Oxygen distribution and consumption in the macaque retina

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Birol G, Wang S, Budzynski E, Wangsa-Wirawan ND, Linsenmeier RA. Oxygen distribution and consumption in the macaque retina. Am J Physiol Heart Circ Physiol 293: H1696–H1704, 2007. The oxygen distribution in the retina of six anesthetized macaques was investigated as a model for retinal oxygenation in the human retina and adjacent to the fovea. PO2 was measured as a function of retinal depth under normal physiological conditions in light and dark adaptation with O2 microelectrodes. Oxygen consumption (QO2) of the photoreceptors was extracted by fitting a steady-state diffusion model to PO2 measurements. In the perifovea, the PO2 was 48 ± 13 mmHg (mean and SD) at the choroid and fell to a minimum of 3.8 ± 1.9 mmHg around the photoreceptor inner segments in dark adaptation, rising again toward the inner retina. The PO2 in the inner half of the retina in darkness was 17.9 ± 7.8 mmHg. When averaged over the outer retina, photoreceptor QO2 (called Qav) was 4.6 ± 2.3 ml O2/100 g−1min−1 under dark-adapted conditions. Illumination sufficient to saturate the rods reduced Qav to 72 ± 11% of the dark-adapted value. Both perifoveal and foveal photoreceptors received most of their O2 from the choroidal circulation. While foveal photoreceptors have more mitochondria, the QO2 of photoreceptors in the fovea was 68% of that in the perifovea. Oxygenation in macaque retina was similar to that previously found in cats and other mammals, reinforcing the relevance of nonprimate animal models for the study of retinal oxygenation, but there was a smaller reduction in QO2 with light than that previously found in cats and other mammals, reinforcing the relevance of nonprimate animal models for the study of retinal oxygenation, but there was a smaller reduction in QO2 with light than in the primate fovea: a region of very high cone density and a foveal pit. In this region, which is 0.5 mm in diameter, the retinal circulation is absent and the retina is thinner because second- and third-order neurons are displaced laterally. The presence of a fovea means that oxygenation of primate retina is certain to be somewhat different from that of the more well-studied animals.

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our knowledge of primate retinal oxygenation. First, we investigated differences in photoreceptor QO2 between light and darkness, which our earlier study (1) had suggested were smaller in monkeys than in cats. Second, we explored differences in oxygenation between the foveal and perifoveal retina. Finally, we wanted to compare several aspects of inner and outer retinal oxygenation in monkeys and cats in order to determine the range over which the more extensive data from cats and other mammals can be used to predict human retinal oxygenation.

METHODS

Animal Preparation and Recording

In these experiments, we adhered to the American Physiological Society’s “Guiding Principles in the Care and Use of Animals” and obtained approval from our Institutional Animal Care and Use Committee. Six adult macaques [4 Macaca fascicularis (cynomolgus) and 2 Macaca mulatta (rhesus)] weighing 4–11 kg were used. The animals were from 11 to >25 yr old and had been used previously for skin tests of allergens. The animals were all in good health when the experiments described here were performed. Monkeys were initially anesthetized with intramuscular ketamine (100–200 mg), followed by intravenous pentothal sodium (5%) as needed during surgery. Two veins and a femoral artery were cannulated, and a tracheal tube was inserted. The head was immobilized in a head holder. A gas-permeable contact lens (MetroOptics, Austin, TX) was used to protect the cornea. The pupil was dilated with topical 1% phenylephrine and 1% atropine. Flurbiprofen sodium ophthalmic solution (0.03%) was also administered topically to block traumatic pupillary constriction mediated by prostaglandins.

Surgery was performed on the right orbit and eye to attach an eye ring and allow introduction of a 15-gauge guide needle at the pars plana to carry the O2 microelectrode (see, e.g., Refs. 25, 30). The electrode was connected to the needle with a boot system that allowed movement but prevented leakage of vitreous humor. After surgery, a mixture of isoflurane (1–2%) and N2O:O2 (0.70:0.30 to 0.75:0.25) was continuously administered. Paralysis with pancuronium bromide (loading dose of 0.5 mg/kg followed by a maintenance dose of 0.2 mg·kg\(^{-1}\)·h\(^{-1}\)) prevented eye movements and facilitated control of blood gases via adjustments of the ventilator. Arterial blood was examined periodically with a blood gas analyzer (model 860, Bayer Diagnostics, New York, NY) to monitor arterial partial pressure of O2 \((PaO_2)\), arterial partial pressure of CO2 \((PaCO_2)\), pH, and blood glucose concentration \((\text{glucose})\), which were maintained at \(PaO_2 = 109.4 \pm 7.40 \text{ mmHg}\), \(PaCO_2 = 40.4 \pm 2.6 \text{ mmHg}\), \(pH = 7.36 \pm 0.02\), and \(\text{glucose} = 122.9 \pm 8.3 \text{ mg/dl during the experiments.}\)

The animal’s body temperature was maintained at 39°C with a feedback-controlled heating blanket. Heart rate and arterial blood pressure were also monitored to evaluate the anesthesia.

