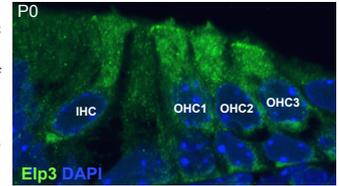


Disruption of the lysine acetyltransferase Elp3 causes planar polarity defects in the mouse organ of Corti

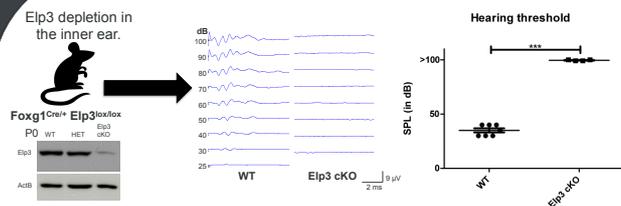
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Introduction

The migration of the kinocilium towards the lateral side of the hair cell orchestrates the orientation and characteristic "V-shape" of the stereocilia hair bundle. Disrupting the genesis and/or migration of the kinocilium causes defects in hair bundle morphology, and also in the planar cell polarity of the organ of Corti (1). Elp3 is the catalytic subunit of Elongator, a six-subunit complex with lysine acetyltransferase activity, and has been shown to target alpha-tubulin, thereby controlling axonal branching and cortical neuronal migration in mice (2). Given the importance of acetylation homeostasis in developmental processes, and that the kinocilium is a tubulin based structure, we investigated the role of Elp3 during inner ear development.



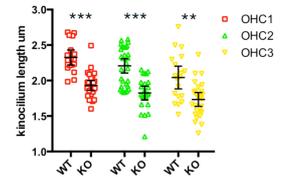
Elp3-deficient mice are deaf



Elp3 depletion perturbs kinocilium length...

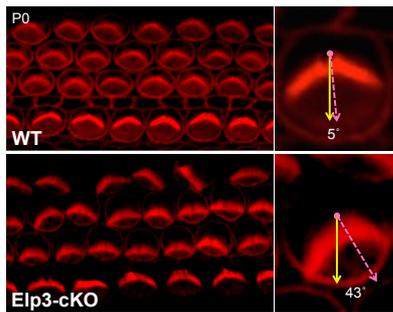
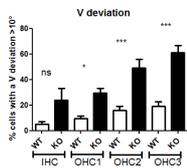
Because planar polarity defects in the organ of Corti have previously been reported in ciliary protein mutants, we assessed the length and position of the kinocilium in our Elp3-cKO mice.

The length of the kinocilia of outer hair cells was significantly reduced in the Elp3-cKO mice.



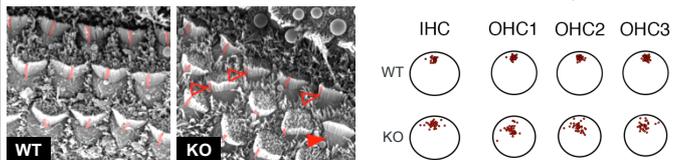
Elp3 depletion causes planar polarity defects...

The hair cells polarise along the medio-lateral plane of the organ of Corti, with their V-shaped hair bundles all pointing laterally. By measuring the deviation of the hair bundle from the medio-lateral axis, we see that Elp3 depletion significantly disrupts the planar polarity of the organ of Corti.

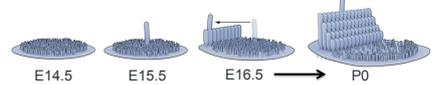


...and also kinocilium localisation

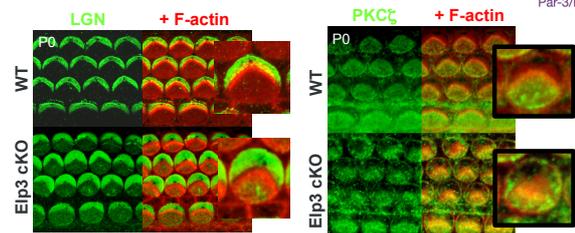
The kinocilia of Elp3 cKO hair cells were often displaced (red-lined arrowheads) and occasionally absent (solid red arrowhead) from the vertex of the V-shaped stereocilia bundle. Plotting the exact position of kinocilia on the apical surface of Elp3 cKO hair cells showed a more scattered distribution. The kinocilia were also more centrally localised.



The kinocilium first emerges from the centre of the apical surface, before migrating to the lateral side of the hair cell.



Its migration relies upon the same molecular machinery that pulls on microtubules and orients the mitotic spindle during cell division. At the hair cell's apical surface, laterally (L) restricted Gai3 and LGN are thought to play a role in pulling on the microtubule aster, moving the kinocilium to the lateral side of the cell. Atypical PKC ζ and Par3/6 define the medial zone (M) of the apical surface (3,4).



In Elp3 cKO hair cells, we found that LGN expression is extended centrally, and also mislocalised to the medial side of the hair cell. Conversely, PKC ζ expression was mislocalised laterally.

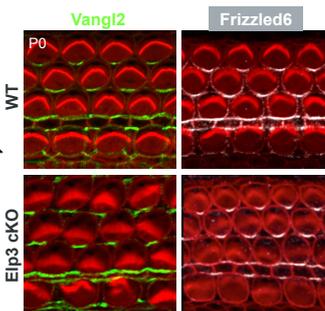
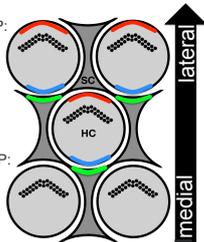
...but the localisation of core PCP proteins is normal

Planar polarity in the organ of Corti is regulated by the asymmetric localisation of the core-PCP proteins across its medio-lateral axis.

We saw that the asymmetric localisation of Vangl2 and Frizzled6 was maintained in Elp3 cKO mice.

Lateral core PCP:
Dvl2
Vangl2

Medial core PCP:
Frizzled3/6



Perspectives

The mislocalisation of LGN could disrupt the lateral pulling forces upon the kinocilium. LGN is tethered to the cell membrane via its binding to Gai3, and this interaction is inhibited by the activity of PKC ζ .

We are currently investigating the potential mechanism through which Elp3 mediates this process. Elp3 may play a general role in trafficking proteins to the apical surface, or alternatively it may interact specifically with one or more of the laterally/medially localised proteins (e.g. LGN, PKC ζ) that control kinocilium migration, ensuring their correct distribution. We are also analysing the developmental timepoints at which kinocilium migration occurs to ascertain the onset of this phenotype.

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- References:
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 2. Creppe et al., Cell (2008)
 3. Ezan et al., Nat. Cell Biol. (2013)
 4. Tarchini et al., Dev. Cell (2013)