Hyperopia and loss of accommodation following ciliary muscle disinsertion in the cynomolgus monkey: physiologic and scanning electron microscopic studies

Paul L. Kaufman, Johannes W. Rohen, and Ernst H. Bárány

Twenty-three cynomolgus monkeys underwent 360-degree disinsertion and retrodisplacement of the ciliary muscle in one eye. Ten to 12 weeks after unilateral disinsertion, resting refraction in the "disinserted" eyes was more hyperopic than in the opposite eyes by 1.12 ± 0.21 (mean ± S.E.M.) diopters (p < 0.001). Accommodative responses to intramuscular pilocarpine (2 or 3 mg/kg) were 0.90 ± 0.14 (mean ± S.E.M.) diopters in the disinserted eyes and 13.88 ± 0.79 diopters in the opposite eyes. The induced hyperopia and loss of accommodation in the disinserted eyes seemed permanent, persisting for at least 14 months in one monkey and 29 months in three monkeys tested periodically after disinsertion. By light microscopy, the ciliary muscle in the disinserted eyes appeared normal and was contracted by pilocarpine. Scanning electron microscopy of the accommodative apparatus revealed retrodisplacement of the ciliary muscle, ciliary processes, and zonular plexus in the disinserted eyes. Structural alterations in the zonular apparatus seemed insufficient to account for the physiological findings. Hyperopia and loss of accommodation following ciliary muscle retrodisplacement are consistent with a new theory of zonular action during accommodation.8

Key words: accommodation, ciliary muscle, ciliary muscle disinsertion, ciliary processes, Macaca fascicularis, monkey eye, refraction, zonule of Zinn

The anterior attachment of the ciliary muscle may be surgically retrodisplaced (i.e., displaced posterolaterally) from the scleral spur to a more equatorial position on the inner scleral wall in the cynomolgus monkey.1 Following retrodisplacement and reattachment, the ciliary muscle retains its normal histologic appearance and its contractility as tested by pilocarpine.5

We report here the refraction, accommodative response to pilocarpine, and scanning electron microscopic appearance of the zonular apparatus in such eyes.

Materials and Methods

Physiological studies. Twenty-four cynomolgus monkeys (Macaca fascicularis) underwent total iris removal5 followed by 360-degree ciliary muscle disinsertion1 in one eye. Following disinsertion, the eye was treated topically with pilocarpine (11 monkeys; 160 μg/hr for 7 days; continuous...
Table I. Effect of ciliary muscle disinsertion and retrodisplacement on baseline refraction

<table>
<thead>
<tr>
<th>No. of monkeys</th>
<th>Disinserted eye</th>
<th>Opposite eye</th>
<th>Disinserted eye − opposite eye</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>$D_1$</td>
<td>$D_2$</td>
<td>$D_{2-1}$</td>
</tr>
<tr>
<td>16$^A$</td>
<td>−0.03</td>
<td>+1.07</td>
<td>+0.09$^e$</td>
</tr>
<tr>
<td></td>
<td>±0.32</td>
<td>±0.37</td>
<td>±0.22$^e$</td>
</tr>
<tr>
<td>7$^b$</td>
<td>+1.04</td>
<td></td>
<td>±0.50$^c$</td>
</tr>
<tr>
<td>23$^c$</td>
<td>+1.06</td>
<td></td>
<td>−0.06</td>
</tr>
<tr>
<td>1$^d$</td>
<td>+1.55</td>
<td></td>
<td>+1.52</td>
</tr>
<tr>
<td>1$^e$</td>
<td>+0.19</td>
<td>+1.34</td>
<td>+1.15</td>
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</tbody>
</table>

Refractions given in dipters (mean ± S.E.M.); $D_1$ = refraction before disinsertion; $D_2$ = refraction 10 to 12 weeks after disinsertion; $D_{2-1}$ = refraction after disinsertion − refraction before disinsertion.

$^a$ Totally iridectomized bilaterally prior to refraction and unilateral ciliary muscle disinsertion. After disinsertion, topical cholinomimetic was pilocarpine in 11 monkeys, echothiophate in 5.

$^b$ Opposite eyes not iridectomized; pupils dilated with topical phenylephrine prior to refraction. After disinsertion, topical cholinomimetic was echothiophate.

$^c$ Monkey 9, totally iridectomized bilaterally. Received topical atropine instead of cholinomimetics after disinsertion.

$^d$ Monkey 7, totally iridectomized bilaterally. Pilocarpine membrane delivery systems not retained in conjunctival sac after disinsertion. Data are included in groups A and C.

