Scanning electron microscopic studies of the zonular apparatus in human and monkey eyes

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Scanning electron microscopic studies of the zonular apparatus in 10 human and 17 monkey eyes revealed two functionally different sets of zonular fibers: the "main fiber" and the "tension fiber" system. The two systems are connected, forming a broad, sagittally oriented "zonular plexus" within the pars plicata of the ciliary body. The zonular plexus is attached to the ciliary epithelium by the tension fibers, which leave the main system and run obliquely forward to the epithelium deep in the valleys of the ciliary processes. Anteriorly, the zonular plexus splits into the two branches of the "zonular fork," which run respectively to the anterior and posterior aspect of the lens. Thus the zonular plexus can act as a fulcrum. The three-dimensional architecture of the zonular apparatus is consistent with a new concept of accommodation.

Key words: accommodation, ciliary body, ciliary epithelium, ciliary muscle, crystalline lens, Macaca fascicularis, zonule of Zinn

Although the zonular apparatus has been intensively studied by many different methods in recent years, no clear-cut picture of the three-dimensional architecture of this complicated fiber system and its functional relationship to the accommodative mechanism has emerged. Most recent theories are still based on von Helmholtz's concept that the posterior part of the zonular apparatus is moved forward during ciliary muscle contraction, thus slackening its anterior portion and allowing the lens to increase its refractive power. Even Coleman, who performed ultrasonic measurements of the changes of various intraocular diameters during accommodation, postulated in his "unified concept of accommodation" that the posterior zonula and the ora serrata move forward and that the vitreous body plays a major role in accommodation. However, we have shown that there is probably little or no movement of the ora and the zonular fiber system in the posterior part of the pars plana, so that the common concepts of the accommodative mechanism are most likely not fully correct. Along these lines, Farnsworth and associates also recently suggested a re-examination of the principal theories of accommodation.

Since it is impossible for an unsupported fiber system under tension to have a curved or bent course (cf. Fig. 8), some arrangement of the zonular fiber strands other than that which is usually described must exist. Light microscopic sections do not reveal the complete architecture of the zonular apparatus. Therefore we studied its morphologic organization by scanning electron microscopy.
Fig. 1. Transition zone between pars plana and pars plicata of the ciliary body. Note the rearrangement of zonular fibers (Z) within the ciliary valleys. At the posterior end of the ciliary processes (CP) the zonular fibers cross in a regular pattern (arrow). (SEM, cynomolgus monkey; A, ×200, B, ×240.)
Fig. 2. Valleys of the ciliary processes (CP), showing the zonular plexus (ZP) and the zonular fork (ZF). (SEM, cynomolgus monkey; A, ×120; B, ×610.)
Fig. 3. Zonular plexus (ZP) located within a ciliary valley. Note the tension fibers (TF) spreading to the lateral wall of the ciliary process (CP). (SEM, cynomolgus monkey; A, ×500; B, ×500.)
Material and methods

The zonular apparatus of five human eyes, freshly enucleated because of a tumor in the posterior segment, and five human autopsy eyes, enucleated 4 to 12 hr after death, were studied by SEM. The age of the patients varied between 26 and 57 years. Twelve normal cynomolgus monkey (Macaca fascicularis) eyes and five surgically aniridic18 cynomolgus eyes were also studied.

Immediately after enucleation, the human and unoperated monkey eyes were immersed in a 2.5% glutaraldehyde solution buffered with Soren sen’s buffer to pH 7.4 for 3 to 5 hr. For better penetration of the fixative, a 5 by 5 mm window was cut out of the sclera in the equatorial region of the eye. After fixation, small pie-shaped sectors of the anterior segment were cut with a razor blade and post-fixed in 1% osmic acid for 3 hr. The specimens were then dehydrated in graded alcohols and acetones and finally dried in a critical-point drier. With a stereomicroscope (M20, Leitz-Wild), the vitreous membrane could easily be pulled away from the dried specimens without disrupting the zonular fibers. Finally, the specimens were coated with gold-palladium and examined with a Stereoscan scanning electron microscope (Cambridge Instruments Co., Ltd., Cambridge, England).

Results

Zonular apparatus. No major differences in the architecture of the zonular apparatus between human and monkey eyes were found with SEM. When the vitreous capsule is gently pulled away, the zonular fibers become clearly visible (Fig. 1, A). Within the pars plana of the ciliary body, the zonular fibers run mainly in a longitudinal direction, interlacing with each other in a regular manner, so that the entire structure has the appearance of a mat or flat network. This mat is attached to the ciliary epithelium by the internal limiting membrane.

