Novel Aspects of the Ultrastructural Organization of Human Corneal Keratocytes

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Purpose. Proper functioning of the endothelium and proper structural organization of the keratocytes and collagen bundles are of ultimate importance for transparency of the cornea. The role of the endothelium has been investigated extensively, whereas the role of the keratocytes is still unclear. Detailed knowledge on the ultrastructural organization of keratocytes and the relationship between keratocytes and collagen bundles is as essential for understanding corneal transparency as is knowledge of endothelial functioning.

Methods. Thirty-five corneas (30 postmortem donor corneas and 5 fresh corneas from the operating theater; age range, 28 to 90 years) were used for light microscopy, transmission electron microscopy, and scanning electron microscopy. Serial frontal sections of the central stroma reaching from epithelium to endothelium and cross-sections were studied. At three levels, reconstructions of the mutual arrangement of keratocytes were made using semithin sections.

Results. Keratocytes have the appearance of highly active cells with an abundancy of organelles. Between the dendritic ramifications of these cells, large amounts of amorphous material is observed. One of the most remarkable observations is the presence of an extensive network of fenestrations along the surface of the keratocytes. Another important observation is the circular arrangement of keratocytes gradually turning clockwise like a corkscrew from epithelium to endothelium.

Conclusions. From the current study, the following conclusions can be drawn: Keratocytes are not quiescent but are highly active cells probably involved in turnover of the extracellular matrix; fenestrations may be of functional relevance with respect to facilitation of diffusion and mechanical attachment of the collagen fibers to the keratocytes; the corkscrew organization of keratocytes suggests that they form completely closed sheets of communicating cells throughout the depth of the cornea, creating equal chances for all light rays to pass one or more keratocytes and thus minimizing variation in light scattering over the entire cornea. Invest Ophthalmol Vis Sci. 1995;36:2557–2567.

Keratocytes in the cornea are responsible for the synthesis of collagen fibrils and intercellular matrix. They also maintain the integrity of fibrils and matrix by a steady turnover. The regular arrangement of the collagen fibrils is essential for transparency of the cornea. There is evidence that because of differences in proteoglycan concentration, the anterior and posterior stroma swell differently when corneas are stored in a preservation medium. Swollen corneas appear to have lost a considerable amount of proteoglycans (PGs). These matrix molecules are responsible for the mutual anchoring of fibrils. Loss of PGs might result in rearrangement of the fibrils and, consequently, in a decrease in transparency. Because of changes in the concentration of PGs during development and aging, it is conceivable that swelling properties of the cornea change with age. How keratocytes counteract the loss in PGs is unknown. In addition, there is no agreement on the age dependency of the decrease in corneal transparency.

In birds, fish, amphibians, and reptiles, the stromal collagen lamellae are characterized by a gradual clockwise shift from a maximal 220° in the anterior part and by unrotated orthogonal lamellae in the pos-
The orthogonal organization was observed with transmission electron microscopy in random cross-sections through the mammalian corneal stroma and with scanning electron microscopy.

Recently, it has been shown, using vital dyes, that in the corneal stroma three types of keratocytes can be distinguished. Two types are differentially located either in the anterior or posterior part, whereas the third type is distributed evenly throughout the stroma. Keratocytes are supposed to be restricted to one collagen lamella and not to traverse other collagen lamellae. Using scanning electron microscopy, a network of large flat cells with many small dendritic bifurcations has been shown in rats. Scanning electron microscopic studies, however, do not give information on the internal features of the keratocytes. Descriptions of the ultrastructural characteristics are based mainly on studies using cross-sections. In these articles, special attention was given to the organization of the collagen fibers, whereas the ultrastructure of the keratocytes largely was ignored. This is conceivable because the small number of cytoplasmic organelles observed in cross-sections suggests that keratocytes are relatively inactive cells. To understand the interaction between keratocytes, collagen fibers, and PGs, it is essential, to document the ultrastructure of stromal keratocytes in more detail. In the current study, a description of the ultrastructure of human keratocytes in the anterior and posterior stroma is presented.