$^e$ $p < 0.001$; $p < 0.05$; NS = not significant, by the two-tailed paired t test.

Table II. Effect of ciliary muscle disinsertion and retrodisplacement on accommodative response to pilocarpine

<table>
<thead>
<tr>
<th>No. of monkeys</th>
<th>Accommodation before disinsertion</th>
<th>Accommodation 10 to 12 weeks after disinsertion</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Disinserted eye</td>
<td>Opposite eye</td>
</tr>
<tr>
<td>16$^A$</td>
<td>13.30 ± 0.95</td>
<td>12.88 ± 1.10</td>
</tr>
<tr>
<td>7$^b$</td>
<td>0.81 ± 0.17</td>
<td>14.40 ± 1.32</td>
</tr>
<tr>
<td>23$^c$</td>
<td>0.90 ± 0.14</td>
<td>13.88 ± 0.79</td>
</tr>
<tr>
<td>1$^d$</td>
<td>9.63</td>
<td>9.16</td>
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</tbody>
</table>

Accommodation given in dipters (mean ± S.E.M.).

$^a$ Totally iridectomized bilaterally prior to refraction and unilateral ciliary muscle disinsertion. After disinsertion, topical cholinomimetic was pilocarpine in 11 monkeys, echothiophate in 5. Intramuscular pilocarpine HC1 dose was 2.0 mg/kg in 8 monkeys and 3.0 mg/kg in 8 monkeys. Each monkey received the same dose on both occasions.

$^b$ Opposite eyes not iridectomized; pupils dilated with topical phenylephrine prior to refraction. After disinsertion, topical cholinomimetic was echothiophate. Intramuscular pilocarpine HC1 dose was 3.0 mg/kg in all 7 monkeys.

$^c$ Monkey 9, totally iridectomized bilaterally. Received topical atropine instead of cholinomimetics after disinsertion. Intramuscular pilocarpine HC1 dose was 3.0 mg/kg.

$^d$ Monkey 7, totally iridectomized bilaterally. Pilocarpine membrane delivery systems not retained in conjunctival sac after disinsertion. Data are included in groups A and C.

Release membrane delivery systems, or with echothiophate iodide (12 monkeys, 40 µg once daily for 10 to 14 days; eye drops), or with atropine sulfate (1 monkey, 500 µg once daily for 21 days; eye drops). The pilocarpine- and echothiophate-treated eyes had all received 2 mg of pilocarpine HC1 by intracameral injection at the conclusion of the disinsertion operation; the atropine-treated eye had not. The opposite eyes were either unoperated (7) or totally iridectomized (17) and received no topical drug treatment. All monkeys received intramuscular penicillin G (40,000 IE/kg) and intramuscular methylprednisolone acetate (0.8 mg/kg) once daily, beginning the day of surgery, for 5 days and 4 weeks, respectively. Refraction and the accommodative response to intramuscular pilocarpine-HCl (2 or 3 mg/kg) were determined with a refractometer in both eyes of all monkeys 10 to 12 weeks after unilateral disinsertion, and in 16 monkeys prior to unilateral disinsertion but 2 to 6 weeks after bilateral iridectomy. In the seven monkeys in which the non-"disinserted" eyes were not iridectomized, the pupil was dilated with topical phenylephrine HCl.
Fig. 1. A and B, Scanning electron micrographs of zonular apparatus in surgically aniridic nondisinserted left eye of monkey 230/76. P, zonular plexus; C, cornea; CB, ciliary body; CP, ciliary processes; L, lens; PP, pars plana with zonular fibers; S, sclera; SC, Schlemm's canal.

(100 μl of a 10% solution) before the refraction experiments, permitting refraction in the presence of pilocarpine. In four monkeys the refraction experiments were repeated up to 29 months after disinsertion. Anesthesia for all experiments was by intramuscular methohexital sodium, 15 mg/kg, followed by intramuscular pentobarbital sodium, 30 to 35 mg/kg.

Morphological studies. Six eyes of three cynomolgus not from the previous group were studied. The surgical technique for disinsertion, except for monkey 40/76, was the same as in the previous group; however, the surgeon was different and less experienced in the technique. Postoperative drug treatment consisted of topical echothiophate and systemic antibiotic and corticosteroid as described above.