Double-barreled PO2 microelectrodes were used to record the intraretinal PO2 according to procedures described by Linsenmeier and Braun (25). PO2 was recorded with a picomammeter (model 614, Keihlery, Cleveland, OH) followed by a buffer amplifier. The intraretinal electroretinogram (ERG) was recorded with the second barrel of the microelectrode in order to gauge the quality of the penetration and the depth of the electrode in the retina (24). The ERG was elicited by 2.5-s flashes of diffuse white light at or above rod saturation \([ \sim 8.6 \log \text{ quanta (555 nm)} \cdot \text{deg}^{-2} \cdot \text{s}^{-1}]\). The maximum illumination available was 11.5 log quanta \((555 \text{ nm}) \cdot \text{deg}^{-2} \cdot \text{s}^{-1}\). In addition to the microelectrode, two Ag-AgCl electrodes were also inserted into the vitreous through sealed 20-gauge needles. One served as the PO2 reference electrode, and the other recorded the vitreal ERG. The voltage barrel of the microelectrode and the vitreal ERG electrode were referenced to an Ag/AgCl scalp electrode, a chloridized silver plate behind the eye. The microelectrode and vitreal signals were recorded through DC coupled unity gain amplifiers (model M7070-A and model M4A, respectively, WPI, Sarasota, FL). After amplification, signals were displayed on a storage oscilloscope and sent to a chart recorder and a computer, which ran a MATLAB data acquisition program.

The microelectrode was positioned to record from the fovea, or the perifovea within \(\sim 2 \text{ mm}\) of the fovea, by the aid of a micromanipulator under visualization through a direct ophthalmoscope. In humans, the parafovea is defined to be a 0.5-mm-wide annular region outside the fovea, which is the central 1.5-mm diameter area, and the parafovea is defined to be a 1.5-mm-wide annulus outside of the parafovea (33). We could not precisely determine whether the electrode was in the parafovea or perifovea, so the term designating the larger area, perifovea, is used here. The microelectrode was advanced through the retina with a hydraulic microdrive (model 1207S, David Kopf Instruments, Tujunga, CA) until it crossed the retinal pigment epithelium (RPE), indicated by a DC shift in the voltage. The microelectrode was then withdrawn at a rate of 2 μm/s to record PO2 profiles from the choroid to the vitreous humor. Recordings were done during both dark and light adaptation. To record a profile during light adaptation, the retina was first penetrated during dark adaptation. Steady illumination with white light at or above rod saturation was initiated with the electrode deep in the retina and maintained for \(\sim 60 \text{ s}\) before the withdrawal was begun. This was sufficient time for the metabolism to completely adjust to the illumination (46). Eight to eighteen profiles were obtained in each animal.

It was assumed that profiles were recorded at a 45° angle to the retinal surface, and distances have been converted to a distance perpendicular to the retinal surface. The withdrawal distances were longer than penetration distances because of electrode drag, but the difference was not statistically significant. The perifoveal penetration distances were 279 ± 85 (in dark) and 300 ± 85 (in light) μm, which were similar to those obtained for cats (25). The foveal penetration distances were 230 ± 49 μm in dark adaptation and 267 ± 34 μm in light adaptation, which were slightly larger than in our previous study \((172 \pm 24 \mu m)\), but the relative difference in thickness between perifovea and fovea was similar to that reported earlier (1).

PO2 Data Analysis

The three layer one-dimensional O2 diffusion model previously used for cat retina (19, 25, 47) and macaque retina (1) was fitted to perifoveal profiles to determine photoreceptor O2 consumption \((QO2)\) per unit volume of tissue. This model represents the outer half of the retina by three layers, corresponding to the outer segments (layer 1), inner segments (layer 2), and outer nuclear layer (layer 3). Only the inner segments contain mitochondria, so \(QO2 = 0\) in layers 1 and 3. Fitting of the model to the data finds optimal values of \(QO2\) in layer 2 \((Q2)\) as well as the location of the boundaries of this layer \((\text{distances } x = L_1\) and \(L_2\) from the choroid, which is at \(x = 0\)). As in previous work, we report \(QO2\) averaged over the outer retina \((Q_{\text{av}})\), which is less affected by the details of the shape of individual profiles and has a lower coefficient of variation than \(Q2\): \(Q_{\text{av}} = Q2 \cdot (L_2 - L_1)/L_2\), where \(L\) is the total thickness of the outer, avascular perifoveal retina, which is usually 50% of the whole profile (19). \(Q_{\text{av}}\) was corrected for differences in the penetration and withdrawal distance (19) by multiplying \(Q_{\text{av}}\) by \((2L/Dp)^2\), where \(2L\) is the retinal thickness during withdrawal and \(Dp\) is the retinal thickness during penetration.