At the posterior end of the ciliary process-
Fig. 5. Zonular apparatus in the region of the ciliary plexus. Within the ciliary valleys many tension fibers (TF) spread obliquely forward and fix the artifactiously elevated main fibers (MF) to the ciliary epithelium. CP, Ciliary process. (SEM, human; A, ×180; B, ×210.)
es, a rearrangement of the zonular fibers takes place (Fig. 1, B). Many finer fiber strands join each other, forming larger anastomosing strands which enter the ciliary valleys. Here, broad interlacing plates or plexuses are formed which are, in contrast to the flat posterior network, orientated mainly in a sagittal plane. These “zonular plexuses” are closely attached to the lateral walls of the ciliary processes (Fig. 2), where some of their fiber bundles split into finer fibers which seem to end at the surface of the ciliary epithelium (Fig. 3). The zonular plexus consists of broad, flattened fiber strands crossing and joining each other in a regular pattern at constant angles (Fig. 2, B).

Anteriorly, the plexus splits into the “zonular fork”, consisting of two main fiber strands running respectively to the anterior and posterior lens capsule (Figs. 2, A, and 4). Finer fiber bundles which run directly to the lens equator and which seem to be of relative...
Fig. 7. Ciliary body showing a transverse fiber system (arrows), probably associated with the vitreous base. CP, Ciliary processes. (SEM, human; ×100.)

unimportance were also seen. The entire zonular apparatus is somewhat bent at the level where the zonular fork is formed (Fig. 4).

From this last observation it appears that the zonular plexuses are firmly attached to the bottom of the ciliary valleys, allowing the zonular fiber bundles to change their direction sharply. There are many coarser and finer fibrils which leave the main strands, run anteriorly in the opposite direction of the main zonular fiber bundles, and become attached to the internal limiting membrane within the depths of the valleys (Figs. 3 and 5). Therefore they cannot be considered to be additional fibers which originate in the pars plicata and join the main fiber system. We assume that these fibers represent a second set of zonular fibers which differ from the first not only in their course and arrangement but also in their function. In previous papers they were called “span-” or “tension fibers.” They run forward in an oblique sagittal direction, often crossing fibers of the zonular fork, which run in the opposite direction, at a right or smaller angle. The crossing pattern of these two sets of fibers can also be seen in thicker than usual light microscopic sections oriented in a sagittal plane. In the untouched zonular apparatus, the system of tension fibers is difficult to recognize by SEM, since it is covered by the main fiber strands. However, after a slight artificial elevation of the zonular fork, the brushlike mass of tension fibers joining the internal limiting membrane throughout the entire length of the ciliary valleys becomes visible (Fig. 5).

The two main fiber strands of the zonular fork lie in a sagittal plane. Near the lens equator these strands again change their orientation, becoming flattened bands which extend mostly parallel to the lens surface; i.e., they change from a sagittal into a frontal plane. The bands split into many fine fibrils which terminate within the zonular lamellae of the lens capsule, where they become firmly attached (Fig. 6). The two main strands attach to the anterior and posterior lens capsule, respectively. However, a small number of mainly finer fiber bundles reach
the equator of the lens, where they spread in a brushlike manner into many fine fibers which are also fixed to the capsule (Fig. 4).

In specimens in which the vitreous membranes have not been completely removed, one can find an additional fiber system, consisting of equatorially oriented fiber bundles fixed to the crests of the ciliary processes (Fig. 7). We assume that these fibers are part of a system of circular bundles which strengthen the vitreous base in this area and by which the vitreous body is firmly attached to the ciliary body.

**Ciliary epithelium.** The ciliary processes project out of the zonular fiber mass, since fibers are only occasionally attached to the crests of the ciliary processes (Figs. 1, 2, A, and 4). The pattern of the pars plicata shows marked individual variations, especially in human eyes, and becomes more complicated with age. The borders of ciliary epithelial cells can be recognized by SEM, so that the cellular pattern of various regions of the ciliary processes can easily be described. At the crests of the processes the epithelial cells show an equatorial orientation (Figs. 1 and 2, A), whereas the lateral walls of the processes exhibit an irregular cobblestone pattern (Figs. 2, A, and 3). The arrangement of the cells within the valleys could not be studied in our specimens. The crests of the ciliary processes often reveal indentations, allowing the crests to be subdivided into “top” and “bottom” regions. These indentations may be caused by the equatorially oriented fiber system of the vitreous body, the main fibers
Fig. 9. Accommodative mechanism schema. A, Nonaccommodating eye. Ciliary muscle (CM) relaxed; anterior zonular fibers (AZ) stretched by traction from posterior (pars plana) zonular fibers (PPZ); lens (L) flattened. B, Accommodating eye. Ciliary muscle contracted, forming an inner edge; the tension fiber system (T) is stretched, taking up the traction from posterior zonular fibers and the choroid. Thus, the anterior zonular fibers become relaxed and the lens more spherical (dotted lines). P, Zonular plexus; I, iris; C, cornea; SC, Schlemm's canal. Arrow indicates direction of ciliary muscle movement during accommodation.

