The inner pillar cells : an unusual cell type of the auditory organ during neonatal development

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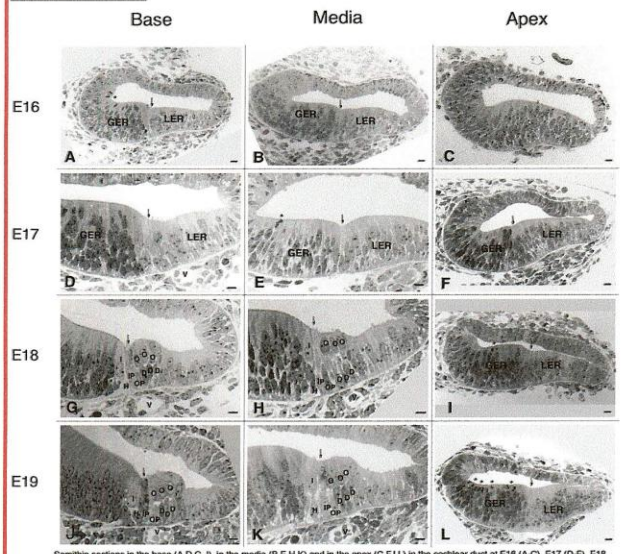
Introduction

The Mammalian organ of Corti is an elongated structure containing a highly ordered cellular pattern. All the cells within the organ of Corti can be identified as one of six unique cell types (inner and outer hair cells, inner and outer pillar cells, inner phalangeal cells, or Deiters' cells). These cells are arranged into a modular cellular unit that is repeated along the length of the organ.

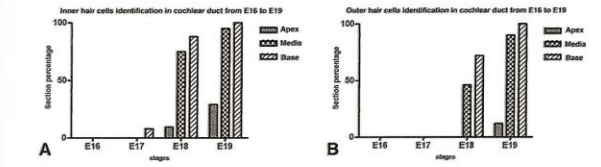
Although the structure of the auditory organ in mature mammals, the organ of Corti, is clearly established, its development is far to be elucidated. Using cytochemical and immunocytochemical methods at the light and electron microscope levels, we examine its spatio-temporal development in rat from embryonic day 16 (E16) to E19.

Morphological analysis ot the dorsal epithelium of the cochlear duct from E16 to E19



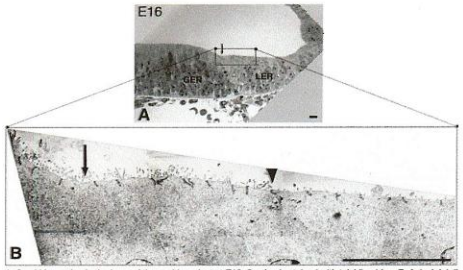
Semithin sections in the base (A,D,G,J), in the media (B,E,H,K) and in the apex (C,F,I,L) in the cochlear duct at E16 (A-C), E17 (D-F), E18 (G-I) and E19 (J-L). All sections are stained with toluidine blue and oriented in the same way with the modiolar region on the left side of the pictures and with the dorsal wall of the cochlear duct at the bottom. Arrow: depression of the dorsal epithelium; star: mitosis; GER: greater epithelial ridge; LER: lesser epithelial ridge; I: inner hair cell; O: outer hair cell; H: phalangeal cell; D: Deiters cell; IP: inner pillar cell; OP: outer pillar cell; V: spiral vessel. Scale bar: 8µm.

Hair cells identification in cochlear duct from E16 to E19



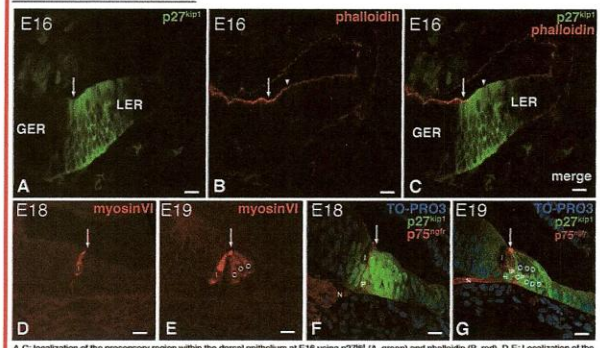
Section percentage exhibiting obvious inner hair cells (A) and outer hair cells (B) at the base, the media and the apex of the dorsal epithelium from rat cochlea.