**Figure 1.** (A,B) Light micrographs of cross-sections throughout the cornea illustrating the apparently less well-organized collagen bundles in the anterior part (B [An]) and the regular organized posterior part (B [P]). A represents only a portion of the anterior stroma. The keratocytes are seen as dark, tapering, slender profiles (asterisks). Some are characterized by bifurcations (arrowheads). (C) Light micrograph of a frontal section through the anterior part of the cornea. The dark outlines represent keratocytes forming circular structures (arrows). The surrounding collagen fibers (CF) are oriented in different directions. E = epithelium; BM = Bowman's membrane; S = stroma; An = anterior; P = posterior; CF = collagen fibers. Bar = 0.05 mm for A, B, and C.
Organization of Human Keratocytes

FIGURE 2. Panel illustrating ultrastructural details of keratocytes. (A) Cross-section through the posterior part of the cornea. The electron-dense keratocytic profile exhibits an oval nucleus with peripheral heterochromatin and a thin rim of cytoplasm. The collagen bundles alternate and are seen in cross-section (*) and in longitudinal section (**). Notice the electron-dense amorphic patches (arrowheads). (B) In frontal sections, nuclei (N) are highly indented and irregular in shape. This nucleus shows a nucleolus (Nu). Note the extended cytoplasm (Cy). (C) Cross-section at the level of the nucleus (N) illustrating many omega-shaped structures (arrows). (D) In frontal sections, the soma shows numerous organelles, e.g., Golgi field (GF), containing electron-dense material (*), vesicle (V), and rough endoplasmic reticulum (ER). (E) Distally, the perikaryon contains many vesicles (V) and a centriole (Ce). The collagen fibers (CF) on either side of the profile run parallel to each other. (F) Gap junctions (arrows) connecting two adjacent keratocytes. N = nucleus; Cy = cytoplasm; Nu = nucleolus; V = vesicle; GF = Golgi field; ER = rough endoplasmic reticulum; Ce = centriole; CF = collagen fibers. Bar = 1 μm (A), 0.25 μm (B), 0.5 μm (C,D), 1 μm (E), and 0.25 μm (F).

corneal keratocytes and of their organization in both cross-sections and frontal sections will be given.

METHODS

Transmission Electron Microscopy

Thirty slit lamp-rejected donor corneas obtained from the Cornea Bank Amsterdam (postmortem times between 2 and 28 hours) and 5 fresh corneas obtained from eyes enucleated because of melanoma (postmortem time <1 hour) were used for this study. Donor age varied from 28 to 90 years. Four samples of approximately 4 mm² of the central, mediolateral, and lateral cornea were dissected. Because of the curvature of the cornea, only 1 to 2 mm² of each sample could be used for analysis. This study is focused on the central part.
After fixation in a mixture of 1.25% glutaraldehyde–1% paraformaldehyde in 0.08 M cacodylate buffer, pH 7.3, for several days, the tissue was rinsed thoroughly with buffer and postfixed in 1% OsO4 supplemented with 1.5% ferrocyanide for 2 hours. The osmolarity of the aldehyde fixatives was kept within the range of 475 to 500 mOsm. The samples were dehydrated in a graded series of ethanols and embedded in epoxy resin. All were flat embedded, and the majority were mounted on epon blanks before sectioning, thus allowing cutting of perfect frontal sections.

Serial frontal, semithin (1 μm) sections, reaching from the anterior to the posterior part of the stroma, were cut with a diamond knife. All sections had the same interference color, indicating that the thickness was more or less the same. They were collected on
FIGURE 4. Panel illustrating the fenestrations along the surface of the keratocyte. (A) A dendritic process of postmortem cornea showing numerous, regularly arranged fenestrations (arrows). Collagen fibers (CF) run parallel to each other at both sides of the profile. At the right side, another collagen bundle runs perpendicularly to the previous one (**). (B) Scanning electron micrograph of postmortem cornea showing numerous fenestrations (arrows) and spine-like protrusions (S). (C) Fenestrations (arrows) at the periphery of large profile in a fresh cornea. (D) Fenestrated network obtained from fresh corneal tissue. CF = collagen fibers; S = spine-like protrusions. Bars = 1 μm (A, D), 3 μm (B), and 0.5 μm (C).

glass slides and stained with toluidine blue. After every 20 μm, ultrathin sections for transmission electron microscopy were made. From all specimens, semithin and ultrathin cross-sections also were cut and studied. The semithin sections were used to determine the ratio in stromal thickness between the anterior and the orthogonal posterior part making use of a grating in a light microscope. Ultrathin sections were stained with uranyl acetate and lead citrate and inspected in Philips EM 201 and CM 12 electron microscopes (Philips Industries, Eindhoven, Netherlands).