Monkey 40/76 represented an early effort at disinsertion without prior iridectomy. Disinsertion was attempted in both eyes; neither was entirely satisfactory technically. The right eye was treated with 50 μl of 3% atropine sulfate and the left eye with 50 μl of 6% pilocarpine HCl approximately 60 min before enucleation in vivo. Immediately following enucleation, several elongated meridional scleral strips were excised, and both eyes were then fixed in 2.5% buffered glutaraldehyde.

Monkey 230/76 underwent disinsertion in the right eye 6 weeks after bilateral total iridectomy. Five months after disinsertion, the monkey received intramuscular pilocarpine HCl, 1.5 mg/kg, and was refracted. Three hours later, both eyes were enucleated in vivo and fixed as in monkey 40/76.

Monkey 241/76 underwent disinsertion in the left eye 6 weeks after bilateral total iridectomy. Seven months after disinsertion, the monkey died during the night after having received intramuscular pilocarpine HCl, 1.5 mg/kg, earlier that day for a refraction experiment. Both eyes were enucleated the following morning and fixed in neutral formalin.

In all six eyes, small, wedge-shaped sectors of
Table III. Permanence of induced hyperopia and accommodation loss after ciliary muscle disinsertion and retrodisplacement

<table>
<thead>
<tr>
<th>Monkey No.</th>
<th>Before disinsertion</th>
<th>After disinsertion</th>
</tr>
</thead>
<tbody>
<tr>
<td>23</td>
<td></td>
<td></td>
</tr>
<tr>
<td>23</td>
<td>+0.31</td>
<td>+0.84</td>
</tr>
<tr>
<td>29</td>
<td>-0.58</td>
<td>-0.70</td>
</tr>
<tr>
<td>35</td>
<td>+0.50</td>
<td>+0.44</td>
</tr>
<tr>
<td>51</td>
<td>+0.86</td>
<td>+1.38</td>
</tr>
</tbody>
</table>

All monkeys were totally iridectomized bilaterally prior to refraction and unilateral ciliary muscle disinsertion. Dis. = disinserted eye; Opp. = opposite eye; Ref. = refraction (diopters) before pilocarpine; Acc. = accommodation (diopters) caused by pilocarpine. Intramuscular pilocarpine HCl dose was 2.0 mg/kg in 1 monkey and 3.0 mg/kg in 3 monkeys. Each monkey received the same dose on all three occasions.

* Endophthalmitis following prior anterior chamber perfusion.
† Possibly low pilocarpine dose.

the anterior segment, containing cornea, sclera, lens, ciliary body, and zonular apparatus, were cut out with a razor blade, refluxed in 2% osmic acid, dehydrated with graded ethanol and acetone, and dried by the critical point method. After drying, the membranous remnants of the vitreous were gently pulled away under stereomicroscopic control, exposing the undisturbed zonular apparatus for scanning electron microscopy (SEM). A few sectors from each eye were embedded in Epon, and semithin sagittal sections were cut and stained with methylene blue or periodic acid–Schiff stain (PAS) and examined by light microscopy (LM).

Results

Physiological studies

Baseline refraction (Table I). Before disinsertion, the baseline refractions were very similar in the two eyes of the individual monkeys. After unilateral disinsertion, the baseline refractions in the "disinserted" eyes were more hyperopic, whether compared to their preoperative selves or to those of the opposite eyes. The change was more than 1 diopter and was statistically significant. We cannot say whether it was permanent (Table III). The disinserted eye of monkey 9, which had been treated postoperatively with atropine instead of the cholinomimetics given to all the other monkeys, was not more hyperopic than its nondisinserted opposite eye. The baseline refractions in noniridectomized, phenylephrine-treated opposite eyes were slightly but not significantly more hyperopic than in iridectomized, non–phenylephrine-treated opposite eyes.

Accommodation. Accommodative responses to pilocarpine were drastically (Table II) and permanently (Table III) reduced after disinsertion. Although the accommodation remaining was on the average less than 1 diopter, it was significantly greater than zero (0.90 ± 0.14 diopters; N = 23; p < 0.001; Table II). The disinserted eye of monkey 9 (postoperative atropine) had by far the most residual accommodation (Table II). The disinserted eye of monkey 7 (nonretention of pilocarpine delivery systems postoperatively) had more residual accommodation than 20 of the 22 other cholinomimetic-treated monkeys (more than 21 of 22 if residual accommodation was expressed as disinserted eye/opposite eye).

Accommodative responses to intramuscular pilocarpine 1.5 mg/kg (a submaximal dose in cynomolgus), were determined in two of the three monkeys studied by SEM. Monkey 230/76: disinserted eye, 0.8 diopters; opposite eye, 1.8 diopters; monkey 241/76: disinserted eye, 0.5 diopters; opposite eye, 5.5 diopters.

