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Observation of Cochlear Sensory Hairs in the Neonatal Hamster

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ABSTRACT. The authors performed the observation of the cochlear sensory hairs in the neonatal hamster by the scanning electron microscope from the first day to the sixteenth day after birth, by surface preparation technique under the magnifying glass.

The results were as follows : The cochlear sensory hairs that were observed as the microvilli in the initial stage grew up to the stereocilia of adult hamster, showing the regular pattern of the development of the cochlear sensory hairs. The time of the completion of those development was judged to be eleven, thirteen and sixteen days after birth, in each basal, middle and apical turn of the basilar membrane.

Key words : Cochlear sensory hairs — Pattern of the development — Neonatal hamster

As the inner ear of the hamster is not developed enough at birth, the neonatal hamster is very useful to study the development of the inner ear.

However, we can hardly find any literatures reporting the development of the inner ear in the neonatal hamster, and then we studied the development of the cochlear hair cells in the neonatal hamster by the phase-contrast microscope (PCM) about ten years ago. At that time we observed the regular pattern[1] of the development of the cochlear hair cells, but it was difficult to observe the cochlear sensory hairs in detail by PCM.

Accordingly we tried to observe the cochlear sensory hairs in more detail by the scanning electron microscope (SEM), and we could observe the interesting pattern of the development of the cochlear sensory hairs in the neonatal hamster.

MATERIALS AND METHODS

Twenty-nine neonatal hamsters were used from the first day to the sixteenth day after birth, and the observation of the cochlear sensory hairs was performed everyday from the first day to the eighth day after birth and every other day from the ninth day to the sixteenth day after birth.

These inner ears were taken out being immersed in the fixative (1% paraformaldehyde and 3% glutaraldehyde solution in phosphate buffer), immediately after decapitation under general anesthesia by ether. On the heels of this procedure, the oval and the round windows were opened and several holes were

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made on these cochlear in the fresh fixative. In this manner the fixative was readily admitted into the inside of the cochleae.

After keeping the cochleae in the fixative overnight at 4°C, they were again fixed with 2% osmic acid (OsO_4) solution, and they were rinsed in 70% alcohol and immersed in 70% alcohol.

After they were microdissected by surface preparation technique under the magnifying glass, the dehydration of their specimens was accomplished in an ascending series of ethanol baths, and then these specimens were placed in a series of baths of isoamyl acetate and dried by the critical point method.

Finally, these specimens, particularly the cochlear sensory hairs were observed by the Hitachi HHS-type 2R scanning electron microscope.[2,3]

RESULTS

The pattern of the development of the cochlear sensory hairs observed by SEM seemed to be fundamentally similar to that of the cochlear hair cells observed by PCM.

Namely, the development of the sensory hairs was much earlier in the basal side than in the apical side of the basilar membrane, except being delayed in about a half of the basal end side of hook area.

In comparison between the inner hair cells (IHC) and the outer hair cells (OHC), the development of the sensory hairs was much earlier in the IHC than in the OHC.

In comparison among each row of the OHC, the development of the sensoy hairs seemed to be delayed as the row proceeded to the outside.

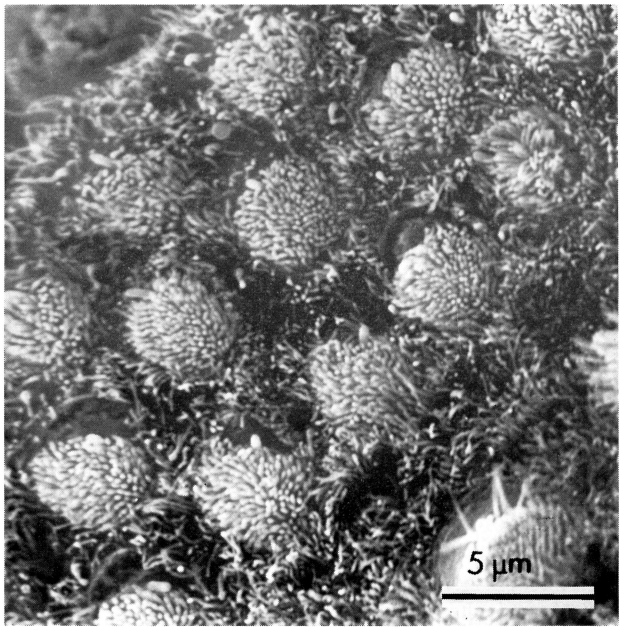

Fig. 1. Sensory hairs on the next day of birth. Sensory hairs are recognized as colonies of microvilli.

By the way, in comparison among each turn of the basilar membrane, the development of the sensory hairs was as follows.

In the basal turn it was recognized as the fairly well advanced stage already at birth, and finished in about eleven days after birth.

In the middle turn it was recognized as the initial stage for one day after birth, and finished in about thirteen days after birth.

