except for IOP, which was referenced at eye level) were measured with P23 transducers (Spectramed, Oxnard, CA), and values were continually displayed and recorded on a chart recorder (2800 S recorder, Gould, Cleveland, OH). Mean arterial pressure (MAP) was calculated from the arterial pressure tracing as MAP = \(\frac{1}{3}\) diastolic + \(\frac{1}{3}\) systolic pressure. Retinal perfusion pressure (RPP) was calculated as MAP – IOP.

Body temperature, measured with an esophageal probe, was maintained at 37°C using a heating blanket (probe, was maintained at 37°C using a heating blanket). Halothane concentration was decreased to 0.2% expired, and chloralose 80 mg/kg and acepromazine 0.1 to 0.3 mg/kg were administered intravenously and repeated in smaller doses to maintain heart rate and blood pressure within 10% to 20% of the values at the end of surgical preparation. Maintenance fluids were provided with normal saline.

### Blood Flow Measurement

Blood flow was measured using radioactive microspheres (DuPont NEN, Wilmington, DE) 15.5 ± 0.1 \(\mu\)m diameter, labeled with Ce 141, Sn 133, Nb 95, Ru 103, Sc 46, or Cr 51, as we described previously. Each time blood flow was measured, 1.75 million microspheres were injected into the left atrium, and a reference blood sample was withdrawn from the abdominal aortic catheter at a rate of 1.36 ml/min using a syringe withdrawal pump (model 22, Harvard Apparatus, Dover, MA). Tissues of interest were removed after the experiment, and their radioactivity levels were counted with a gamma counter (Minaxi Autogamma 5000 series, Packard, Downers Grove, IL). Counts in both retinas and the choroids were combined, as we have described. Average blood flow was measured in the retina, choroid, and cerebral hemispheres (including gray and white matter). The linear matrix method and standard formulae were used to convert raw counts into blood flow in ml/100 g per minute.

### Ischemia Experiments

Baseline blood flow was measured, and ocular ischemia was induced for 1 hour as previously described. The needle in the anterior chamber was connected via a three-way stopcock to a pressure transducer and to an elevated saline reservoir. Intraocular pressure was increased in both eyes to a value 75 to 100 mm Hg above systolic arterial pressure. In initial experiments, we confirmed blanching of retinal vessels during ischemia upon ophthalmoscopic examination. After 1 hour of ischemia, the saline reservoir was lowered to restore IOP to preischemic levels. In all cases, IOP returned to the preischemic baseline within 3 minutes. Four experiments were performed (Table 1). Experiment 1 examined blood flow at baseline (before ischemia), and at 15, 60, 120, and 240 minutes after normal IOP was restored. Experiment 2 examined blood flow at baseline, at 30 minutes of ischemia, and at 5, 10, and 15 minutes after normal IOP was restored. To serve as controls for experiments 1 and 2, animals in experiments 3 and 4 were not rendered ischemic. Blood flow was measured at times corresponding to those in the ischemia experiments.

After completion of the experiments, animals were killed by injection of KCl and sodium thiomyal into the left atrial catheter. Eye and brain (cerebral cortical) tissues were removed and analyzed as previously described.

### Data Analysis

Data were analyzed using SYSTAT v 5.2 (SYSTAT, Evanston, IL). In all instances, \(P < .05\) was considered statistically significant, and all values are reported as mean ± SEM. Blood flow was compared over time in each of the four experiments using repeated measures analysis of variance followed by Student’s paired \(t\)-tests. The major comparison of interest was whether a significant change versus baseline (preischemia) could be detected. Next, we calculated the mean change from baseline to the succeeding four data points in each of the four experiments. To account for the effect of time and repeated injection of microspheres on blood flow, we subtracted change from baseline to a
given data point in the controls (experiments 3 and 4) from the corresponding changes from baseline in the ischemic experiments (experiments 1 and 2, respectively). The mean changes obtained represent the corrected mean change from baseline for both retinal blood flow (RetBF) and choroidal blood flow (CoBF). (Cerebral blood flow [CBF] values were not corrected for control). The SEM for these values was calculated as the square root of the sum of the squares of SEM for mean change from baseline of experiment and control values, respectively. The corrected mean change from baseline values was compared for significance versus 0 (i.e., representing a significant change from baseline after correction) using t-tests. Systemic hemodynamic values, such as MAP and arterial blood gas tension, as well as CBF were compared to baseline also using repeated measures analysis of variance and paired t-tests.

