Effects of mechanical stimulation on bone formation

Katja Hrovat [1,2], Margalida Fontcuberta-Rigo [1,3], Pere Puigbò [1], Miho Nakamura [1]
[1]University of Turku, [2]University of Ljubljana Biotechnical Faculty, [3]University Rovira i Virgili

INTRODUCTION

- Osteoblasts regulate ossification and dysregulation can result in changes of mineralisation. It is important to understand osteoblast biology to help treat diseases as osteoporosis (1), which is the most common chronic metabolic bone disease, characterized by increased bone fragility (2). Due to aging population and longer life span, it is increasingly becoming a global epidemic (2).
- Increased mechanical stimuli (physical exercise) enhances bone mass and bone strength (3). Mechanical loading also promotes osteoblast differentiation, inhibits osteoclast formation, migration and adhesion *in vivo* (3).
- The aim of our research is to investigate the effects of mechanical stimulation on osteoblast differentiation in comparison to static conditions, since it is known that mechanical stress stimulates bone formation.

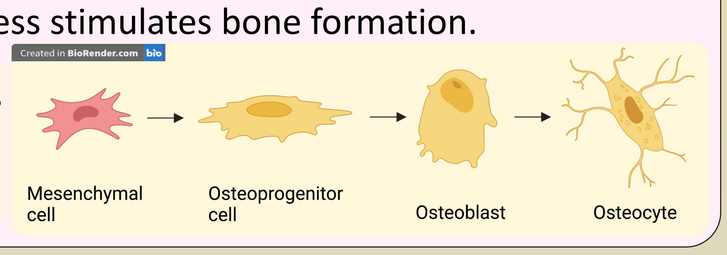
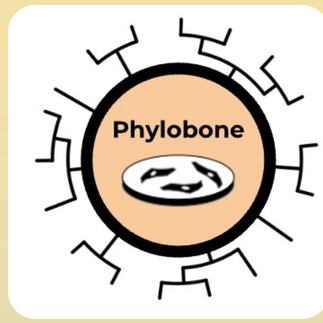
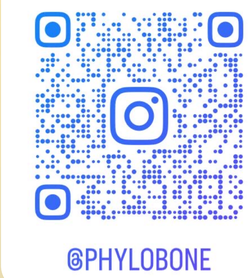


Figure 1: Differentiation process of osteoblasts (simplified), bone-building cells of mesenchymal origin. They differentiate from mesenchymal progenitor cells and can also differentiate in osteocytes, cells present in bone matrix (1).





This reserch is part of the Phylobone research project to study bone regeneration in the light of (biological) evolution. Visit us at phylobone.com

HIGHLIGHTS

- Mechanical stimulation promotes osteoblast differentiation in comparison to static differentiation.
- Two weeks differentiation enhances osteoblast differentiation in comparison to one week differentiation.
- Longer exposure to the mechanical stimulation and analysis of expression of other proteins still needs to be investigated in the future.