In the extreme end of the apical turn it was not recognized for three days after birth, though a ciliary process was found in the center of each cochlear sensory cell that was judged only by the anatomical position in the basilar membrane and the ciliary process seemed to grow into kinocilium in the future. Hereafter the development of the sensory hairs in the apical turn began to be recognized to finish in about sixteen days after birth.

Now, we would like to state the development of the sensory hairs of the OHC in the middle turn, as a concrete example to show the course of the development of the cochlear sensory hairs.

In the middle turn the initial stage of the development of the sensory hairs of the OHC was recognized as colonies of microvilli on the day and the next day of birth (Fig. 1).

The microvilli of the OHC at the row of the axial side (inside) seemed to grow slightly earlier than those at the row of the opposite side (outside). A kinocilium was recognized barely at the outside of each colony of the microvilli.

On the 4th day after birth (Fig. 2), the OHC showed so rapid growth that they seemed to have been pushed up from the bottom.

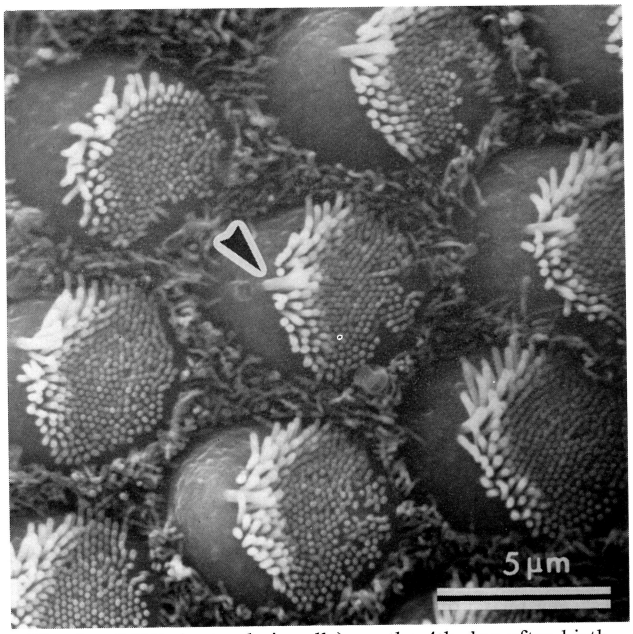

Fig. 2. OHC (outer hair cells) on the 4th day after birth. Microvilli of the axial side become shorter and a kinocilium is clearly recognized at the opposite side (arrow).

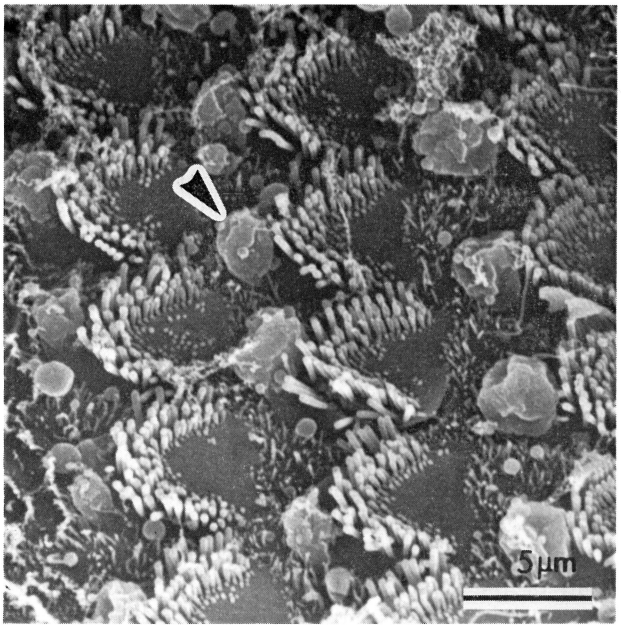

Fig. 3. OHC on the 10th day after birth. Microvilli of the axial side almost disappear and a kinocilium begins to disappear showing a balloon-like form (arrow). Microvilli of Deiters' cells still remain.

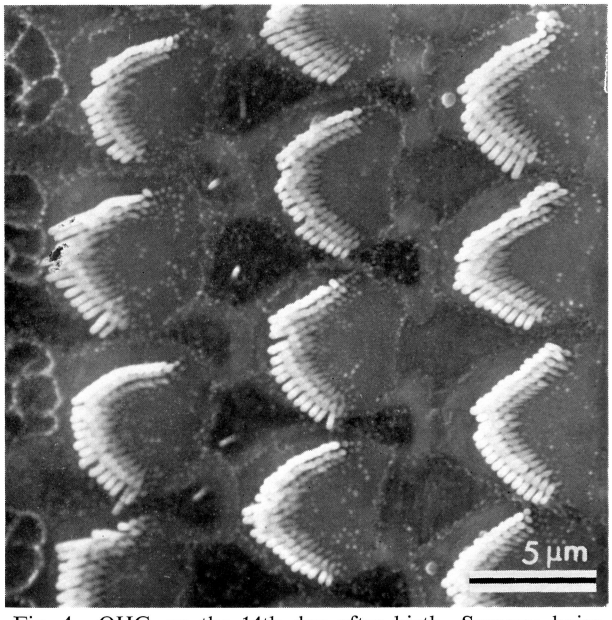

Fig. 4. OHC on the 14th day after birth. Sensory hairs are almost matured. They are recognized as a step-like form of three rows and the opened side of "W" faces to the axis. Microvilli of Deiters' cells almost disappear.