RESULTS
Systemic Hemodynamic Measurements
Because at nearly all time points in the four experiments arterial blood pH, PO2, PCO2, hematocrit, and CBF did not change significantly from the baseline measurement, hemodynamic data are only presented for experiment 1 (Table 2). Although PO2 was relatively high (160 to 170 mm Hg) in our experiments, all values were within range of 100% O2 saturation of hemoglobin, and blood flow values we obtained (see below) were similar to those in our previous studies and to those of others.8,9

Retinal perfusion pressure significantly increased after ischemia ended in experiments 1 and 2. This increase was accounted for primarily by a trend toward increased MAP. In experiment 2, as a result of the large induced increase in IOP, RPP was −110 ± 7 mm Hg during ischemia, which was significantly different than the baseline value (69 ± 8 mm Hg). In the control experiments (experiments 3 and 4), there were no significant changes in RPP.

Cerebral blood flow generally remained constant during these experiments. Only in experiment 4 was a significant change detected from baseline. Cerebral blood flow was 25.6 ± 1.7 ml/100 g per minute at the last measurement versus 30.3 ± 1.9 ml/100 g per minute at baseline.

Retinal Blood Flow
Retinal blood flow increased significantly (200%) 15 minutes after ischemia ended, and, compared to baseline in experiment 1, the values are 17.3 ± 8.6 ml/100 g per minute at the last measurement versus 17.3 ± 8.6 ml/100 g per minute.

### TABLE 2. Mean Arterial Blood Pressure, Retinal Perfusion Pressure, Arterial Blood Gas Tension, Hematocrit, and Cerebral Blood Flow

<table>
<thead>
<tr>
<th>Measurement Period</th>
<th>Baseline</th>
<th>15</th>
<th>60</th>
<th>120</th>
<th>240</th>
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</thead>
<tbody>
<tr>
<td>MAP (mm Hg)</td>
<td>110 ± 6</td>
<td>113 ± 7</td>
<td>122 ± 8</td>
<td>124 ± 10</td>
<td>98 ± 13</td>
</tr>
<tr>
<td>RPP (mm Hg)</td>
<td>90 ± 7</td>
<td>97 ± 7</td>
<td>111 ± 8†</td>
<td>111 ± 10*</td>
<td>85 ± 11</td>
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<tr>
<td>pH</td>
<td>7.35 ± .01</td>
<td>7.33 ± .01</td>
<td>7.32 ± .01</td>
<td>7.33 ± .01</td>
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</tr>
<tr>
<td>PCO2 (mm Hg)</td>
<td>30.3 ± 0.8</td>
<td>30.7 ± 0.6</td>
<td>28.9 ± 0.5</td>
<td>29.6 ± 0.8</td>
<td>30.9 ± 0.6‡</td>
</tr>
<tr>
<td>PO2 (mm Hg)</td>
<td>177 ± 5</td>
<td>165 ± 8</td>
<td>166 ± 7</td>
<td>175 ± 5</td>
<td>164 ± 5</td>
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<tr>
<td>Hct (%)</td>
<td>27 ± 1</td>
<td>28 ± 2</td>
<td>28 ± 1</td>
<td>27 ± 1</td>
<td>27 ± 1</td>
</tr>
<tr>
<td>CBF (ml per 100 g/min)</td>
<td>28.4 ± 3.3</td>
<td>29.2 ± 2.3</td>
<td>29.8 ± 1.9</td>
<td>31.5 ± 2.6</td>
<td>34.8 ± 1.4</td>
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Experiment 1: n = 6 cats. Data are shown as mean ± SEM.
MAP = Mean arterial pressure; RPP = retinal perfusion pressure (MAP−IOP); PCO2 = arterial carbon dioxide tension; PO2 = arterial oxygen tension; Hct = hematocrit; CBF = cerebral blood flow. Statistical significance: *P < .05 versus baseline; †P < .05 versus 15 minutes; ‡P < .05 versus 60 minutes; §P < .05 versus 120 minutes.
FIGURE 1. Retinal blood flow (RetBF, ml/100 g per minute) in experiments 1 and 3 (A) and 2 and 4 (B). Data are displayed as mean ± SEM, and number of animals per experiment is indicated in Table 1. Dotted line (RetBF control) represents control (no ischemia) experiments (3 and 4), whereas solid line (RetBF exptl) represents ischemia experiments 1 and 2. On the x-axis, postischemia refers to time after the end of 1 hour of ischemia. *Significant difference from baseline.