Gonioscopic and histologic studies of the disinserted eyes reported here had revealed in most eyes a few scattered, localized endothelial and/or connective tissue bands in
the zonular apparatus. However, several of the eyes demonstrating a hyperopic refractive change and loss of the accommodative response to pilocarpine were entirely free of such bands. There was very little noncorneal astigmatism (i.e., astigmatism not correctable by a spherical contact lens) or astigmatic accommodation in either disinserted or nondisinserted eyes.

**Morphological studies**

**Light microscopy.** The disinserted eye of monkey 9 (treated postoperatively with atropine instead of cholinomimetics) showed little or no retrodisplacement of the ciliary muscle over essentially the entire circumference. The disinserted eye of monkey 7, in which the pilocarpine membrane delivery systems were not retained postoperatively, showed a large retrodisplacement in one quadrant and much less retrodisplacement in the other three. Histologic examination of 10 of the other 22 disinserted eyes studied physiologically revealed in each case a large retrodisplacement over essentially the entire circumference. The two cholinomimetic-treated disinserted eyes which had more residual accommodation than monkey 7 were not studied histologically. The disinserted eyes of monkeys 230/76 and 241/76 showed marked ciliary body retrodisplacement. In the few sections studied of the disinserted eyes of monkey 40/76, retrodisplacement was minimal in one eye and greater in the other. In virtually all sections of all disinserted eyes examined previously and in the present study, the attachment between the retrodisplaced ciliary muscle and the sclera consisted of a small area of scar tissue between the sclera and the anterior part of the outer longitudinal muscle fibers overlying the pars plicata; scar tissue never extended as far posteriorly as the anterior pars plana. The reticular and circular portions of the muscle appeared entirely normal.

The ciliary muscle in the disinserted eyes treated with pilocarpine prior to enucleation (monkeys 40/76L, 230/76R, 241/76L) seemed more contracted and anteriorly translocated than the atropine-treated disinserted eye (monkey 40/76R). However, the differences were not dramatic, and no real relaxation was seen in the atropine-treated eye.

**Scanning electron microscopy**

**ZONULAR APPARATUS, CONTROL EYES.** Individual zonular fibers, starting from the region of the ora, join into bands, the broad sides of which are parallel with the pars plana. When these bands enter the valleys between the ciliary processes, they rearrange to a "plexus" oriented sagittally and thus parallel to the sides of the ciliary processes (Fig. 1). In the plexus, the bands fuse edgewise forming weblike sagittal sheets (Fig. 1, B). At the anterior end of the valley, the sheets or plexus split into two main groups of fibers which run to the anterior and posterior lens capsule, respectively. In the depths of the valleys, the plexus splits off fibers obliquely which run anteriorly-corneally-scleralward, terminating in the floor and lateral walls of the valleys (Fig. 2). Thus these fibers, which anchor the zonular plexus to the ciliary body, allow the direction of zonular tension to change from parallel with the pars plana to a more inward direction. They have been called "tension fibers."

**ZONULAR APPARATUS, DISINSERTED EYES.** The ciliary muscle, ciliary processes, and the zonular plexus were all retrodisplaced relative to the scleral spur. However, the zonular plexus was more retrodisplaced than were the processes (Fig. 3), especially in monkey 241/76. Scar tissue binding the outer longitudinal muscle bundles to the sclera extended no farther posteriorly than the level of the pars plicata.

The zonular apparatus itself showed only three relatively minor changes. (1) Fewer
Fig. 3. Scanning electron micrograph of ciliary body in aniridic nondisinserted left eye (A) and aniridic disinserted right eye (B) of monkey 230/76. Apparent loss of substance of ciliary muscle in (B) is related to difference in plane of section between A and B. As verified histologically, it does not represent atrophy of ciliary muscle. CB, ciliary body; L, lens; P, zonular plexus; PP, pars plana; SC, Schlemm's canal. Arrows indicate scar tissue firmly attaching retrodisplaced ciliary muscle to sclera.

zonular fibers were present over the pars plana of the disinserted eyes than in controls, so that the pars plana epithelium could be seen between the zonular fiber bundles in the disinserted eyes (Fig. 4). The zonular plexus was also rarefied. (2) Over the region of the pars plana there were occasional deposits of a condensed material to which some of the zonules seemed to adhere. These zonules were lifted out of the plane of the rest (Fig. 5). (3) Some of the anterior strands pressed against the ciliary processes, causing deep grooves in the epithelium.