The same model was fitted to foveal profiles. In many of the foveal profiles there were peaks in the inner portion of the retina that indicated the presence of capillaries, so the modeled region was the outer \(\sim 60\%\) of the retina. In other cases the recordings did not contain such peaks within the retina, and recordings therefore appeared to be from the avascular foveal pit. The entire retina could be modeled in those cases. The identification of a profile as foveal was based on the characteristics of the ERG, as previously described (1). Total \(QO2\) per unit area of retinal surface, \(Q_{\text{tot}} = Q2(L_2 - L_1)\), was used to compare

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the fovea and perifovea, because the thickness of the photoreceptor layers is greater in the fovea and a measure based on consumption per unit volume would not have revealed whether the fovea or perifoveal photoreceptors used more O2.

Because the inner retina has a three-dimensional vasculature and the \( \text{PO}_2 \) profiles obtained were one-dimensional, the model was only used in the outer avascular part of the retina. For the inner retina, \( \text{PO}_2 \) values were averaged over the inner 50% of each profile. This average value is called \( P_{IR} \).

**Statistics**

Each of the parameters reported here was averaged across profiles within an animal before the average across macaques was computed, so that each macaque would be equally weighted. All values are reported as means \( \pm \) SD. Statistical significance was determined in most cases with a paired Student’s \( t \)-test and was defined as a \( P \) value \(<0.05.

**RESULTS**

**Perifoveal Retina**

**\( P_{O2} \) profiles.** Figure 1 shows examples of perifoveal \( P_{O2} \) profiles from one macaque. The shapes of the profiles were similar to those observed in cats. Under dark-adapted conditions (Fig. 1A), the \( P_{O2} \) started at 75 mmHg in the choroid (100% retinal depth) and decreased to 0 mmHg at 82% retinal depth. The \( P_{O2} \) increased again toward the inner retina. The shape of the profile indicated that \( O_2 \) was supplied to photoreceptors from both choroidal and retinal circulations. The average inner retinal \( P_{O2} \) (\( P_{IR} \)) was 24 mmHg in this example, and the \( P_{O2} \) at the retinal/vitreal boundary was 10 mmHg.

Under light-adapted conditions, the shape of the profile was similar, but there was a higher \( P_{O2} \) in the distal retina compared with the dark-adapted profile. The profile started with a choroidal \( P_{O2} \) of 72 mmHg and decreased to a minimum of 20 mmHg. The \( P_{O2} \) at the retinal/vitreal boundary was 25 mmHg (Fig. 1B).

**Choroidal and minimum \( P_{O2} \).** Figure 2A shows average choroidal \( P_{O2} \) (\( P_C \)), which reflects the choroidal \( O_2 \) supply, obtained from each profile and averaged within each macaque under dark- and light-adapted conditions. The data on primate
retina are so limited that in this figure and in Figs. 3, 5, and 7, we show values for each animal to illustrate the interanimal variability as well as light/dark differences. Across animals, $P_C$ was $48 \pm 13$ mmHg (mean and SD; $n = 6$ macaques) in darkness. It was unaffected by light adaptation ($P_C = 50 \pm 14$ mmHg; $n = 6$). Although $P_C$ was somewhat variable among animals (average values of 36–68 mmHg under dark- and 33–72 mmHg under light-adapted conditions), it was stable within each animal.

Figure 2B shows the average minimum $P_O2$ obtained from each profile and averaged over each macaque under dark- and light-adapted conditions. The minimum $P_O2$ always occurred in layer 2, the photoreceptor inner segments. In cynomolgus, it was $4.2 \pm 1.8$ mmHg ($n = 4$ animals) in dark adaptation and significantly higher ($12.6 \pm 1.8$ mmHg) in light adaptation ($P = 0.009$; $n = 4$). The minimum $P_O2$ was not different between dark and light adaptation in the two rhesus (macaques R1 and R2). The rhesus were older (at least 25 yr old) than the cynomolgus (average age of $\sim 12$ yr), so whether this is an age difference or a species difference is not clear. When all six animals were included in the analysis, the minimum was $3.8 \pm 1.9$ mmHg in dark adaptation and the difference between dark and light was not significant ($P = 0.068$; $n = 6$ macaques).

