

# Primary Cilium, a Potential Target of Microgravity Induced Bone Loss

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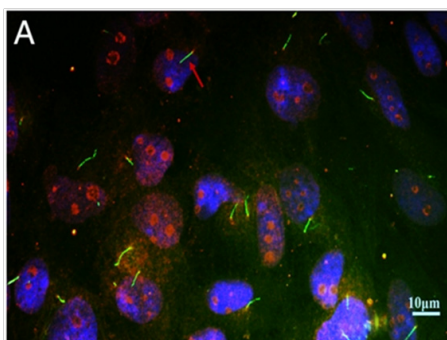
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## Abstract

Microgravity in space can cause various problems in different biological systems. One of the most prominent and well recognized physiological challenges accompanying an extended spaceflight is the reduction in bone mass [1]. However, the underlying mechanisms of this phenomenon are still elusive. Primary cilium is a solitary and special organelle that emanates from the surface of most mammalian cells, which is anchored to the cell by mother centriole during the interphase and G0 of cell cycle [2]. For a long time, primary cilium was considered as a vestigial organelle. Until recently it was found that primary cilium provided a means of sequestering the centriole, so as to inhibit cell division. More significantly, a variety of receptors, ion channels and transporter proteins have been localized to the cilium, which has been proved as a key coordinator of signaling pathways to respond mechanical and chemical stimuli [3,4]. Primary cilium has been proved as a mechanosensor to regulate bone formation both in osteocytes and osteoblasts. It can act as a sensory organelle to receive extracellular signals and change its orientation to translate mechanical stimuli into biochemical and transcriptional changes, and as a result, bone formation is activated [5,6]. Inspired by established role as a mechanosensor, the role of primary cilium in microgravity induced bone loss must be studied. In here we presents the structure and function of primary cilium in bone metabolism process, and prospects the importance of primary cilia in microgravity stimulated osteoporosis.

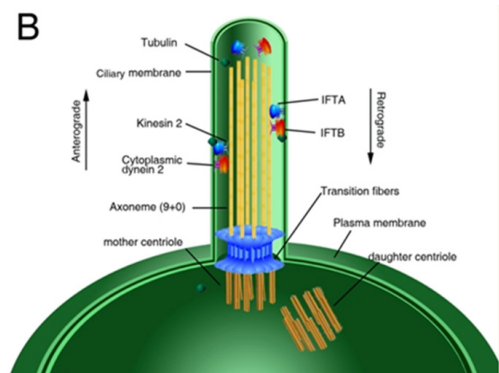
## Primary Cilium Structure and Growth



**Figure 1:** Immunofluorescence image of primary cilium. Primary cilium were stained by acetylated  $\alpha$ -tubulin (green). The basal bodies were stained by  $\gamma$ -tubulin (red), and the nuclei were stained with DAPI (blue).

Primary cilium consists of an axoneme of nine doublet microtubules that extends from the mother centriole of the centrosome, which is enclosed by the ciliary membrane, an extension of the cell membrane (Figure 1). The ciliary compartment is separated from the cytoplasmic matrix by transition fibers which connect the basal body (mother centriole) to the plasma membrane [2,7]. To build a primary cilium, the axonemal microtubules polymerize is delivered by intraflagellar transport (IFT). The IFT

system is consisted by two associated cargo proteins, IFT-A, which is transported along axonemal microtubules by kinesin 2 motor proteins in the anterograde (base-to-tip) direction, and IFT-B, which is delivered by cytoplasmic dynein 2 in the retrograde (tip-to-base) direction [8]. The overall protein composition of cilia is very complex, considering the function and structure of cilia. It has been estimated that cilia contain over 1,000 different proteins [9]. The tasks to confirm the functions of these proteins are still on the way (Figure 2).



**Figure 2:** The schematic structure of primary cilium.

## New Signaling Pathways Related With Primary Cilium

The signaling pathways coordinated by primary cilia are quite diverse and new pathways are identified by researchers every year. Except signaling like platelet-derived growth factor receptor (PDGFR), Wingless/Int-1 and Hedgehog (Hh)[10-12], more new primary cilium-located proteins were reported, including Transforming growth factor  $\beta$  (TGF- $\beta$ ), protein kinase A, anoctamin and septin [13-15]. Xie et al. [16] reported that BMP-Smad1/5/8 signaling in osteoblasts was regulated by primary cilium through BMP receptor II (BMPRII) which was localized at the bases of cilia. Kwon et al. [17] found that dynamic flow transiently decreased cAMP production in a primary cilium dependent manner in osteocytes and adenylyl cyclase 6 (AC6) preferentially localized to the cilium [17]. Clement et al. [18] reported that TGF- $\beta$  receptor localized to the ciliary tip and endocytic vesicles at the ciliary base in fibroblasts and that TGF- $\beta$  stimulation increases receptor localization and activation of SMAD2/3 and ERK1/2 at the ciliary base [13]. Besides, many studies have shown that receptor tyrosine kinase (RTK) signalling pathways were involved in primary cilium, including PDGFR $\alpha$  signalling, IGF-1R signalling, EGFR signalling and FGFR signalling [18].