MATERIALS AND METHODS

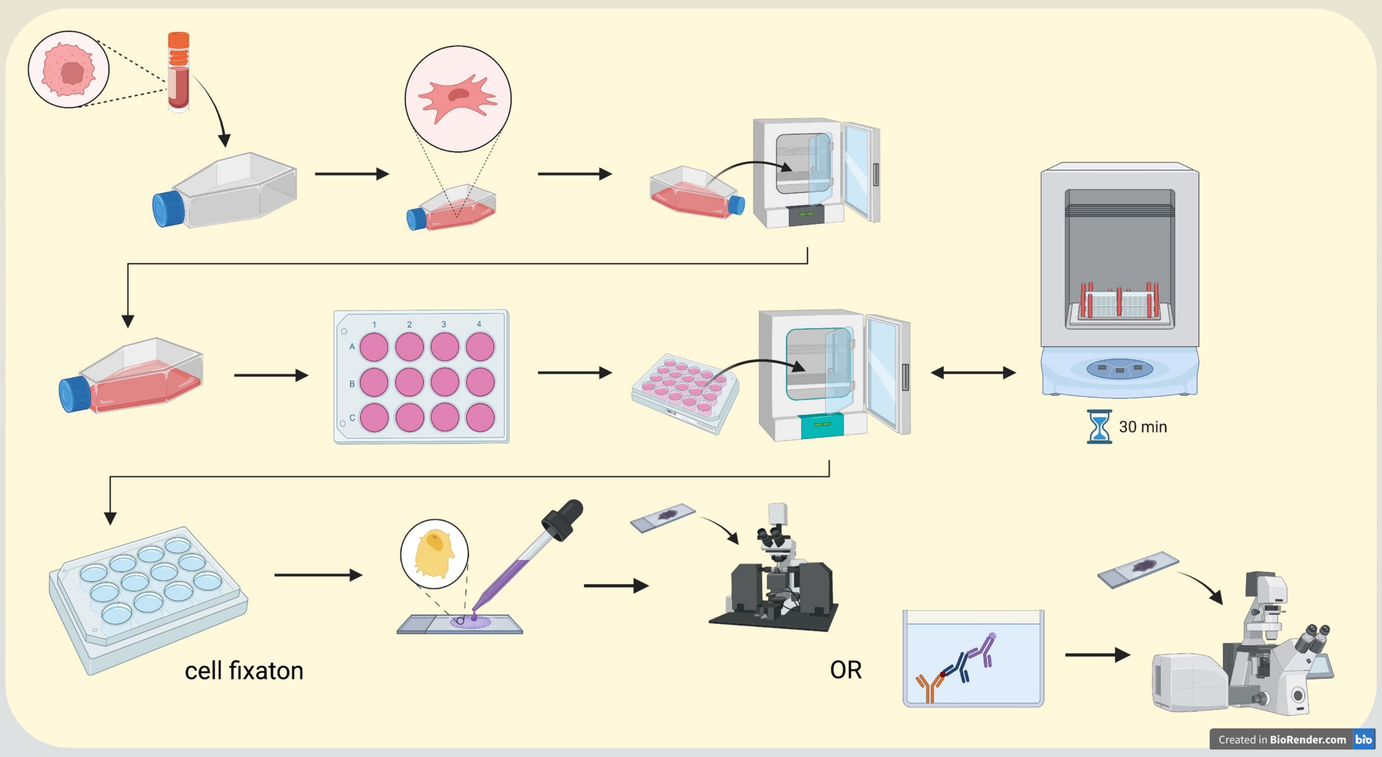


Figure 2: Cells were grown on culture flasks, seeded on 12-well culture plates and cultured until 100% confluency. The cells were differentiated by adding osteogenic differentiation factors. Parallelly, we divided plates into two groups: only treated with differentiation medium, and one treated also with mechanical stimulation. Cells were fixed at 1 and 2 weeks after starting differentiation and mechanical stimulation. After fixation, the analysis of the samples was performed.