The length of the microvilli of the OHC decreased stepwise proceeding to the inside from the longest of the outermost side, and these shortened microvilli began to disappear one by one in the inside, while a kinocilium of each OHC was clearly recognized at the outside of the colony of microvilli.

On the 10th day after birth (Fig. 3), these microvilli in the inside of the OHC almost disappeared and those in the outside remained. The remained microvilli were partially similar to the stereocilia, but they were not so regular and not enough in the growth.

On the other hand, each kinocilium began to disappear showing a balloon-like form. Though the microvilli of Deiters' cells still remained becoming shorter, they had a tendency to disappear showing a small balloon-like form.

On the 14th day after birth (Fig. 4), the image of the microvilli was completely lost, that is, the microvilli grew up so enough that they were appropriate to be called the cochlear sensory hairs (stereocilia).

The sensory hairs of the OHC were almost matured and recognized as a step-like form of three rows that the opened side of "W" faced to the axis. The microvilli of Deiters' cells almost disappeared.

At this stage these sensory hairs were almost similar to these of the adult hamster (Fig. 5), and then the development of the sensory hairs of the OHC was judged to have finished.

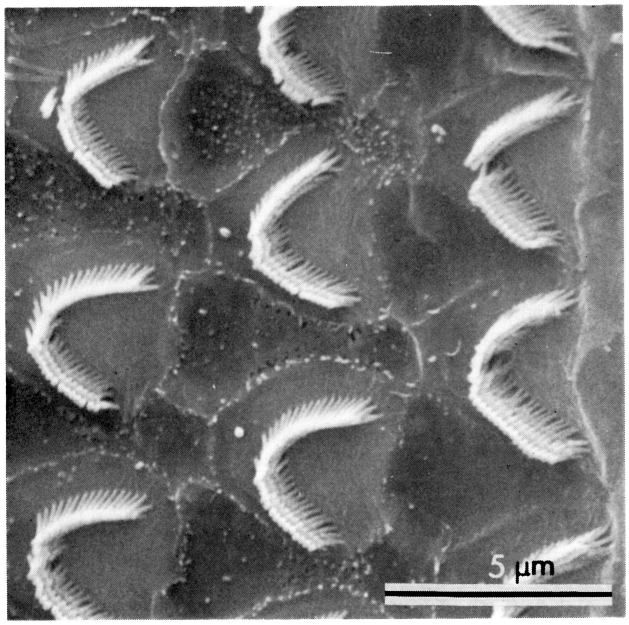

Fig. 5. OHC of an adult hamster.

DISCUSSION

Using the neonatal hamster, the time of the completion of the development of the cochlear sensory hairs after birth was judged to be eleven days in the basal turn, thirteen days in the middle turn and sixteen days in the apical turn

in the observation by SEM, though the time seemed to be four, six and nine days in each basal, middle and apical turn in the observation by PCM.

These differences of the observations by SEM and PCM were considered to be based on the difference of the ability of SEM and PCM, because it was possible by SEM to observe the microvilli that were impossible to be concretely observed by PCM.

Furthermore, the above-mentioned pattern of the development of the cochlear sensory hairs is very interesting in relation to the time of the appearance of the auditory sense in the neonatal hamster and in comparison with human and other mammals.[4–6]

For instance, Lindeman *et al.*[5] have reported that in the one week old kitten, there had been already a clear-cut reduction in the size of the microvilli, which at sixteen days had appeared of normal, adult size, and within a few weeks after birth all the kinocilia had disappeared, after they had observed the cochleae, especially the organ of Corti, in nine kittens aged from two days to six weeks by SEM. In addition to their observation, about these kinocilia, it is generally known that the sensory cells in the mature organ of Corti have no kinocilium.

Kraus *et al.*[6] have discussed the morphological development of the organ of Corti in connection with the onset and the incresase of auditory sense or the development of auditory thresholds, after they had studied the cochleae of five, ten, twelve and fifteen days old mice and adults by light and electron microscopy, from three important sites, that is, the basilar membrane, pillar cells and tectorial membrane.

Hence, we would like to perform continuously the study of the development of the inner ear in the neonatal hamster, with regard to the onset and the increase of auditory sense.

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