Osteoblastic micro-RNAs regulate cortical bone formation

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Introduction

Micro-RNAs are short RNA molecules which regulate gene expression in various eukaryotic cellular processes. The RNAse III enzyme Dicer generates most of the micro-RNAs by processing premature miRNAs.

As of recent, many micro-RNAs have been reported to be involved in the regulation of gene expression during bone formation and remodeling. To study the role of osteoblast specific micro-RNAs, we have generated and characterized a novel mouse strain (Dicerosx). It is an inducible Cre strain. In this strain, by administering tamoxifen in Cre positive animals, Dicer can be inactivated in osterix expressing osteoblasts and cells differentiated out of them.

Using this novel strain, the effect of Dicer inactivation in mature osteoblasts was studied in the context of skeletal maturation and fracture healing.

Materials and Methods

Dicerosx strain was generated by crossing Dicer(Flox/Flox) strain (Harfe et.al) and OsxCreERT2 strain (Maes et.al). Animal studies were approved by the Finnish Animal Ethics committee. Standard genotyping procedures were carried out.

Two groups of animals were used in this study. Young mice (aged 3 weeks) and older mice (aged 5 weeks). Tamoxifen was administered to the animals for 3 days and then the animals were followed for 5 weeks. After 5 weeks, the animals were sacrificed and femora were analysed by MicroCT analysis.

Standardized tibial fractures were generated in Cre positive and Cre negative males. The mice were scarificed at d14 and the calluses were analysed by MicroCT.