**Oxygen consumption.** Perifoveal outer retinal $Q_O2$ ($Q_{av}$) was obtained by fitting a three-layer one-dimensional diffusion model to the avascular region of the $O2$ profiles as described in METHODS. Figure 3 shows the mean and SD of $Q_{av}$ for each macaque under dark- and light-adapted conditions. The light-evoked changes shown in Fig. 3 are the maximum reductions in $Q_O2$ that can be produced by light. $Q_{av}$ was $4.6 \pm 2.3$ ml O$_2$/100 g tissue-min under dark-adapted conditions, and 72 $\pm$ 11% of the dark value under light adaptation. This difference was statistically significant ($P = 0.004$; $n = 6$ macaques). These results are in agreement with previous studies on light-evoked changes in $Q_O2$ in both macaques (1) and other vertebrates (e.g., Refs. 3, 21, 25, 38, 55), although as noted in DISCUSSION, the light-dark difference was smaller in macaques than in cats.

Both $P_C$ and $Q_{av}$ varied considerably across animals. In cats, these variables are strongly related, with $Q_{av}$ being limited by the $O2$ supply available from the choroidal circulation, which is the major $O2$ source for the photoreceptors (25, 30). To evaluate whether a similar relation exists in macaques, $Q_{av}$ was plotted against $P_C$ obtained from each profile, allowing us to treat each profile equally (Fig. 4). $Q_{av}$ was found to be linearly dependent on $P_C$, and the slope was significantly different from zero in both dark-adapted ($P < 0.01$; $n = 40$ profiles, $R^2 = 0.361$) and light-adapted ($P < 0.01$; $n = 27$ profiles, $R^2 = 0.646$) conditions. This dependence was even stronger when the regression was done on the average $Q_{av}$ and $P_C$ values from each macaque rather than treating each profile individually ($P < 0.05$; $n = 6$ macaques, $R^2 = 0.698$ in dark, $R^2 = 0.900$ in light). Thus much of the variance in $Q_O2$ in the outer retina was explained by variation in $P_C$. The rhesus did not appear to differ from the cynomolgus on these measures.

The $O2$ used by the photoreceptors is supplied by both the choroidal and retinal circulations. To quantify the $O2$ contribution from each source, the diffusion equations were used to compute the $O2$ fluxes into the consuming region from the choroidal supply and retinal supply as described elsewhere (25). The average percentage of $O2$ supplied by the retinal circulation in perifovea varied between 0 and 23% among macaques, averaging 15 $\pm$ 8% in dark adaptation. It varied between 0 and 22%, averaging 11 $\pm$ 8%, in light adaptation. Hence, the choroid was the main $O2$ source for the perifoveal photoreceptors, supplying 85% of the $O2$ in dark and 89% of the $O2$ in light.

**Average inner retinal $P_O2$.** Average inner retinal $P_O2$ ($P_{IR}$) was always lower than $P_C$ and was somewhat heterogeneous depending on the proximity of the microelectrode to retinal vessels (peaks in Fig. 1). Figure 5 shows $P_{IR}$ for each macaque. In cynomolgus, $P_{IR}$ was $20.4 \pm 7.6$ mmHg ($n = 4$ macaques) in dark adaptation and slightly increased to 24.9 $\pm$ 7.4 mmHg in light adaptation ($P = 0.013$; $n = 4$ macaques). In rhesus, the change in $P_{IR}$ was in the opposite direction, decreasing from 10.6 $\pm$ 4.6 mmHg in darkness to 5.8 $\pm$ 2.9 mmHg under light. When all animals were included, the...
grand average in the dark was 17.9 ± 7.8 mmHg, and the change in inner retinal P\textsubscript{O2} with light was not significant (P = 0.388; n = 6).

**Foveal Retina**

**P\textsubscript{O2} profiles.** We were able to record from the fovea in two cynomolgus [macaque C3 (dark-adapted conditions) and macaque C4 (dark- and light-adapted conditions)]. The foveal pit, where there is no inner retina and no retinal circulation, is <0.5 mm in diameter in humans and monkeys (10, 33, 40, 51). Just outside this region, in a 0.5-mm annulus, the retina gradually becomes thicker (33, 37) and has a rich retinal vasculature (40). This 1.5-mm-diameter region is all considered part of the fovea. It was difficult to keep the electrode confined to the avascular center of the fovea when penetrating the retina at an angle, and many of the profiles showed evidence of the retinal circulation that exists outside the central 0.5 mm. Figure 6 shows examples of foveal P\textsubscript{O2} profiles from macaque C4, which were similar in shape to the perifoveal profiles in both light and dark adaptation.

