INVESTIGATION OF JUNCTIONAL APPARATUS BETWEEN COLLAGEN FIBERS IN THE CILIARY BODY STROMA AND THE BASEMENT MEMBRANE OF THE CILIARY PIGMENTED EPITHELIUM IN RABBITS

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Abstract : We investigated the junctional apparatus between collagen fibers in the ciliary stroma and the basement membrane of pigment epithelium in the pars plana area in rabbits, in order to clarify how the power of the ciliary body stroma is transmitted to the pigment epithelium during accommodation.

Six eyes of three pigmented rabbits were enucleated and immediately fixed in a half Karnovsky's solution. They were processed for transmission electron microscopic study. In the pars plana area, the basement membrane of the ciliary pigment epithelium had a connection with collagen fibers of the ciliary body in three ways : 1) by direct adhesion ; 2) by way of microfibrils ; 3) by way of cytoplasmic processes of the ciliary pigment epithelium that had protruded in the stroma. The unique structure described above may contribute to flexible and strong mechanical adhesion between the two adjacent structures to withstand frequent mechanical stretch and relaxation during accommodation. The association of the processes with microfibrils has not been reported in the past.

Key words : collagen fibers, ciliary stroma, basement membrane, pigment epithelium, pars plana

INTRODUCTION

Between the inner side of the ciliary muscle and the ciliary muscle lies the so-called ground plate. It can be subdivided into the cuticular lamina representing the basement membrane of the ciliary epithelium, the intermediate connective tissue layer which is composed of loosely arranged collagen fibers, the elastic lamina which is the continuation of the elastic lamina of Bruch's membrane of the choroid, and the vessel layer consisting mainly of numerous capillaries[1].

This layer is the space through which the power of the ciliary muscle is transmitted, thus powering the accommodative mechanism. The elastic microfibrils of the ciliary body have varying architecture regionally, suggesting different vectors of tractional forces in the anterior and posterior ciliary body related to accommodation[2].

Ultrastructual observations on the elastic lamina of Bruch's membrane of the rabbit and human eyes reveal connections to the ciliary epithelium and choroidal elastic tissue. The connections to the ciliary epithelium are in the form of bundles of microfibrils, that peel off the anterior extension of Bruch's membrane beyond the ora serrata and insert into the

basement membrane of the pigmented epithelium of the pars plana. The connections onto the pars plana of the ciliary body implicate Bruch's membrane in disaccommodation when the ciliary muscle relaxes[3].

In the presenting study, we investigated the ciliary body stroma of the pars plana region of pigmented rabbits by transmission electron microscopy, in order to clarify what kind of junctional apparatus or power-transmitting units were present between the basement membrane of the ciliary pigmented epithelium and the collagen fibers of the ciliary body stroma.

MATERIAL AND METHODS

All procedures were conducted in compliance with the ARVO Statement for the Use of Animals in Ophthalmic and Vision Research. Three pigmented rabbits (aged three months) were sacrificed by intravenous injection of pentobarbital sodium (150mg/kg of body weight). The eyes were immediately enucleated, and fixed in a solution of 2.5% glutaraldehyde and 2% paraformaldehyde in 0.1mol/L of phosphate buffer (Half Karnovsky's solution) at a temperature of 4℃. Thirty minutes later, full thickness 3×3mm windows were cut in the posterior sclera adjacent to the optic nerve and in the central cornea using a razor-blade knife, and the globe was fixed overnight. After fixation, tissue slices containing the ora serrata and ciliary body were processed for routine transmission electron microscopic study.

RESULTS

All eyes studied showed similar findings. The ciliary body is poorly developed, due to scarcity of muscle fibers. Fig. 1 shows an example of the area of the ciliary pigmented epithelium and adjacent ciliary body stroma in the pars plana area. Examination by more highly magnified photographs reveals that the basement membrane of the ciliary pigmented epithelium has three kinds of junctions with the stromal collagen fibers. Fig. 2 shows an area where the stromal collagen fibers attach to the basement membrane of the ciliary pigmented epithelium at various angles. Fig. 3 shows an area where microfibrils attach to the basement membrane of the ciliary pigmented epithelium. Fig. 4 shows a peculiar area where cytoplasmic processes of the ciliary pigmented epithelium protrude through the basement membrane to the stroma. Parallel to these process microfibrils and collagen fibers run. Some of the microfibrils appear to stem from the process.

DISCUSSION

It has been reported that rabbits have a narrow range of accommodation measuring 2 to 3 diopters [4,5]. The scarcity of the ciliary muscle may contribute to the much narrower range of accommodation compared with primates.

The collagen fibers in the ciliary body stroma are arranged in an irregular way so that one finds both longitudinally, cross cut, and obliquely cut fibrils on the same section[4]. Lutjen-Drecoll et al[6] studied ciliary muscle topography and connective tissue distribution in atropinized, pilocarpinized, and untreated monkey eyes. They quantatively showed antero-inward movement of the ciliary epithelium in young, pilocarpinized eyes. In our study of rabbits, the collagen fibers attach to the basement membrane of the ciliary pigmented

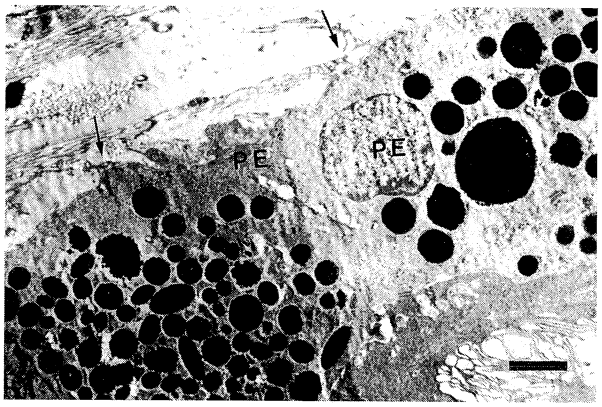

Fig. 1. A transmission electron microscopic study of the pars plana area of the rabbit. The ciliary pigmented epithelium are pulled forward via attachment apparatus (arrows). Bar= 2μm