Scanning Electron Microscopy
After fixation in the glutaraldehyde–paraformaldehyde solution, small pieces of the central part were dissected into three parts for scanning electron microscopy: the anterior stroma, the mid stroma, and the posterior stroma. The pieces were dehydrated in a graded series of ethanols and were critical point dried using CO₂. The pieces were mounted on small metal plates with conducting carbon cement and coated with platinum. The specimens were studied in a Philips SEM 505 scanning electron microscope (Philips Industries).

Reconstruction
Samples of the central part of two fresh corneas were reconstructed. The outlines of all keratocytic profiles present in 20 consecutive semithin sections in the anterior, mid, and posterior stroma were drawn, making use of a drawing tube attached to a light microscope. The drawings were copied on transparent sheets and superimposed, reflecting the three-dimensional organization of the keratocytes.

RESULTS
General Observations
The samples of rapidly fixed fresh tissue and those of postmortem tissue showed great ultrastructural simi-
laries. Furthermore, in cross-sections, the ratio in thickness of the anterior stroma and of the orthogonal posterior stroma appeared to be 1:1.3 (SD ± 0.3) in both cases (Fig. 1B, An versus P).

**Light Microscopy**

In cross-sections, the corneal stroma is characterized by an apparently less well-organized anterior, where keratocytes and collagen bundles are not strictly parallel aligned to the corneal surface (Figs. 1A, 1B [An]), and a well-organized posterior (Fig. 1B [P]). Keratocytes are seen as long, slender, tapering profiles (Figs. 1A, 1B, asterisks). Bifurcating keratocytes often are observed (Figs. 1A, 1B, arrowheads), preferentially located in the most anterior part adjacent to Bowman's membrane, suggesting that anterior keratocytes are not restricted to one collagen lamella.

In frontal sections (Fig. 1C), the keratocytes appear to be arranged in a circular fashion. Whether the circular structures represent one or more keratocytes cannot be deduced from the light micrographs. Despite this fact, the cytoplasmic outlines of the keratocytes are 10 to 20 times larger than those observed in cross-sections. Keratocytes in the anterior stroma are surrounded by relatively short collagen bundles. These bundles, often arranged in a starlike fashion, do not disturb the more or less parallel alignment to the corneal surface. As in plywood, the lamellae are organized strictly at right angles, and the anterior stroma can be compared with rotated plywood. In all sections, only parallel collagen bundles, not a single cross-sectioned collagen bundle, was found (Figs. 2E, 6A, 6B).

**Transmission Electron Microscopy**

In ultrathin cross-sections, the nucleus (smallest diameter, approximately 0.5 to 0.7 μm) of the keratocyte occupies the main part of the cell body and is surrounded by a thin rim of cytoplasm containing few organelles (Fig. 2A). The surrounding collagen bundles are orthogonally piled up (Fig. 2A, Nu). Between the collagen fibers, amorphous material is found (Figs. 2A, 2C, 3A, 3D, arrowheads). This material is more abundant between distal branches of keratocytes in cross-sections (Fig. 3E). The small dendrites have diameters between 0.05 and 0.13 μm, and the large dendrites have diameters between 0.26 and 0.90 μm. Frontal sections show tremendous amounts of amorphous matrix, especially in more proximal parts (Fig. 3F). In addition, the nucleus appears to be indented and irregular (Fig. 2B). Its smallest diameter, excluding indentations, measures between 7 and 13 μm, which is approximately 20 times larger than nuclei in cross-sections. Keratocytes usually have one nucleolus (Nu). The perikaryal cytoplasm measures between 4 and 6 μm. In the absence of a nucleus, profiles of up to 18 × 4 μm were observed. Therefore, it is not surprising that the total number of organelles in en face sections (Figs. 2D, 5A, 6A) is higher than that in cross-sections (Figs. 2A, 2C, 3D). However, when comparing the number of organelles in relation to the surface area, the number in en face sections also exceeds that in cross-sections. Notice mitochondria (M), rough endoplasmic reticulum (ER), large Golgi fields (GF), and different types of vesicles (V) (Fig. 2D). In the anterior stroma, the keratocytes contain approximately twice as many mitochondria than in the mid or posterior stroma. Furthermore, omega-shaped structures were observed along the plasma membrane (Fig. 2C, arrows). They measure approximately 40 to 60 nm and also are observed, though less frequently, along the membranes of distal branches. Microtubules often were found to be attached to centrioles (Ce) (Fig. 2E). Regularly, cell