Discussion
Our studies of the three-dimensional architecture of the zonular apparatus by light and transmission electron microscopy as well as SEM suggest a new concept of the tissue mechanics involved in accommodation. The theories of Helmholtz and his followers cannot fully explain the morphological data, as Farnsworth and associates also pointed out recently. Coleman believes that there is an anterior movement of the ciliary epithelium near the ora serrata during accommodation. However, in a light microscopic study on the ciliary epithelium of monkey eyes fixed in different stages of accommodation, we did not find any such morphological changes, although there were some changes in the shape and size of the cells.

In addition, most recent authors believe that the zonular fiber system has a curved or bent course in running from the ora toward the lens. However, a fiber system...
which is stretched between the lens and the ora by the elastic forces of the choroid could not run in a curved manner without intermediate fixation; the fibers would try to follow the shortest course between the terminal fixation points and would be pulled into the vitreous body (Fig. 8). The existence of a second set of zonular fibers, the tension fibers, is consistent with and essential to a new concept of accommodation. These fibers run in a direction opposite to the main fibers, attaching the posterior part of the zonular apparatus to the ciliary body and bracing the zonular fiber system at that point (Fig. 8). The zonular plexus is affixed to the apex of curvature of the main zonular fiber system by the tension fiber system, the latter thus functioning as a fulcrum.

This type of construction is lacking in rats, which have practically no accommodation, but is well developed in monkey and human eyes, which have a large accommodative amplitude. Some "tension fibers" were seen by Coleman in stained whole-mount preparations of bleached human ciliary body, but he thought them to be functionally unimportant accessory fibrils. Recent SEM studies have not mentioned the existence of either the tension fibers or the zonular plexus. Bornfeld et al. described two different groups of zonular fibers, one originating from the pars plana and going to the anterior aspect of the lens and another coming from the ciliary valleys and inserting into the posterior aspect of the lens. Farnsworth and associates on the other hand, described four major tracts of zonular fibers. According to these authors, all these fiber groups show a virtually uninterrupted path except the fourth group, which originates at the posterior aspect of the lens and inserts at the ciliary processes. We also found morphological differences between the zonular fiber strands affixed to the anterior lens surface and those of the posterior lens surface. However, according to our findings, it is of primary importance to recognize that these two major fiber groups intermingle within the ciliary valleys to form the zonular plexus and are anchored there by the tension fibers. Therefore, these structures could function as a fulcrum.

Our earlier and present findings lead us to postulate the following mechanism for accommodation:

1. When the ciliary muscle is relaxed, the elastic network of the choroid retracts the muscle so that the ciliary body becomes stretched. The fulcrum (zonular plexus—tension fibers) moves posterolaterally, allowing the pars plana zonular fibers to exert traction directly on the anterior zonular fibers. The lens capsule is stretched laterally over 360 degrees, flattening the lens and decreasing its refractive power (Fig. 9, A).

2. When the ciliary muscle contracts, its fiber bundles reorient themselves, and the number of circular bundles increases. The ciliary muscle and ciliary body move forward and inward. The tension fiber system is stretched and assumes the traction of the posterior zonular fibers and the elastic network of the choroid, thereby relaxing the fiber strands anterior to the zonular fork. The normal elasticity of the lens capsule, no longer counteracted by zonular traction, molds the lens to a more spherical shape, increasing its refractive power (Fig. 9, B).

Measurements and calculations of changes in lens shape and capsular energy during accommodation should take into account the existence of a zonular fulcrum. Following disinsertion and retrodisplacement of the ciliary muscle insertion in cynomolgus monkeys, the accommodative response to intramuscular pilocarpine is nearly completely abolished despite muscular contraction. Posterolateral displacement of the zonular plexus—tension fiber system fulcrum would stretch the anterior zonular fibers taut regardless of the state of contraction of the muscle and can thereby account for this physiological finding. Indeed, these findings can well be considered as being a further support of our concept that in the absence of a zonular fulcrum anchored in the ciliary body, retrodisplacement of the muscle should have no effect on accommodation.

Details of the attachment of the zonular fibers to the ciliary epithelium and of the
epithelial junctional complexes require further study. This will be the subject of an additional paper.

I am greatly indebted to Mrs. G. Link, whose technical assistance for the scanning electron microscopic preparations was greatly appreciated.

REFERENCES