FIGURE 2. (A) Change in retinal blood flow (Δ RetBF, ml/100 g per minute) and (B) choroidal blood flow (Δ CoBF, ml/100 g per minute) from baseline, or before ischemia, to 240 minutes after the end of the ischemic period and the restoration of normal IOP. RetBF or CoBF at each time point was corrected for control (nonischemia, experiments 3 and 4). Blood flow values from ischemia and 5, 10, and 15 minutes after ischemia (35, 70, 75, 80 minutes on x-axis) are derived from experiments 2 and 4, whereas values at 60, 120, and 240 minutes after ischemia (125, 185, and 305 minutes on x-axis) are derived from experiments 1 and 3. Data are displayed as mean ± SEM. On the x-axis, postischemia refers to time after the end of 1 hour of ischemia. *Significant difference from 0 (solid line).

At this point, RetBF was 29.4 ± 5.4 ml/100 g per minute compared to the baseline value of 18.6 ± 4.9 ml/100 g per minute (P < .042). RetBF increased again at the last measurement (corresponding to 240 minutes after ischemia ended in experiment 1) to 36.9 ± 7 ml/100 g per minute, without achieving statistical significance. In experiment 2, we measured RetBF within 5 to 15 minutes after IOP was restored to normal as well as during ischemia (Fig. 1B, solid line). Compared to
baseline (16.2 ± 2.9 ml/100 g per minute), significant changes were found at each subsequent measurement: ischemia (1.1 ± 0.7 ml/100 g per minute, \(P < .002\)), 5 minutes after the end of the ischemic period (49.4 ± 10.5 ml/100 g per minute, \(P < .02\)), 10 minutes after the end of the ischemic period (45.0 ± 7.3 ml/100 g per minute, \(P < .005\)), and 15 minutes after the end of the ischemic period (45.9 ± 6.5 ml/100 g per minute, \(P < .003\)). During ischemia, RetBF was detectable at approximately 6% of baseline. At 15 minutes after ischemia ended, RetBF was increased 183% above baseline, nearly the same percent increase as that in experiment 1 at the same time period (Fig. 1A). The changes in RetBF in the corresponding control experiment (experiment 4) are also shown (dotted line) in Figure 1B. The overall trend was for RetBF to increase compared to baseline, but only the last measurement (corresponding to 15 minutes after ischemia ended in experiment 2) was significantly different from baseline (16.8 ± 2.1 ml/100 g per minute versus 12.1 ± 1.3 ml/100 g per minute at baseline, \(P < .005\)).

Figure 2A illustrates changes in RetBF relative to baseline when experiments 1 and 2 were corrected for their corresponding controls (experiments 3 and 4, respectively). Although two measurements of blood flow were obtained at 15 minutes after the end of ischemia, the value shown in Figure 2A was derived from experiments 1 and 3 (the same statistical significance was found for the 15-minute value derived from experiments 2 and 4). Corrected mean change from baseline values was compared to 0 (solid horizontal line). Values significantly different from 0 were: ischemia (-16.1 ± 2.7 ml/100 g per minute, \(P < .001\)), 5 minutes after the ischemic period ended (28.8 ± 10.1 ml/100 g per minute, \(P < .02\)), 10 minutes after the ischemic period ended (25.9 ± 6.5 ml/100 g per minute, \(P < .002\)), and 15 minutes after the ischemic period ended (25.1 ± 5.7 ml/100 g per minute, \(P < .001\)). There were no significant differences between these latter three data points.