No clear-cut differences could be found between the zonular architecture in disinserted eyes treated before fixation with pilocarpine (monkeys 40/76L, 230/76R, 241/76L) and atropine (monkey 40/76R). This agrees with the findings of LM.

CILIARY PROCESSES, CONTROL EYES. The sur-
face of the lateral walls had a cobblestone pattern, and the crests had parallel folds running across them (Fig. 6).

CILIARY PROCESSES, DISINSERTED EYES. There were many normal-looking processes, but in some processes the surface cell pattern was markedly irregular and disorganized. However, there were no denuded areas. This alteration in surface cell pattern was the only difference visible by SEM between the ciliary processes in control and disinserted eyes.

Discussion

Following surgical disinsertion and retrodisplacement of the ciliary muscle from the scleral spur, there was a slight but significant hyperopic shift in the resting refraction. This could be due to flattening of the lens and/or a posterior shift of its position. We cannot say whether disinsertion-induced hyperopia was permanent (Table III). The operation causes some anterior capsular and subcapsular opacities, the latter migrating deeper into the anterior cortex as time passes. Additionally, the four long-term monkeys underwent anterior chamber perfusion for outflow facility measurement in both eyes many times during the 14 to 29 months they were followed. Such insults to the lens might well cause myopic refractive changes, both in disinserted and control eyes.

There was also a profound and permanent diminution of the accommodative response to pilocarpine.

Neither of these effects seems explainable by the relatively moderate structural changes in the ciliary muscle-scleral attachment or the zonular apparatus seen in the disinserted eyes by gonioscopy, LM, or SEM. The involvement of the anterior outer longitudinal muscle fibers by scar tissue at the new scleral attachment does not seem to hinder the forward-inward movement of the main mus-
Fig. 7. Ciliary body and zonular apparatus (schematic). A, Normal eye, nonaccommodating. Pars plana zonules (PPZ), pulled posteriorly by elastic forces of choroid, exert traction on anterior zonules (AZ), with zonular plexus (P), anchored in ciliary valleys by tension fibers (T, inset), acting as fulcrum; lens (L) is stretched and flattened by pull of anterior zonules. CM, Ciliary muscle. B, Disinserted eye, nonaccommodating. Ciliary body is retrodisplaced (ST, scar tissue); entire zonular apparatus is made taut; lens is stretched and flattened more than in A; anterior chamber is deeper and refraction more hyperopic than in A. C, Normal eye, accommodating. Ciliary muscle (CM) contracts, moving forward and inward; tension fibers take up traction of pars plana zonules; anterior zonules slacken; elastic lens capsule assumes relaxed, more spherical shape; anterior chamber shallows slightly. D, Disinserted eye, attempting to accommodate. Ciliary muscle contracts as in C, but retrodisplaced muscle cannot carry zonular plexus sufficiently far forward and inward to slacken anterior zonules; lens remains stretched and flattened; little or no change in refraction or chamber depth occurs.

Both physiologic effects can be explained by retrodisplacement of the ciliary body and zonular plexus. According to the new theory of accommodation put forward by one of us, 7, 8, 9 the lens is kept in the nonaccommodated flat state (Fig. 7, A) by the pull of the pars plana zonules, which in turn are pulled posteriorly by elastic forces of the choroid. This pull is transmitted to the lens capsule by way of the two anterior branches of the zonular apparatus, with the zonular plexus, fixed in the valleys of the ciliary body by the tension fibers, acting as a fulcrum. When the ciliary muscle contracts and moves forward and inward (Fig. 7, C), the tension fibers are carried forward and take up the pull of the pars plana zonules. Thus the elastic pull of the choroid is no longer transferred to the anterior zonules, which therefore slacken, allowing the lens to become more spherical.

In disinserted eyes, there is a postero-lateral displacement of the ciliary body as a whole, along with the zonular plexus. If this retrodisplacement is large enough and, at the same time, the lens is kept in place by the vitreous body, the anterior zonules will be tautened, and an extra flattening of the lens will occur. This would explain the hyperopic shift in resting refraction in the disinserted eyes (Fig. 7, B). Furthermore, if the postero-lateral displacement is large enough, even the contracted muscle cannot carry the zonular plexus far enough forward and inward to slacken the anterior zonules. This would explain the observed lack of accommodation (Fig. 7, D). In monkey 9, where retrodisplacement was minimal, resting refraction was essentially the same in the two eyes, and a moderate amount of accommodation remained in the disinserted eye.

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REFERENCES


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