Because we appeared to be recording at the edge of the avascular region, we also calculated the contribution of the retinal circulation and found that 23 ± 8% (macaque C4) of the O\textsubscript{2} was supplied by the retinal circulation under both dark- and light-adapted conditions. Hence, the choroid was still the main O\textsubscript{2} source, supplying ~77% of the O\textsubscript{2} to the photoreceptors in the region where recordings were made.

**Perifoveal and foveal comparison.** In the center of the fovea, the only retinal layers present are photoreceptor layers. Thus the same diffusion model can be used for the fovea as for the perifovea, but the entire retina is included in modeling. As noted above, profiles at the edge of the fovea can include regions where the inner retina exists, and this vascularized region must be excluded from the fits. Outer retinal thickness for the foveal profiles (L) ranged between 60% and 100% of the foveal retina, depending on whether there was evidence of retinal circulation.

Figure 6 shows model fits to the foveal profiles. Q\textsubscript{tot} was calculated as described in METHODS to allow a fair comparison between Q\textsubscript{O2} of the foveal and perifoveal regions, since the inner segments are longer in the fovea (see, e.g., Refs. 20, 33). The parameter Q\textsubscript{tot} represents all the Q\textsubscript{O2} per unit volume of the photoreceptors (Q\textsubscript{av}). Q\textsubscript{tot} obtained from each profile was averaged over all profiles within each animal and is shown in Fig. 7 for dark and light adaptation for two macaques from this study (macaques C3 and C4) and for one macaque from our earlier study (macaque 93, M. nemestrina; Ref. 1). The earlier data were collected by exactly the same methods and are included because the unusually complicated and expensive nature of these experiments makes limited data available. The foveal Q\textsubscript{tot} decreased under light-adapted conditions, but by a smaller amount than the decrease observed in perifoveal Q\textsubscript{tot}. It is possible that in these recordings from cone-dominated regions, we did not have enough light to produce the maximum reduction in Q\textsubscript{O2}. The main finding was that in all three animals perifoveal Q\textsubscript{tot} was higher than foveal Q\textsubscript{tot}, and this was significant when both dark- and light-adapted Q\textsubscript{tot} were considered together (P = 0.0009; n = 5 pairs of perifoveal-foveal data, paired t-test), or when the dark-adapted data alone were considered (P = 0.022; n = 3 macaques). Averaged across all animals and conditions shown in Fig. 7, foveal Q\textsubscript{tot} was 68% of
perifoveal \( Q_{tot} \). Yu et al. (53) also found that foveal \( QO_2 \) was lower than perifoveal \( QO_2 \). The value of \( Q_2 \) itself, the \( QO_2 \) of the inner segments themselves per unit volume, was also always lower in the fovea than in the perifovea.

**DISCUSSION**

The aim of this study was to provide information on retinal oxygenation in macaques, because primate data provide a bridge from cats (and other species) to humans. The measurements and analysis used here for retinal oxygenation in macaques are identical to the methods used previously in cats, and they allow quantitative comparisons between the two species that have not been possible before.

**Perifoveal Retina**

Table 1 summarizes parameters of retinal oxygenation of the perifovea in macaques obtained in this study and the area centralis of cats obtained in previous work (8, 19, 25, 30). In all cases, these are values for normal physiological conditions, with blood gases in the normal range. The same diffusion model works equally well for the outer retina of both animals, and, in most respects, retinal oxygenation is very similar in the two species. First, in the outer retina both species have a relatively high \( P_{O_2} \) at the choroid and a low \( P_{O_2} \) in darkness in the inner segments, approaching zero in many animals. Thus, in darkness, cats and monkeys should be almost equally vulnerable to conditions that would tend to reduce availability of \( O_2 \) to the photoreceptors, such as hypoxemia or retinal detachment. As discussed below, \( QO_2 \) is reduced in both species when choroidal \( P_{O_2} \) decreases. However, they may also be equally well adapted to deal with this situation by increasing anaerobic glycolysis during hypoxia, which we know is true of cats (31) and other species (3, 14, 50). Second, photoreceptors in both species are supplied primarily by the choroidal circulation, with only 10–15% of the supply coming from the retinal circulation. Third, the absolute value of \( Qav \) is similar for cat area centralis and monkey perifovea in dark adaptation. On the basis of the regression of \( Qav \) on \( P_{C} \), one would expect \( Qav \) of \(-4.1 \text{ ml} \cdot 100 \text{ g}^{-1} \cdot \text{min}^{-1}\) in cat and \(4.9 \text{ ml} \cdot 100 \text{ g}^{-1} \cdot \text{min}^{-1} \) in monkey at \( P_{C} = 50 \text{ mmHg} \). Fourth, the \( P_{O_2} \) values in the inner half of the retina are also quite similar, 15–20 mmHg on average. The two rhesus had lower values than the cynomolgus, but the dark values in rhesus were within the distribution of \( P_{O_2} \) values observed in the normal cat inner retina (26). The lower inner retinal \( P_{O_2} \) values in the rhesus could be a result of impairments in vascular regulation with age, but a larger sample of animals of the same species would be needed to examine this point. As might be expected from the regulatory properties of the retinal circulation, the \( P_{O_2} \) values in the inner retina are similar to those in the brain (see e.g., Ref. 32).