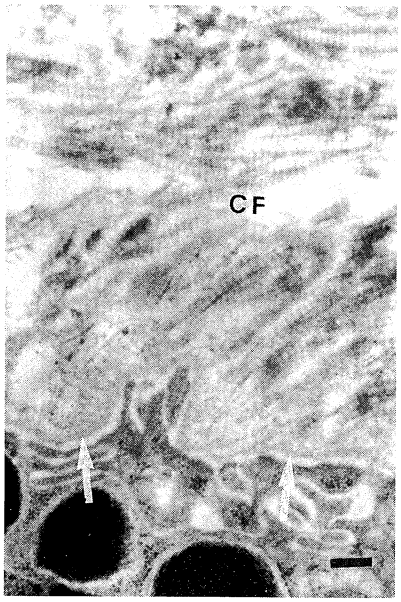

Fig. 2. A transmission electron micro-scopic study of the pars plana area of the rabbit. The basement membrane of the ciliary pigmented epithelium is attached to the collagen fibers of the ciliary body stroma at various angles (arrowheads). Bar= 500nm

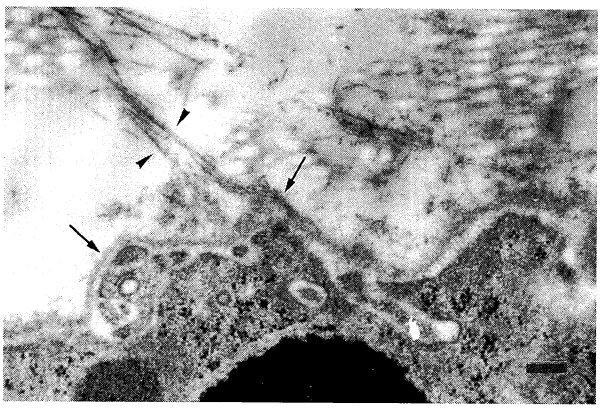

Fig. 3. A transmission electron microscopic study of the pars plana area of the rabbit. The basement membrane of the ciliary pigmented epithelium is attached to the collagen fibers of the ciliary body stroma via microfibrils(arrows). Bar= 500nm

Fig. 4. A transmission electron microscopic study of the pars plana area of the rabbit. There is a cytoplasmic process of the ciliary pigmented epithelium that protrudes through the basement membrane (arrows). This process send microfibrils(arrowheads) to the collagen fibers of the stroma. Bar= 500nm

epithelium at various angles, suggesting that they are playing similar roles to those of monkeys during accommodation.

Our observation of the attachment of the ciliary stromal collagens to the basement membrane of the ciliary pigmented epithelium reveals that at least three types of junction are present.

The first type of connection is direct attachment. The wide area of attachment may facilitate the power of the ciliary muscle to be efficiently transmitted to the ciliary pigmented epithelium. This type of power transmission may most contribute most to the accommodation.

The second type of connection is microfibrils. Korte et al[3] reported that microfibril connections between the ciliary pigmented epithelium and collagen bundles are an important connecting apparatus. This type of connection may contribute to fine adjustment of accommodation or disaccommodation.

Club- and plug-like processes of the pigment epithelium and fingerlike stromal infoldings into the pigmented epithelium were observed in past reports[7,8], which appeared to strengthen the attachment of the epithelium to the stroma. Smaller cytoplasmic protrusions of the pigmented epithelium to the stroma through the basement membrane, as we show here, were reported previously, but to our knowledge, microfibrillar association with these protrusions had not been reported. These peculiar connections may be an additional anchoring apparatus for accommodation, or may play an important role, for example, in preventing mechanical dehiscence or slippage of the ciliary pigmented epithelium and stroma.

REFERENCES

1) **Tamm, E. R.** and **Lutjen-Drecoll, E.** : Ciliary body. Microscopy Research and technique, **33** : 390-439, 1996.

2) **Streeten, B. W.** and **Licari, P. A.** : The zonules and the elastic microfibrillar system in the ciliary body. IOVS **24** : 667-681,1983.

3) **Korte, G. E.** and **D'Aversa, G.**: The elastic tissue of Bruch's membrane. Connections to choroidal elastic tissue and the ciliary epithelium of the rabbit and human eyes. Arch. Ophthalmol. **107** : 1654-1658, 1989.

4) **Prince, J. H.** : The rabbit in research. Charles C Thomas Publisher, Springfield,Illinois, USA, 1964.

5) **Duke-Elder, S. S.** : The eye in Evolution. In:System of Ophthalmology Vol. 1, Henry Kimpton, London, 1958.

6) **Lutjen-Drecoll, E., Tamm, E. R.** and **Kaufmann, P. L.** : Age-related loss of morphologic responces to pilocarpin in rhesus monkey ciliary muscle. Arch. Ophthalmol. **106** : 1591-1598, 1988.

7) **Hara, K., Lutjen-Drecoll, E., Prestele, H.** and **Rohen, J. W.** : Structual differences between regions of the ciliary body in primates. IOVS **16** : 912-924, 1977.

8) **Rohen, J. W.** and **Zimmerman, A.** : Altersveranderungen des Ciliarepithels beim Menschen. Albrecht V. Graefes Arch. Klin. Exp. Ophthal. **179** : 302-317,1970.