Understanding CXXC Finger Protein 1 Action in Osteoblast Differentiation

Ahmed Al Saedi[1,2], Katelyn Healey[1], Molly Persky[1], David Maridas[3], Lijie Jiang[1], Vicki Rosen[3,4] and Diana L. Carlone[1,2,4]

[1]Division of Endocrinology, Boston Children's Hospital, Boston, MA, 02115, USA.
[2]Department of Pediatrics, Harvard Medical School, Boston, MA, 02115, USA.
[3]Department of Developmental Biology, Harvard School of Dental Medicine
[4] Harvard Stem Cell Institute, Cambridge, MA 02138, USA

Skeletal development, remodeling and regeneration are dependent on the activity of mesenchymal progenitor cells (MPCs). Recent data have implicated epigenetics in the regulation of MPC function with links to altered bone formation in both mice and humans. Precisely how epigenetics controls MPC activity and function, including the ability of MPCs to participate in bone formation, remains less clear. Here, we focus on CXXC Finger Protein 1 (CFP1), an epigenetic regulatory factor that mediates transcriptional activation through targeted H3-Lys4 methylation of chromatin. While CFP1 has been identified as a regulator of progenitor cell differentiation in other systems, its role in bone formation remains to be defined. To address this, we deleted *Cfp1* in MPCs using *Prx1*-Cre (cKO^{Prx1}), which resulted in halted chondrocyte differentiation, maturation and primary ossification, implicating this factor as a critical regulator of endochondral ossification. In addition, a decrease in intramembranous ossification within the calvaria was detected in mutant mice consistent with a role for CFP1 as a direct regulator of osteoblast differentiation. To investigate this further, we performed adenoviral deletion of *Cfp1* in bone marrow stromal cells (BMSCs) isolated from homozygous floxed-*Cfp1* mice. Quantitative RT-PCR (qPCR) analysis revealed a dramatic reduction in the expression of osteoblast markers (*Runx2*, *Osx*, *Col1a1*, *Ocn*) supporting a role for Cfp1 in bone formation. To further define its role in osteoblast differentiation, we generated Cfp1 knockout MC3T3-E1 cells using CRISPR/Cas9 and mice using *Osx1*-Cre (cKO^{Osx1}). Studies investigating if changes in osteoblast differentiation occur in the absence of Cfp1 are currently underway. In summary, our recent findings link CFP1 action to both endochondral and intramembranous bone formation.