## The Role Of Primary Cilia In Bone Homeostasis

Over the past decade the primary cilium has emerged as a novel extracellular sensor in bone, being required to transduce changes in the extracellular mechanical environment into biochemical responses regulating bone adaptation. Malone et al. [6] report that bone cells possess primary cilia that project from the cell surface and deflect during fluid flow and that these primary cilia are required for osteogenic and bone resorptive responses to dynamic fluid flow. Hoey et al. [19] demonstrated that mechanically stimulated osteocytes secrete soluble factors which act in a paracrine manner to enhance osteogenic gene expression in MSCs. For in vivo studies, Temiyasathit et al. [20] demonstrated that deficient primary cilia in osteoblasts and osteocytes results in reduced loading induced bone formation and verifies earlier in vitro. Lee et al. [21] reported that knockout of AC6, which located in primary cilium, significantly reduced response to ulnar loading compared to WT rat. In addition, primary cilium also plays a key role in other bone promoters induced osteogenetic progress. For example, Yan et al. [22] found that primary cilia of osteoblasts play an indispensable role in mediating pulsed electromagnetic fields (PEMFs) osteogenic effect in vitro. Ma et al. [23] reported that primary cilia are important in mediating icaritin-stimulated osteogenic differentiation and may be a novel target for pharmacological therapies for bone loss.

## Primary cilium may be responsible for microgravity induced bone loss

Inspired by its established role as a sensory organelle, we hypothesize that primary cilium is a critical target of microgravity stimuli based on the following facts. Firstly, primary cilium is a mechanical stimulation sensor, which can change its orientation

and length after the treatment of many force, like fluid shear stress and electromagnetic force [2]. If cells are exposed into microgravity condition for a long time, the primary cilium may change its length or morphology to adapt new environment. Secondly, on the one hand, it was found that primary cilium played an indispensable role in mediating normal bone formation induced by bone promoters. On the other hand, many studies have shown that microgravity can inhibit the differentiation, maturation and mineralization of osteoblasts and promoted bone loss [24].

Thus hypothesis that microgravity leads to bone loss because of the changes of primary cilium may be reasonable. Thirdly, reports had shown that the primary cilium can be as a gravitational force transducer. Moorman et al. [25] reported that primary cilium can detect the cyclical changes in the Earth's gravitational field and transduce those changes into changes in the variability (stochastic nature) of gene expression. Finally, it has been found that microgravity affects the cytoskeleton in various cells, and it has been known that there are intrinsic connections between cytoskeleton changes and primary cilia. Dai et al. [26] reported that actin microfilaments participate in BMP-2 activity in activating Runx-2 and that their disruption might be an important contributor to microgravity-induced inhibition on BMP-2 activity in osteogenic induction [26]. Sharma et al. [27] reported that cytochalasin D increased the cilia length through changes in the actin network and in levels of soluble tubulin. Therefore, microgravity can affect actin cytoskeleton and it may change the primary cilium because of their intrinsic connections. In summary, future works need imminently to determine whether microgravity affects the length, orientation or morphology of primary cilia and whether these changes is responsible for the microgravity induced bone loss.

## References

- Grimm D, Grosse J, Wehland M, Reseland JE, Sundaresan A, Corydon TJ, et al. (2016) The impact of microgravity on bone in humans. *Bone* 87: 44-56.
- Satir P, Pedersen L B, Christensen S T (2010) The primary cilium at a glance. *Journal of Cell Science* 123(4): 499-503.
- Shearer R F, Saunders D N (2016) Regulation of primary cilia formation by the ubiquitin-proteasome system. *Biochemical Society Transactions* 44(5): 1265-1271.
- Pedersen L B, Mogensen J B, Christensen S T (2016) Endocytic Control of Cellular Signaling at the Primary Cilium. *Trends in Biochemical Sciences* 41(9): 784-797.
- Hoey D A, Kelly D J, Jacobs C R (2011) A role for the primary cilium in paracrine signaling between mechanically stimulated osteocytes and mesenchymal stem cells. *Biochemical & Biophysical Research Communications* 412(1):182-187.
- Malone A M, Anderson C T, Tummala P, Ronald Y Kwon, Tyler R Johnston, et al. (2007) Primary cilia mediate mechanosensing in bone cells by a calcium-independent mechanism. *Proceedings of the National Academy of Sciences of the United States of America* 104(33): 13325-13330.
- Malicki J J, Johnson C A (2016) The Cilium: Cellular Antenna and Central Processing Unit. *Trends in Cell Biology* 27(2): 126-140.
- Vertii A, Bright A, Delaval B, Hehnlly H, Doxsey S (2015) New frontiers: discovering cilia-independent functions of cilia proteins. *EMBO Rep* 16(10): 1275-1287.