**FIGURE 5.** (A) A curved keratocyte showing an indented nucleus (N) with a nucleolus (Nu) and numerous cytoplasmic organelles. At the upper side, the cytoplasm becomes thin and bends backward to form two bulges. (B) Scanning electron micrograph illustrating a complex keratocyte comparable to that described in A. Note spine-like protrusions (S) and fenestrated region (F). N = nucleus; Nu = nucleolus; M = mitochondrion; ER = rough endoplasmic reticulum; S = spine-like protrusion; K = keratocyte. Bar = 4 μm (A), 5 μm (B).
bodies and branches of keratocytes were found to be connected by gap junctions (Fig. 2F, arrows). These attachment sites rarely were observed in cross-sections. Often, the bifurcations, shown in Figures 1A and 1B, proved to be branches of different cells running parallel to each other (Fig. 3A, asterisk) and showing many mitochondria (M) (Fig. 3A). In other instances, they turn out to be real bifurcations of one keratocyte (Fig. 3B). Distal cell processes are aligned closely, giving the impression of one smooth dendritic process. However, close inspection proved them to consist of several parallel aligned processes (Fig. 3C, arrow). Ultrastructurally, two types of keratocyte can be distinguished in fresh and postmortem corneas. Most keratocytes have a dark nucleus with sparse peripheral heterochromatin and a dark cytoplasm (Figs. 3A, 3B, 5A). Primarily in the anterior stroma, however, cells are observed with a light cytoplasm and a nucleus containing a considerable amount of peripheral heterochromatin (Fig. 3D). In almost all cross-sections of cell bodies and proximal dendrites, bordering collagen bundles are more or less aligned parallel. At more distal parts, a similar parallel arrangement is observed, and the space between the terminal branches is filled with amorphous matrix (Fig. 3E, arrow). Frontal sections do not show such a parallel alignment. All profiles are irregular (Figs. 2B, 3F, 5A, 6B, 6C) and are sometimes filled with amorphous matrix (Fig. 3F).

**Fenestrations**

A remarkable observation of this study is the presence of fenestrations both in postmortem (Figs. 4A, 4B, arrows) and fresh corneas (Figs. 4C, 4D, arrows). Be-
Human Corneal Stroma

Anterior Mid Posterior

Figure 7. Reconstruction of central keratocytes in the anterior, mid, and posterior stromas.

![Image of keratocytes in different layers of the cornea]

As indicated, it is difficult to deduce whether the circular structures seen in frontal sections represent one or more cells, as also illustrated in the scanning electron micrograph (Fig. 5B). Sometimes only cytoplasmic elements without clear borders can be observed (Fig. 6B), whereas in other cases the circular structure turns out to be just one cell (Fig. 5A). Because two nuclear profiles are present in the cytoplasm of this cell, the nucleus must be highly indented (see Fig. 2B). The irregular cytoplasm tapers at the upper side, turns backward, and broadens in two areas showing many organelles. Keratocytes at all levels, from epithelium to endothelium, display fenestrations that measure between 80 and 160 nm (Figs. 6A arrows, 6B, 6C, 6D, 6E). They are more pronounced in cells located in the anterior. The location of the collagen fibers creates the impression that they are connected with the keratocytes at the sites of the fenestrations (Figs. 6A, 6C, 6D, 6E). This was indeed the case for a few of the inspected pairs of stereomicrographs. However, other stereo pairs did not give clear-cut evidence for a real attachment.

Reconstruction

Figure 7 illustrates the superimposed keratocytes when viewed in frontal sections. The density of keratocytes is highest in the anterior stroma and lowest in the posterior stroma. Surprisingly, these reconstructions show that there is hardly any overlap of keratocytes in consecutive sections. At all three levels, it is obvious that keratocytes are not randomly distributed but turn clockwise, like a corkscrew. This means that at a particular distance from the anterior and posterior surfaces, the keratocytes will be seen as closed sheets.

DISCUSSION

Functional Aspects of Keratocytes

The current study shows that keratocytes are not the quiescent cells suggested by the relatively small number of organelles observed in cross-sections. The presence of many stacks of rough endoplasmic reticulum, large Golgi fields surrounded by numerous different type of vesicles, and numerous omega-shaped structures along the membranes indicates that keratocytes are highly active cells involved in the synthesis and storage of proteins and in release and uptake of materials to and from the extracellular matrix. The large quantities of amorphic material in close vicinity of the cellular branches may indicate the presence of a stock of procollagen and/or PG precursors available for turnover.