Results

Fig 1: Schematic of the Dicerosx strain

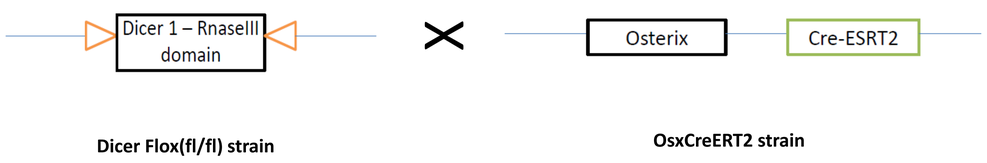


Fig 2: Cortical parameters in 8 week old females

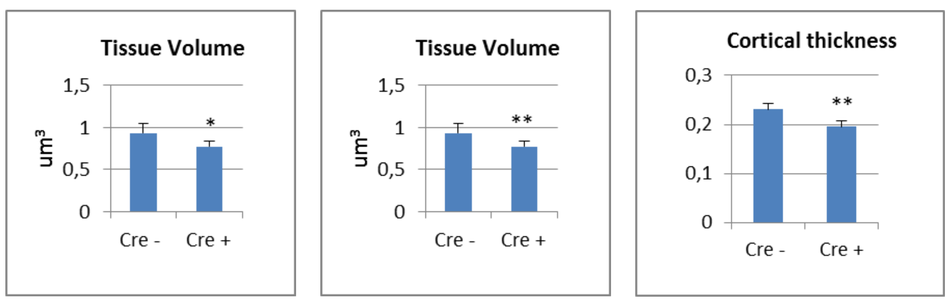


Fig 3: Cortical parameters in 8 week old males

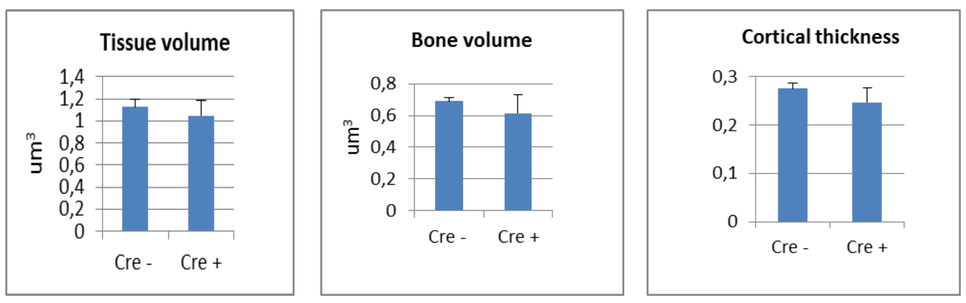


Fig 4: Cortical parameters in 15 week old females

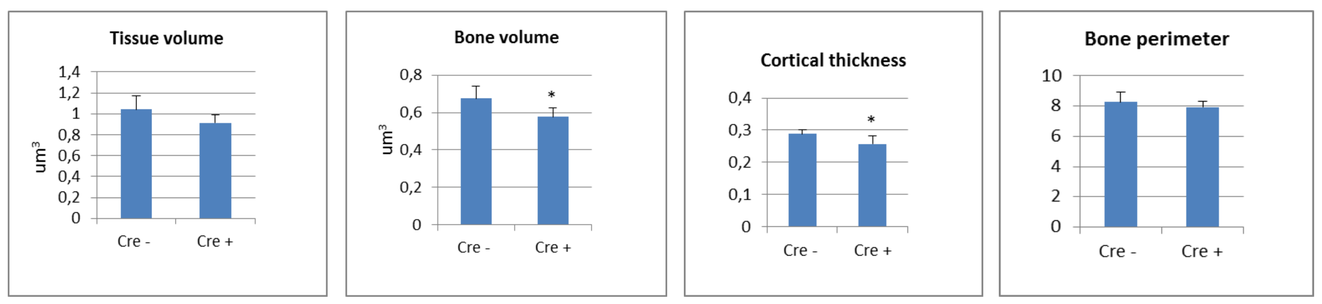


Fig 5: Cortical parameters in 15 week old males

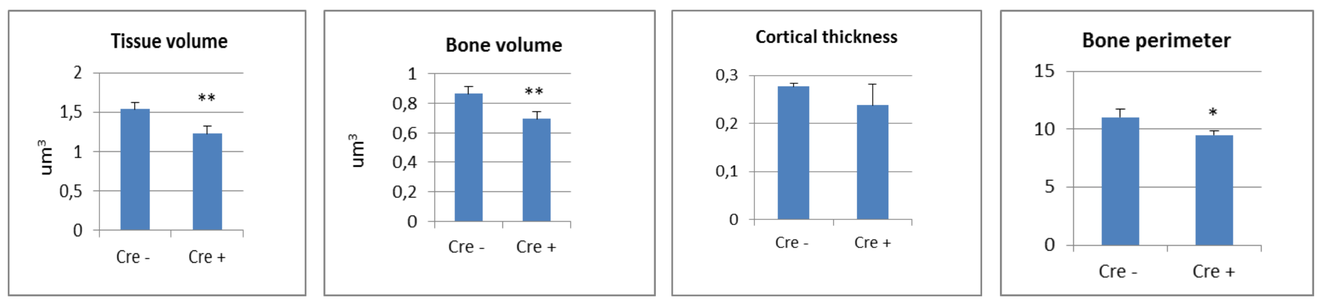
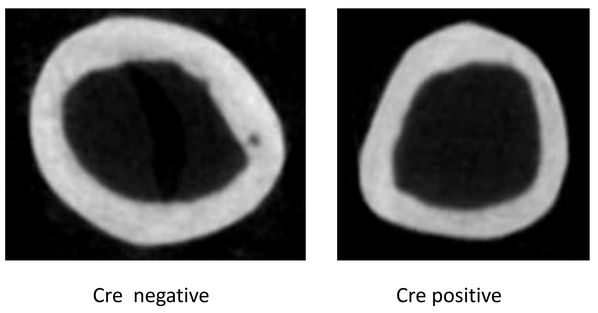
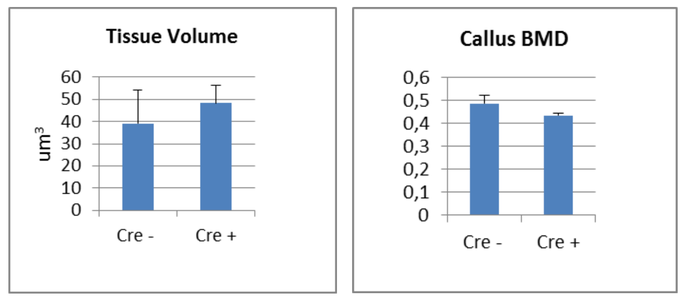


Fig 6: pictorial representation of cortical parameters in 15 week old animals.



Cre negative Cre positive

Fig 7: MicroCT analysis of Callus in Fracture healing



Conclusions

- **Significant Reduction in cortical bone volume and thickness in Cre + animals.**
- **Reduced Bone perimeter in 15 week old male mice but not in female mice.**
- **No significant difference in the amount of mineral deposited in the Cre+ callus as compared to Cre- controls.**
- **The average callus volume appeared to be increased in the Cre+ mice but the variation was high in both the groups. Further analysis is ongoing.**
- **Dicer processed microRNAs seem to be important in the regulation of cortical bone formation and in periosteal osteoblastic cells.**

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References

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