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Intraflagellar transport in skeletal development

R. Serra

Department of Cell Biology, University of Alabama at Birmingham, Birmingham, AL, USA

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Primary cilia are non-motile microtubule based appendages extending from the surface of almost all vertebrate cells. The process of Intraflagellar Transport (IFT) is responsible for building and maintaining the structure and function of primary cilia. Disruption of *Kif3α*, a component of the Kinesin-II motor complex, disables anterograde IFT and leads to failure in the formation and maintenance of cilia. Likewise, the absence of *IFT88/Tg737/Polaris*, a core component of the IFT particle, results in the loss of cilia. The functional significance of primary cilia has only recently been uncovered. We recently used the Cre-Lox strategy to target deletion of IFT proteins to skeletal tissue to determine the role of primary cilia in embryonic and post-natal skeletal development[1-3].

Deletion of Kif3α or IFT88 in Prx1 expressing cells results in defects in embryonic endochondral bone formation including a dramatic reduction in bone length and accelerated hypertrophic differentiation as early as E14.5 days[1]. Prx1- Cre is expressed in early limb mesenchyme starting at E9.5 days and thus recombination is present in chondrocytes and perichondrium/periosteum[4]. Defects in endochondral bone formation were at least in part due to disruption of Indian Hedghog (Ihh) signaling as measured by the expression of Ptc1 and Gli1, direct targets of Hh signaling. Ectopic cartilage was found in the periosteum along the diaphysis in E18.5 day embryos. This phenotype is distinct from that seen in Ihh-null mice and suggests that additional signaling pathways, for example, canonical Wnt signaling, may also be affected by the loss of IFT.

In contrast, deletion of Kif3α or IFT88 in Col2α expressing cells did not affect embryonic skeletal development; however, defects in the post-natal growth plate were seen starting at about P7 days[3]. The differences in the two mouse models are likely due to the localization and timing of Cre-mediated recombination. Col2α-Cre is expressed later in the limbs (E12.5 days), after cells have committed to the chondrocyte lineage, and the perichondrium/periosteum is not efficiently targeted[5,6]. Even though alterations in skeletal development were not observed until P7 days, Ptc1 expression was dramatically reduced in chondrocytes as early as E15.5 days. Ptc1 was maintained in the perichondrium. There is significant evidence for Ihh signaling in the perichondrium during embryonic development[7-9] and this may in part explain the lack of an embryonic phenotype in *Col2αCre;Kif3α$^{fl/fl}$* mice. In the post-natal growth plate, disruption of IFT resulted in reduced proliferation and altered hypertrophic differentiation. Cell shape and columnar orientation in the growth plate were also disrupted suggesting a defect in the process of rotation. Alterations in chondrocyte rotation were accompanied by disruption of the actin cytoskeleton and alterations in the localization of activated FAK to focal adhesion-like structures on chondrocytes. Alterations in Hh signaling as measured by Ptc1 expression were not detected in the post-natal growth plate even though the alterations in part resembled those seen in mice with a post-natal disruption of Ihh[10]. The results suggest that additional signaling pathways may be involved. For example, imaging studies have suggested that chondrocyte cilia could transmit mechanical force through their interaction with the surrounding ECM thus acting as mechanosensors[11].

Alterations were also detected in the cranial base synchondroses in post-natal *Col2αCre;Kif3α$^{fl/fl}$* mice, including growth retardation, disorganization of the growth plate and alterations in chondrocyte differentiation[2]. Ptc1 expression was dramatically reduced in growth plate chondrocytes but the expression domain of Ptc1 was expanded in the perichondrium suggesting increased Hh signaling in this tissue. Excessive intramembranous ossification as well as ectopic cartilage was seen in perichondrial tissue. Cranial base defects in mice deficient in Ihh only minimally resembled those in which Kif3α was deleted suggesting that cilia affect signaling through additional factors.

Together, the results indicate that primary cilia/ IFT are involved in multiple signaling pathways within the skeleton.

The author has no conflict of interest.

Corresponding author: Rosa Serra, University of Alabama at Birmingham, Department of Cell Biology, 660 MCLM, 1918 University Blvd, Birmingham, AL 25294-0005, USA
E-mail: rserra@uab.edu

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