Twice the number of mitochondria in the anterior stroma indicates a higher metabolism in the anterior stroma than in the mid stroma and posterior stroma. A higher number of mitochondria also was observed in the anterior stroma of mouse and sheep. Differences in mitochondrial densities might reflect the known gradient of oxygen tension from the anterior to the posterior side of the stroma. In addition, the density of keratocytes is higher in the anterior stroma. Also, the corneal innervation is restricted to the anterior stroma. The significance of this anterior–posterior difference is unclear. It has been suggested that different types of keratocytes are located in the stroma. In the current study, two types of keratocytes (light and dark) were discerned, and it can be argued that these cells are in different states of activity. However, activity would be reflected in different numbers of organelles. This was not the case. Both light and dark cells contained comparable numbers of organelles. Although precise comparison is difficult, the study by Poole and coworkers and the current study indicate that more than one type of kerocyte exists. Different keratocytes may produce different substances. Biochemical studies have demonstrated differences in the amount and type of PGs in the anterior and posterior stromas. The ratio of keratan sulphate to dermatan sulphate appeared to be smaller in the anterior stroma than in the posterior stroma. This ratio might be related to the distribution...
of the different types of keratocytes, resulting in the anterior-posterior organization of the cornea.

Why keratocytes contain such high numbers of organelles responsible for protein synthesis remains to be answered. The cornea is irradiated continuously by ultraviolet light. It contains chromophores absorbing in the ultraviolet region between 200 and 295 nm. The proteins of the corneal ground substance would be responsible for the absorption of ultraviolet radiation in the 275-nm to 295-nm region. Ultraviolet absorption leads to protein damage. Maintenance of transparency requires a repair mechanism. Knowledge of PGs and collagen turnover in healthy human corneas is unknown. Detailed studies have been carried out in rabbits. These studies reported that PGs change in corneal scar tissue and that collagen turnover is low because incorporation of radioactive proline was low. Keratocytes might replace the damaged proteins by synthesizing new precursors. Because there is still a discrepancy in the literature concerning an age-dependent decrease in the corneal transmittance of light, one might speculate that the maintenance of corneal light transmission is dependent on the corneal repair mechanism, not on age.

Cell-to-cell communication takes place through gap junctions. In our study, gap junctions were observed regularly in frontal sections but rarely in cross-sections. In rabbits and in humans, gap junctions also were present in cross-sections. The occurrence of many gap junctions in frontal sections suggests that keratocytes communicate in both lateral and anterior-posterior directions. Nishida and coworkers digested collagen fibers in rat tissue and showed by scanning electron microscopy that the remaining keratocytes form an integrated system of closely connected keratocytes. The numerous gap junctions found in our study indicate that keratocytes may indeed be involved in a synchronized system. Recently, it has been shown that gap junctions are indeed functional in human corneas. In many theoretical studies on transparency, the volume of the keratocytes is not taken into account because keratocytes would occupy only 2% to 5% of the total volume. Quantification has shown that the total volume of keratocytes, approximately 10% of the stromal mass, consists of 2.4 million cells. Transparency was always considered to be dependent on the parallel organization of the collagen bundles, the regular distance between the collagen fibers, and the constant diameter of the fibers. These parameters definitely will be of great importance for transparency, but, on the basis of our study and on the studies of Nishida and Watksy, it seems plausible to propose that keratocytes, acting in synchrony, regulate these parameters by controlling the turnover of matrix proteins.

Density of Keratocytes

Reconstruction of keratocytes gave evidence for a larger cellular volume in the anterior part of the corneal stroma. Because no discrimination was made between cytoplasm and nuclei, it is not possible to deduce from these data whether a larger cellular volume represents a higher density of keratocytes or whether individual keratocytes have a more extended cytoplasm. Studies in humans and in rabbit support our finding on different cell densities throughout the stroma. Making use of confocal microscopy, these authors have shown that the number of nuclei and the amount of DNA are significantly higher in the anterior corneal stroma than in the mid stroma and posterior stroma. Therefore, the conclusion seems justified that larger cellular volumes, such as those found in the current study in the anterior stroma, are caused by a higher number of keratocytes and not by keratocytes with more cytoplasm.