**Choroidal Blood Flow**

Choroidal blood flow increased 15 minutes after ischemia (1551 ± 303 to 1814 ± 200 ml/100 g per minute, respectively, Fig. 3A, solid line), although the difference did not achieve statistical significance. There was a subsequent trend for decrease in CoBF, but only the final measurement, at 240 minutes after ischemia ended, was significantly different from baseline (521 ± 129 ml/100 g per minute, \(P < .01\), 34% of baseline). The changes in CoBF in the corresponding control experiment (experiment 3) are also shown in Figure 3A (dotted line). There was a downward trend in CoBF relative to baseline, with the second measurement (corresponding to 15 minutes after ischemia ended in experiment 1) significantly decreased (969 ± 187 versus 1428 ± 270 ml/100 g per minute, \(P < .027\)). The last two measurements (corresponding to 120 and 240 minutes after ischemia ended, respectively) were 685 ± 130 and 779 ± 152 ml/100 g per minute, but the differences from baseline did not achieve statistical significance.

In experiment 2, we examined CoBF within 5 to 15 minutes after IOP was restored to normal, as well as during ischemia (Fig. 3B, solid line). Compared to baseline (1482 ± 503 ml/100 g per minute), significant changes were found at the two subsequent measurements: ischemia (9.2 ± 6.9 ml/100 g per minute, \(P < .004\)) and 5 minutes after the end of the ischemic period (3083 ± 464 ml/100 g per minute, \(P < .001\)). Minimal CoBF was detectable during ischemia, approximately 0.6% of baseline (\(1/10\) the decrease in
This study demonstrates that a dramatic increase in DISCUSSION was found for the 15-minute value derived from experiments 1 and 3 (the same statistical significance is ischemia. In these experiments, the increase in CBF increased RetBF was maintained at 15 minutes after blood flow, as indicated by significant increases in hour of combined retinal and choroidal ischemia. We from baseline (0) were not significant, although there P < .014), and 15 minutes after (641 ± 134 ml/100 g per minute, P < .002). The last measurement was 37% of baseline.

Figure 2B illustrates changes in CoBF relative to baseline when experiments 1 and 2 were corrected for their corresponding controls (experiments 3 and 4, respectively). Although two measurements of blood flow were obtained at 15 minutes after the end of ischemia, the value shown in Figure 2B is derived from experiments 1 and 3 (the same statistical significance was found for the 15-minute value derived from experiments 2 and 4). All values were compared to 0 (solid horizontal line). Values significantly different from 0 were: ischemia (−991 ± 340 ml/100 g per minute, P < .02), 5 minutes after the end of ischemia (2311 ± 317 ml/100 g per minute, P < .001), 10 minutes after (1051 ± 333 ml/100 g per minute, P < .01), and 15 minutes after ischemia ended (1038 ± 311 ml/100 g per minute, P < .01). The subsequent changes from baseline (0) were not significant, although there was a trend for a decrease over time.

**DISCUSSION**

This study demonstrates that a dramatic increase in blood flow (hyperemia) is seen in the retina after 1 hour of combined retinal and choroidal ischemia. We did not evaluate the precise onset of this phenomenon, but it took place soon after the restoration of blood flow, as indicated by significant increases in RetBF 5 minutes after IOP returned to normal. Increased RetBF was maintained at 15 minutes after normal IOP was restored. By 1 hour, RetBF had returned to nearly the baseline (preischemic) level. The effects of ischemia on CoBF were similar. In neither case, however, was there evidence of hypoperfusion after the restoration of normal IOP.

Hyperemia after cerebral ischemia has been reported in animal models of both focal 

TABLE 1

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<th>CoBF</th>
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ability and damage to cerebral blood vessel endothelium. At present, the relationship between visual outcome and hyperemia after retinal ischemia is not known.

We used a model of retinal ischemia in which IOP was elevated to a value exceeding systolic arterial blood pressure by approximately 75 to 100 mm Hg and is presumed to result in cessation of blood flow in the retinal and choroidal circulations. Our model does not completely resemble the usual clinical situation in patients, that is, ischemia in retinal and choroidal circulation does not ordinarily occur simultaneously. Nonetheless, our model for retinal ischemia is easily produced, reproducible, and reversible. It allowed us to produce ischemia for 60 minutes, a period known to produce electrical and histopathologic evidence of ischemia. The precise degree of decrease in induced blood flow had not been previously determined. We confirmed in experiment 2 the nearly complete cessation of blood flow in the retina and choroid at 30 minutes of ischemia. The persistence of some blood supply, in fact, resembled the clinical situation in which flow does not usually stop completely.