A few properties need more explanation. In darkness, photoreceptor \( QO_2 \) is strongly dependent on choriocapillaris \( P_{O_2} \) \( (P_{C}) \) in both animals, and the slope of the relation is similar. (There was no evidence that \( QO_2 \) or the dependence of \( P_{O_2} \) on \( P_{C} \) was different in cynomolgus and rhesus.) This dependence occurs because the limiting factor in photoreceptor \( QO_2 \) is usually the flux of \( O_2 \) to the inner segments, which is largely controlled by the \( P_{O_2} \) at the choroidal boundary. In both species, there is little variation in \( P_{C} \) within an animal but substantial variation across animals. Variations in \( P_{C} \) from animal to animal are likely to result from variations in choroidal blood flow, which is autononically controlled (11). (Unfortunately, this assertion has never been tested directly, because choroidal blood flow is very difficult to assess simultaneously with intraretinal \( P_{O_2} \).) A lower blood flow leads to a higher choroidal \( O_2 \) extraction and therefore lower \( P_{C} \), but as \( P_{C} \) decreases the flux of \( O_2 \) to the photoreceptor is compromised, and \( QO_2 \) decreases (27). The dependence of \( QO_2 \) on \( P_{C} \) extends only to values of \( P_{C} \) in the normoxic and hypoxic ranges. Yu et al. (53) found that there was no dependence of \( QO_2 \) on \( P_{C} \) in the macaque retina when \( P_{O_2} \), was elevated above

### Table 1. Comparison of macaque and cat retinal oxygenation

<table>
<thead>
<tr>
<th>Characteristics of Oxygen Distribution and Consumption</th>
<th>Macaque Perifovea</th>
<th>Cat Area Centralis</th>
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</thead>
<tbody>
<tr>
<td>Minimum ( P_{O_2} ) in dark, mmHg</td>
<td>3.8±1.9</td>
<td>5.25±5.12 (25)</td>
</tr>
<tr>
<td>Choroidal ( P_{O_2} ), mmHg</td>
<td>48±13</td>
<td>54±11.7 (8), 41.2±16.2 (30)</td>
</tr>
<tr>
<td>Fraction of ( O_2 ) supplied to photoreceptors by choroid</td>
<td>0.85 (dark), 0.89 (light)</td>
<td>0.91 (dark) (25), 0.93 (dark) (30), ~1.0 (light) (25)</td>
</tr>
<tr>
<td>Dependence of photoreceptor ( QO_2 ) on choroidal ( P_{O_2} )</td>
<td>Linear (dark and light)</td>
<td>Linear (dark and light) (25), 30)</td>
</tr>
<tr>
<td>( Qav_{light}/Qav_{dark} )</td>
<td>0.72, 0.75 (1)</td>
<td>0.33 (25), 0.51 (8), 0.61 (19)</td>
</tr>
<tr>
<td>Inner retinal ( P_{O_2} ) in dark, mmHg</td>
<td>20.4±7.7 (cynomolgus), 10.6±4.6 (rhesus)</td>
<td>18.5±1.8 at ( P_{O_2} &gt; 85 \text{ mmHg} ) (25), 12.1±4.4 (30), 15.5 (8)</td>
</tr>
<tr>
<td>Inner retinal ( P_{O_2} ) in light, mmHg</td>
<td>24.9±7.4 (cynomolgus), 5.8±2.9 (rhesus)</td>
<td>12.6±0.8 at ( P_{O_2} &gt; 85 \text{ mmHg} ) (25), 18.5 (8)</td>
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Values are means ± SD. Numbers in parentheses indicate original reference. \( Qav \), oxygen utilization; \( Qav \), \( QO_2 \) averaged over outer retina; \( P_{O_2} \), arterial partial pressure of \( O_2 \); \( P_{C} \), average choroidal \( P_{O_2} \). *Values from regression of \( Qav \) on \( P_{C} \) at \( P_{O_2} = 50 \text{ mmHg} \).
normal, which is not surprising because under those conditions the photoreceptors have a surplus of O₂. The result of Yu et al. (53) during hyperoxia is similar to the finding in cats (8, 28), pigs (34), and rats (52) that hyperoxia does not increase photoreceptor QO₂.