Speculation on the Functional Relevance of Fenestrations

This is the first study demonstrating fenestrations along the surface of keratocytes in human corneas. Their presence in fresh human tissue (postmortem time <1 hour), in fresh transcardially perfused mammalian tissue (cynomolgus monkey, data not shown), and in postmortem tissue, irrespective of postmortem time, exclude them as postmortem artefacts. Another possibility is that fenestrations are fixation artefacts. Because no significant differences were observed in the osmolarity of the fixatives and no fenestrations were found in the stromal and subepithelial nerve fibers (Müller, submitted for publication), this possibility can be excluded as well. Although their significance remains to be elucidated, they are most likely involved in the maintenance of the overall organization of the corneal stroma. Some speculations can be made:

1. Fenestrations (Fig. 4) might be confused with omega-shaped structures and may be involved in the turnover of protocollagen and/or PG precursors. Keratocytes indeed have sufficient organelles for protein synthesis and transport (see Functional Aspects of Keratocytes). However, the size of the real omega-shaped (Fig. 2C) structures measures between 50 and 70 nm, whereas the fenestrations measure between 80 and 160 nm. Therefore, it seems unlikely that fenestrations and omega-shaped structures are identical.

2. Because the cornea has no blood supply, nutrients and metabolites have to diffuse from the anterior chamber to the tear film through the stroma, and vice versa. The keratocytes form completely closed covering sheets of cells when viewing from the tear film or anterior chamber. This implies that nutrients and metabolites either have to pass through the keratocytes or pass between the overlapping parts of the cells. If they pass through the keratocytes, one would expect a large number of transcytotic vesicles as described for most endothelial cells of capillaries. These
were not observed. Passing between the overlapping parts would increase the diffusion pathway. Fenestra-
tions could represent pathways for free diffusion, thus
facilitating this process. The observation that fenestra-
tions are present on the more distal parts of overlap-
pling branches (Fig. 3C) supports this proposed func-
tion.

3. In a developmental study on matrix morphogene-
sis by chick corneal keratocytes, Birk and Trelstad44 pre-
sented a scheme in which they proposed large surface-
associated compartments of the extracellular space in
which the cells control the important events of collagen
fiber, bundle, and lamellar organization. These authors
did not find fenestrations, probably because of superpo-
sition in the semithin sections. In another study, in which
they also used high-voltage electron microscopy, it was
suggested that collagen fibers in human tissue emerge
perpendicularly from pits on the surface of the kerato-
cytes.45 Strikingly, the location of the fibers at right
angles to the keratocytes is exactly the same as observed
in the current study (Figs. 4, 6). The pits suggested by
Binder and coworkers45 may be equivalent to the fenes-
trations viewed from the frontal side. This indicates an
intrinsic interaction between fibers and keratocytes. The
search for mechanical attachment sites, for example, was
unsatisfactory. Pairs of transmission electron micro-
graphs, taken at +5° and −5°, did not give clear-cut
evidence on this point when inspected with a stemicro-
scope. However, fenestrations must have functional sig-
nificance because they were observed more frequently
in the anterior stroma than in the mid and posterior
stromas.

Reconstruction
Serial semithin sections and drawings of the profiles of
keratocytes suggested an arrangement of keratocytes
throughout the stroma that was more complex than
circular. Three-dimensional reconstruction of the ker-
atocytes indeed revealed a corkscrew-like arrangement
resulting in completely closed sheets when viewed
from the frontal side. Closed sheets of keratocytes min-
imize variation in light scattering and may attribute to
corneal transparency.3,5,6

In humans, it has not been proven that collagen
bundles gradually turn clockwise. In chicks,29,46 it has been
shown that collagen laminae describe a gradual clockwise
shift of a maximal 220° in the anterior stroma, and they
remain unrotated orthogonally in the posterior stroma.
Our reconstruction data demonstrating that keratocytes
turn in a corkscrew-like fashion in the upper half of the
stoma suggest that collagen bundles gradually turn clock-
wise in humans as well.

These conclusions can be drawn from the current
study: Keratocytes are not quiescent but are highly active
cells probably involved in the turnover of the extracellular
matrix; fenestrations may be of functional relevance with
respect to the facilitation of diffusion and mechanical
attachent of the collagen fibers to the keratocytes; the
corkscrew organization of keratocytes suggests that they
form closed sheets of communicating cells throughout
the depth of the cornea, creating equal chances for all
light rays to pass one or more keratocytes and minimizing
variation in light scattering over the entire cornea.

Key Words
corkscrew organization, fenestrations, human cornea, ker-
atocytes, ultrastructure

Acknowledgments
The authors thank Teja Wesseling, Ben Willeekens, and
Anneke de Wolf for technical assistance; Dr. B. Nunes Cardozo
and Paul Felten for discussions; and Niko Bakker, Ton Put,
and Marina Danzman for photographic assistance. They also
thank all clinicians—especially Dr. M. Kliffen—at the De-
partment of Ophthalmology, Erasmus University, Rotterdam,
for their efforts to obtain fresh corneal tissue from
melanotic eyes.

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