It is of interest that RetBF during ischemia was 6% of the value before ischemia, whereas CoBF was approximately 0.6% of the value before ischemia. There are several possible explanations for and implications of these findings. One is that the effect of elevated IOP is clearly transmitted to a greater extent in choroidal than in retinal circulation. Even at high IOP values, blood flow may not be completely obliterated because of incomplete transmission of elevated pressures to the retinal and choroidal circulations. Support for such a hypothesis comes from two sources. There is histopathologic evidence of heterogeneous injury in the retina after ischemia in rabbits by elevation of IOP. A homogeneous distribution of injury suggests a more uniform transmission of elevated external pressure. Results similar to ours were obtained in a pressure-induced ischemia model in the brain. Cerebral ischemia was produced in cats by infusing artificial cerebrospinal fluid into the cerebral ventricular system to elevate intracranial pressure above systolic arterial pressure. Persistence of cerebral blood flow was detected, the degree of which varied according to region of the brain examined. The period of ischemia was far shorter than ours (reflecting the lower tolerance of the brain to ischemia than the retina). During a longer ischemic period, elevated intracranial pressure may have been further transmitted to other brain regions. We only measured RetBF and CoBF at one point during ischemia; we do not know if the degree of ischemia varied during the 1-hour period.

There are limitations to this study related to the use of radioactive microspheres. Microspheres are best used for blood flow measurements during
steady-state conditions. It is possible that conditions were not steady state in the immediate postischemic measurements of experiment 2. That the RetBF measurements obtained at 5, 10, and 15 minutes after the end of the ischemic period were not significantly different from one another suggests steady-state conditions may have been present. It has been shown that after injecting microspheres into the left atrium for 20 seconds, as we did, 98% of the spheres collected through the femoral arterial catheter are collected within the first minute.19 During or immediately after injection of microspheres, an audible signal was detected with a Doppler probe over pial arteries.13 We presume rapid distribution in the retina would be similar. Although microspheres may not be ideal for measuring RetBF and CoBF changes in the first minute after ischemia, we think the time course for distribution is rapid enough to measure changes for the next 15 minutes and more.

Another limitation related to number of microspheres impacted in the retina and in other tissues with relatively low blood flow has been discussed previously by us9 and by others.20,21 This limitation explains why 1.75 million spheres were used per injection. Ideally, impaction of 400 microspheres in the tissues is desirable, but accurate measurements are still possible in experiments when 100 to 200 microspheres are impacted per injection.8,19 Increasing the number of microspheres injected to obtain greater degree of impaction has been shown to result in significant hemodynamic effects and alterations in regional blood flow in cats.22 We combined counts from both eyes (with eyes from each animal treated similarly) to ensure greater accuracy in our data, as described previously.8,9 An alternative might have been to use one eye in each animal as an experimental eye and the other as a control; this would have conserved animals, but our data might have been less accurate.

When we observed decreases in blood flow in the choroid and increases in the retina at later time periods in ischemia experiments, we speculated that repeated microsphere impaction altered blood flow over time in the retina and choroid. Another possible explanation, a change in systemic hemodynamics, was eliminated by the careful control of arterial blood pressure and retinal perfusion pressure, hematocrit, and arterial CO2 tension, all demonstrated to alter RetBF and CoBF.8,5,23 We also maintained a constant anesthetic state during measurements. No significant changes were detected in CBF during the experiment. Taken together, these findings imply that the alterations in RetBF and CoBF found in our experiments were not due to a deterioration of the experimental preparation.

Because impaction of microspheres can alter regional blood flow in cats,24 we performed control experiments 3 and 4 in nonischemic cats with the same number of microspheres injected at time points comparable to those in ischemic experiments 1 and 2, respectively. In the control experiments, we determined the effects of repeated microsphere injection on RetBF and CoBF: CoBF showed a downward trend, and RetBF exhibited a trend toward increased blood flow. These changes also could not be accounted for by alterations in systemic hemodynamics or variations in anesthetic depth. Similarly, CBF did not vary significantly from baseline, except for an alteration at the last measurement in experiment 4. It is unlikely that the changes represent a “time effect” because CoBF decreased over the much shorter time period used in experiment 4 compared to experiment 3. Thus, we conclude that the repeated impaction of 1.75 million microspheres decreases delivery of microspheres upon subsequent injection, resulting in lower blood flow over time in the choroid, similar to the results obtained for the carotid body in cats.22,25 The increase in RetBF with repeated microsphere impaction was unexpected, but this result was also replicated in experiments 3 and 4. These changes in measured blood flow could potentially have been avoided by measuring blood flow only one or two times per animal and using multiple sets of animals to produce data at each time point of interest. We elected not to choose this method because of the significantly greater expense and the disadvantage of being unable to compare subsequent blood flow to baseline in each cat, which, in turn, would likely have resulted in increased variability in blood flow.