9. Pazour G J, Agrin N, Leszyk J, Witman GB (2005) Proteomic Analysis of a Eukaryotic Cilium. *Journal of Cell Biology* 170(1):103-113.
10. Schneider L, Clement C A, Teilmann S C, Satir P, Christensen ST, et al. (2005) PDGFR alpha signaling is regulated through the primary cilium in fibroblasts. *Cur Bio* 15(20): 1861-1866.
11. Goetz SC, Ocbina PJ, Anderson KV (2009) The primary cilium as a Hedgehog signal transduction machine. *Method Cell Biol* 94: 199-222.
12. Liu B, Chen S, Helms J A (2014) The primary cilium as an integration center for Hedgehog and Wnt signalling. *Reproductive Toxicology* 48(48):19-19.
13. Clement C A, Ajbro K D, Koefoed K, Andersen CY, Larsen LA, et al. (2013) TGF- $\beta$  signaling is associated with endocytosis at the pocket region of the primary cilium. *Cell Reports* 3(6):1806-1814.
14. Hwang S H, Mukhopadhyay S (2015) G-protein-coupled receptors and localized signaling in the primary cilium during ventral neural tube patterning. *Birth Defects Res A Clin Mol Teratol* 103(1):12-19.
15. Rania G, Qicong H, Marion F, Marie-Christine R, Benjamin S, et al. (2013) Septins 2, 7 and 9 and MAP4 colocalize along the axoneme in the primary cilium and control ciliary length. *J Cell Sci* 126(12): 2583-2594.
16. Xie Y F, Shi W G, Zhou J, Xian CJ, Chen KM, et al. (2016) Pulsed electromagnetic fields stimulate osteogenic differentiation and maturation of osteoblasts by up regulating the expression of BMPRII localized at the base of primary cilium. *Bone* 93: 22-32.
17. Kwon R Y, Temiyasathit S, Tummala P, Quah CC, Jacobs CR (2010) Primary cilium-dependent mechanosensing is mediated by adenylyl cyclase 6 and cyclic AMP in bone cells. *FASEB J* 24(8): 2859-2868.
18. Christensen S T, Clement C A, Satir P, Pedersen LB (2012) Primary cilia and coordination of receptor tyrosine kinase (RTK) signalling. *Journal of Pathology* 226(2):172-184.
19. Hoey D A, Kelly D J, Jacobs C R (2011) A role for the primary cilium in paracrine signaling between mechanically stimulated osteocytes and mesenchymal stem cells. *Biochem Biophys Res Commun* 412(1): 182-187.
20. Temiyasathit S, Tang W J, Leucht P, et al. (2012) Mechanosensing by the Primary Cilium: Deletion of Kif3A Reduces Bone Formation Due to Loading. *PLoS one* 7(3): e33368.
21. Lee K L, Hoey D A, Spasic M, Hammond HK, Jacobs CR, et al. (2014) Adenylyl cyclase 6 mediates loading-induced bone adaptation in vivo. *FASEB J* 28(3): 1157-1165.
22. Yan J L, Zhou J, Ma H P, Xian CJ, Chen KM, et al. (2015) Pulsed electromagnetic fields promote osteoblast mineralization and maturation needing the existence of primary cilia. *Mol Cell Endocrinol* 404:132-140.
23. Ma X N, Ma C X, Shi W G, et al. (2016) Primary cilium is required for the stimulating effect of icaritin on osteogenic differentiation and mineralization of osteoblasts in vitro. *J Endocrinol Invest* 40(4): 1-10.
24. Chatani M, Morimoto H, Takeyama K, et al. (2016) Acute transcriptional up-regulation specific to osteoblasts/osteoclasts in medaka fish immediately after exposure to microgravity. *Scientific Reports* 6: 39545.
25. Moorman SJ, Shorr AZ (2008) The primary cilium as a gravitational force transducer and a regulator of transcriptional noise. *Dev Dyn* 237(8):1955.
26. Dai Z, Tan Y, Yang F, Lina Qu, Hongyu Zhang, et al. (2011) Altered Actin Dynamics and Functions of Osteoblast-Like Cells in Parabolic Flight may Involve ERK1/2. *Microgravity Science and Technology* 23(1):19-27.
27. Sharma N, Kosan Z A, Stallworth J E, Berbari NF, Yoder BK (2011) Soluble levels of cytosolic tubulin regulate ciliary length control. *Molecular Biology of the Cell* 22(6): 806-816



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