In both cat and monkey, and all other animals that have been studied by various techniques (3, 18, 29, 38, 42, 49, 55), illumination causes a decrease in photoreceptor QO₂ (Qav). Surprisingly, the changes in Qav in light were considerably smaller in macaques (≈30% reduction) than in cats (50–70% reduction). It is conceivable that this difference resulted from some property other than a species difference. For example, the macaques were all at least several years older than the cats, and they were anesthetized with N₂O-isoflurane rather than urethane. While these factors cannot be ruled out, we propose that a difference in photoreceptor physiology between two species causes the difference in the net effect of light in cats and monkeys. It is known that underlying the change in QO₂ in light are changes in two photoreceptor processes (3, 18, 21). Under light-adapted conditions, the influx of Na⁺ decreases, which decreases the metabolic demand for Na⁺/K⁺ pumping, reducing QO₂. At the same time, the turnover of cGMP in the outer segment increases. Because cGMP is produced from GTP, and in turn from ATP, QO₂ for this process increases. The measured QO₂ is a summation of the two opposite effects of light on Na⁺/K⁺ pumping and cGMP turnover (3, 18). The relative values should depend on rates of enzymatic processes, numbers of channels and pumps, and the geometry of the photoreceptors, so it would not be surprising if the balance was different in macaques and cats. This would lead to a difference in the net effect of light on QO₂.

The only additional evidence that bears on the question of light-dark differences in primate photoreceptor metabolism is measurements of retinal deoxyglucose uptake in dark and light. Bill and Sperber (Ref. 6; also cited in Ref. 11) showed that deoxyglucose uptake was greater in the outer retina in darkness. Taking the lowest value of optical density in the autoradiograms (11) as zero uptake, the outer retinal peak uptake in light was 69% of the value in darkness. Interpreting this value in terms of QO₂ requires the assumption that the autoradiogram density was linear with glucose concentration, and that light-dark changes in glucose uptake reflect changes in oxidative as well as glycolytic metabolism. This is uncertain, since lactate production uses >80% of the glucose in pig (45) and rat (49) retina. However, the deoxyglucose measurements appear to be at least consistent with the relatively small effect of light found in the present study.

One implication of the smaller change in QO₂ with light in monkeys is that human retinal therapies based on the use of illumination to modulate retinal PO₂ may be less effective than would have been expected from the work on cats. For example, initial studies reported that increased light in nurseries for premature infants increased the incidence of retinopathy of prematurity (ROP) (16). This is consistent with the expectation that light exposure should increase the amount of O₂ reaching the inner retina from the choroid, providing the inner retina with enough O₂ from that source that it would not need to develop the retinal circulation. Removal of infants from the high-O₂ environment of the nursery would then stimulate neovascularization as the retina became hypoxic. However, an extensive clinical study [Light Reduction in Retinopathy of Prematurity (LIGHT-ROP); Ref. 36] showed that development of ROP was not dependent on light level, and this now seems consistent with the modest effect of light in changing outer retinal PO₂ in macaques. While many investigators emphasize the role of hypoxia in ROP (39), some results from animal studies of ROP suggest that hypoxia is never the sole factor in causing neovascularization (54). Another situation in which light may influence inner retinal PO₂ is diabetic retinopathy. Increased retinal illumination at night has been promoted as a way to reduce inner retinal hypoxia in diabetes (4, 5), a condition in which the retinal circulation is often compromised (15, 43). Hyperoxia (i.e., inspiration of gas with an elevated fraction of O₂) does improve retinal and visual function during diabetes (13, 17, 22), implying that the diabetic retina is hypoxic, and there is also more direct evidence of inner retinal hypoxia in diabetes (12, 26). However, it now seems questionable whether illumination alone can produce a sufficiently hypoxic condition to be beneficial.