We can only speculate about possible mechanisms for alterations in blood flow with repeated injections of microspheres. One possibility is release of vasodilatory substances upon choroidal vessel occlusion that fail to dilate an occluded choroidal circulation but produce vasodilatory effects upon RetBF. Alternatively, the increase may be caused by the purely mechanical shunting of blood to the retinal circulation when the choroidal circulation is occluded. Whatever the mechanism, the use of repeated microsphere injections (greater than two) to determine CoBF and RetBF requires concomitant control experiments. Thus, caution is in order when using multiple injections of microspheres because even with corrections for control, the results obtained may be more qualitatively than quantitatively accurate.

Regardless of the mechanisms involved in the changes in blood flow with repeated injections, with correction for control experiments, it is apparent that both RetBF and CoBF increased significantly from baseline at 5, 10, and 15 minutes after ischemia ended. It is of note that although there was a tendency for RetBF and CoBF to decrease over time after correction for control (Fig. 2), there was no hypoperfusion.
In models of global or focal cerebral ischemia, hypoperfusion occurred between 30 minutes and 6 hours after reperfusion. The mechanism of hypoperfusion may be related to leukocyte adhesion or to alterations in vasomotor tone, particularly with alterations in the endothelium or in its response to vasoactive substances. Possibly, this phenomenon occurs later after retinal ischemia, and we did not measure late enough after reperfusion to detect its onset. Another possibility is a difference in the effect of ischemia on the retina if endothelium is preserved or if there is less leukocyte adhesion. Although leukocyte alterations have been shown after retinal ischemia in a preliminary study, further investigation is required to determine their precise nature.

We did not address the mechanism of postischemic hyperemia, whose origin after cerebral ischemia remains controversial. Cerebral and retinal ischemia result in the activation of excitatory amino acids and the release of vasoactive substances, including prostaglandins, adenosine, and free-radical species. An increase in local adenosine concentration was found after cerebral ischemia in rats, and adenosine is known to produce vasodilatation in the cerebral vasculature. Others have shown that decreasing \(O_2\) free-radical formation did not affect hyperemia after cerebral ischemia in cats. In contrast, inhibition of free-radical formation did not affect hyperemia after cerebral ischemia in rats. In models of global or focal cerebral ischemia, hypoperfusion occurred between 30 minutes and 6 hours after reperfusion. The mechanism of hypoperfusion may be related to leukocyte adhesion or to alterations in vasomotor tone, particularly with alterations in the endothelium or in its response to vasoactive substances. Possibly, this phenomenon occurs later after retinal ischemia, and we did not measure late enough after reperfusion to detect its onset. Another possibility is a difference in the effect of ischemia on the retina if endothelium is preserved or if there is less leukocyte adhesion. Although leukocyte alterations have been shown after retinal ischemia in a preliminary study, further investigation is required to determine their precise nature.

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Arterial \(PO_2\) was relatively high in these experiments (160 to 170 mm Hg) to ensure adequate oxygenation during the lengthy open chest preparation. Although this may have resulted in some vasoconstriction in the retina, it is unlikely that it significantly altered our results because only insignificantly larger amounts of dissolved \(O_2\) would have been present compared to \(PO_2\) values of 80 to 100 mm Hg. Furthermore, arterial \(PO_2\) was maintained at a constant level throughout the experiments. Therefore, any effect it might have had would have applied to all our measurements.

In conclusion, we found significant hyperemia in the retina in cats after 1 hour of ischemia. The mechanisms of this effect and its significance for ultimate visual outcome are issues that remain to be investigated.

**Key Words**

blood flow, ischemia, retina, choroid, hyperemia

**References**


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