Morphological differences within the dorsal epithelium of the cochlear duct at E16



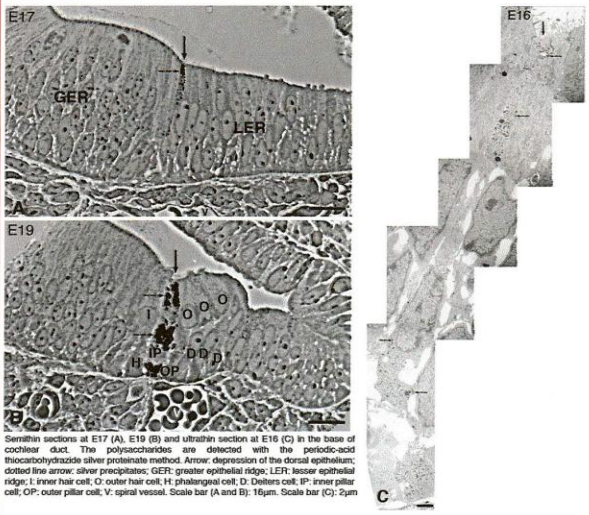
A: Semithin section in the base of the cochlear duct at E16. Section is stained with toluidine blue. B: Apical detail of a serial section of A at the ultrastructural level. Arrow: depression of the dorsal epithelium; arrowhead: transition between the zone with numerous microvilli and the zone with few microvilli; GER: greater epithelial ridge; LER: lesser epithelial ridge. Scale bar: 8µm.

Immunolabellings ot the dorsal epithelium of the cochlear duct between E16 and E19



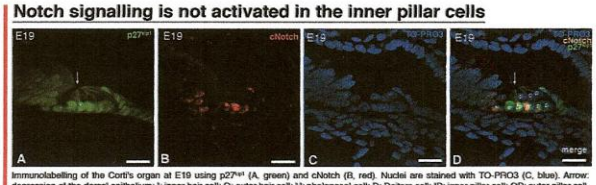
A-C: localization of the prosensory region within the dorsal epithelium at E16 using $p27^{kip1}$ (A, green) and phalloidin (B, red). D-E: Localization of the *hair cells* using *myosinVI* (red) from E18 (D) to E19 (E). F-G: Localization of the pillar cells using $p75^{ngfr}$ (red) from E18 (F) to E19 (G). $p27^{kip1}$ is used to localized the prosensory region (F) and the supporting cells (G). Nuclei are stained with TO-PRO3 (G-F, blue). Arrow: depression of the dorsal epithelium; arrowhead: transition between the zone with numerous microvilli and the zone with few microvilli; GER: greater epithelial ridge; LER: lesser epithelial ridge; I: inner hair cell; O: outer hair cell; H: phalangeal cell; D: Deiters cell; IP: inner pillar cell; OP: outer pillar cell; N: extensions of the spiral ganglia. Scale bar: 16µm.

Characterization of the first identifiable cell of the Corti's organ at the light and electron microscope levels



Semithin sections at E17 (A), E19 (B) and ultrathin section at E16 (C) in the base of cochlear duct. The polysaccharides are detected with the periodic-acid thiocarbohydrazide silver proteinate method. Arrow: depression of the dorsal epithelium; dotted line arrow: silver precipitates; GER: greater epithelial ridge; LER: lesser epithelial ridge; I: inner hair cell; O: outer hair cell; H: phalangeal cell; D: Deiters cell; IP: inner pillar cell; OP: outer pillar cell; V: spiral vessel. Scale bar (A and B): 16µm. Scale bar (C): 2µm

Notch signalling is not activated in the inner pillar cells



Immunolabelling of the Corti's organ at E19 using $p27^{kip1}$ (A, green) and cNotch (B, red). Nuclei are stained with TO-PRO3 (C, blue). Arrow: depression of the dorsal epithelium; I: inner hair cell; O: outer hair cell; H: phalangeal cell; D: Deiters cell; IP: inner pillar cell; OP: outer pillar cell. Scale bar: 16µm.

Conclusions

We demonstrate that the Corti's organ develops from a non-proliferating cell zone that is located in the junctional region between two edges of the dorsal epithelium of the cochlear duct. We also reveal that the first cells to develop in this zone are the inner pillar cells, a particular nonsensory supporting cell type; they arise in the base of the cochlear duct at the boundary between the two ridges at E16. The cell differentiation in this prosensory region continues according to a base-to-apex gradient, the inner hair cells appears in the greater epithelial ridge at E17 and the outer hair cells in the lesser epithelial ridge at E18. At E19, the different cell types of the Corti's organ are in place. Finally, we show that unlike the development of all the supporting cell types of the Corti's organ, the development of inner pillar cells within the prosensory domain does not involve Notch1 signalling. These results highlight the central role that could play the inner pillar cells in the Corti's organ development.

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