In cats, the large reduction in QO₂ with light elevates the minimum PO₂ substantially, so the photoreceptors have an excess of O₂ in light, which makes QO₂ almost independent of PC (25). In monkeys, however, the smaller change in QO₂ caused by light leaves QO₂ still strongly dependent on PC during illumination. Essentially, this means that during illumination it would take a smaller change in choroidal PO₂ to have an impact on photoreceptor metabolism in monkeys than in cats.

**Foveal Retina**

From our previous study and from the structure of the retina, the shape of the foveal PO₂ profiles was expected to be somewhat different than those in the perifovea. As with perifoveal profiles, these profiles were expected to start with a high PO₂ at the choroid/retinal boundary and decrease across the retina with a distinct bend corresponding to the region of high QO₂ in the inner segments, but then, in contrast to the perifoveal profiles, PO₂ would decrease linearly most of the way toward the vitreous, since there is no retinal circulation in the foveal pit (1). We are confident that the microelectrode was recording from the fovea, because intraretinal ERG measurements (1) proved this in addition to visual inspection. However, the PO₂ in the inner part of the foveal profiles was higher than the PO₂ in the outer retina, which suggested that in its angular trajectory the microelectrode passed through the edge of the fovea, outside the central 0.5 mm, where there is a rich retinal circulation (40). Here, at the edge of the avascular region, the retinal circulation, in addition to the choroidal circulation, supplied O₂ to at least some of the foveal photoreceptors. Thus the avascular fovea may not be as exclusively dependent on the choroid as has been assumed previously.

Photoreceptors in the fovea had lower QO₂ than those in the perifovea in our study and in that of Yu et al. (53). This was surprising, because the mitochondrial density in foveal cone inner segments is ~60% higher than that in perifoveal rods (20) and the inner segments are longer in the fovea (20). To ensure a fair comparison between perifovea and fovea, we and Yu et al. (53) based this conclusion on the QO₂ of the photoreceptors per unit area of outer retina (Qtot), rather than the more conventional basis of unit volume of tissue (Qav). Qtot gives a better estimate of the total amount of O₂ used by the fovea.
outer retina in each region because it is the consumption per unit volume of the inner segments themselves, Q₂, times the length of the inner segments, L₂ = L₁. While the foveal cone inner segments are somewhat longer than peripheral inner segments of either rods or cones (20), this is more than offset by a considerably smaller Q₂ in the fovea. The resulting difference in Q₉₀ was not large, but it was consistent. This result may suggest that rods use more O₂ than cones, but it was impossible to calculate the Q₀₂ per rod or cone because of steep changes in photoreceptor density with eccentricity in and near the fovea, and because the ratio of rods to cones was unknown at the recording sites. Thus whether an individual rod or an individual cone uses more O₂ is still an open question.

Yu et al. (53) also reported Q₀₂ values for “inner fovea” and stated further that Q₀₂ in this region steadily increased as the animals inspired higher percentages of O₂. It is not clear what was meant by “inner fovea.” In the central avascular 0.5 mm of the fovea, there is no inner retina at all. The innermost layer in this part of the fovea is the outer nuclear layer, which does not consume oxygen. A three-layer model rather than the five-layer model they used would be adequate for this region. If they were referring to the region outside the avascular foveal pit on the sloping sides of the fovea, where the inner retina does exist, the inner retina in this region is vascularized and may have several layers of retinal capillaries (40). It is impossible to incorporate this complex O₂ supply into a valid one-dimensional model of diffusion and consumption for two reasons. First, it is not possible to model only the inner retinal layers between vessels, because exactly what part of the electrode track corresponds to these very small avascular regions is unknown. Second, it is not clear how lateral gradients in the inner retina would influence the P₀₂ along an electrode track. Thus, while it seems from the work of Yu et al. (53) that the P₀₂ increases across the entire thickness of the fovea during hyperoxia, we question whether it is valid to also conclude that there is increased Q₀₂.

**Conclusions**

In conclusion, most oxygenation parameters of the perifoveal retina obtained for macaques in this study were very similar to those obtained from cat retina. The major difference is that light had a smaller effect on outer retinal Q₀₂ and P₀₂ in macaques, and the explanation for this will require further investigation. Thus the cat is a good animal model for most studies of retinal oxygenation. Because of the similarities of cats to pigs and rats, at least these species also provide good models for primate, and presumably human, retina. Oxygenation of the fovea is somewhat different from that of the perifovea, because the retinal circulation is absent in the most central region and because the O₂ demand is slightly lower, but with this knowledge properties of the fovea are largely predictable from the data and model for perifoveal retina. This will allow simulations of the oxygenation of this region under conditions that are difficult to study experimentally.

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