RESULTS

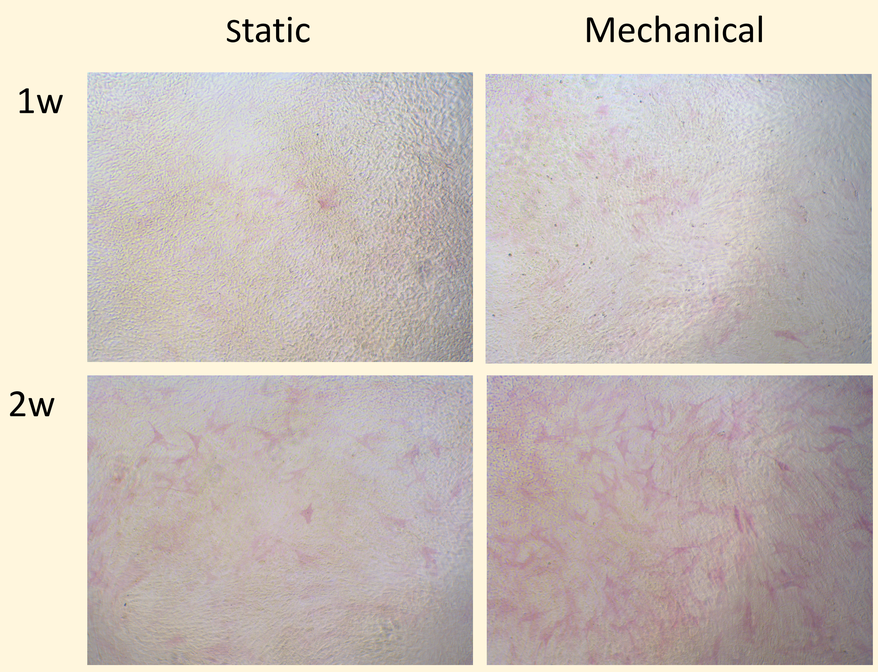


Figure 3: Results of the alkaline phosphatase (ALP*) show that after 2 weeks more cells differentiated into osteoblasts in comparison to 1 week. In addition, differentiation of the cells was accelerated under exposure to mechanical stimulation.
*ALP: molecular marker of osteoblast differentiation

RESULTS

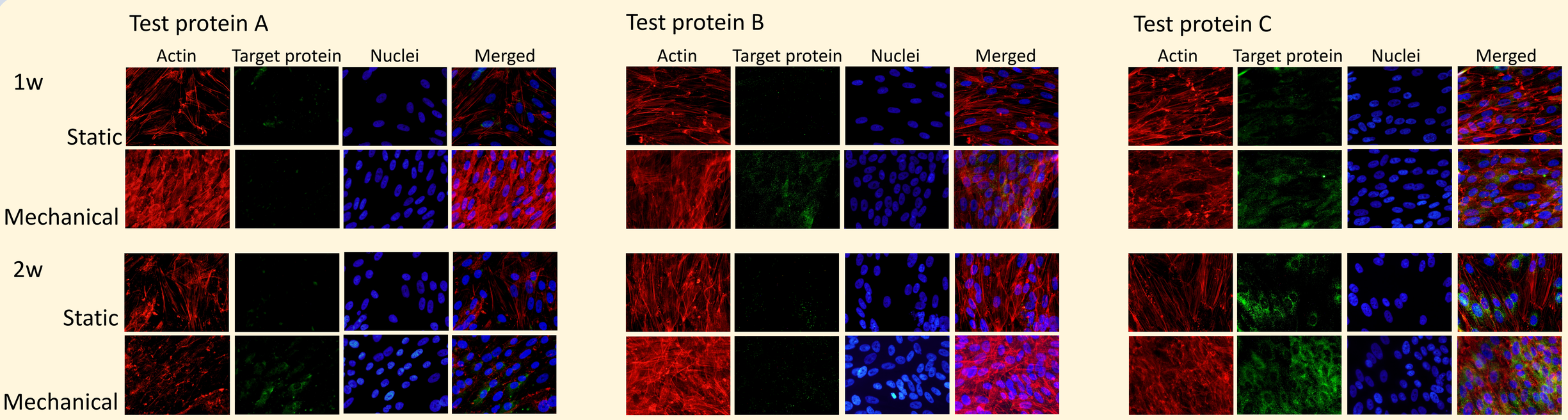


Figure 4: Results of the immunohistochemical analysis of three different proteins show higher expression of the proteins A and C after two weeks in comparison to one week and higher expression of protein B after one week in comparison to two weeks. The expression of all three proteins, important for bone formation, was observed to be higher with samples exposed to mechanical stimulation in comparison to static differentiation.

References

1. Rutkovskiy, A., Stensløkken, K. O., & Vaage, I. J. (2016). Osteoblast differentiation at a glance. *Medical science monitor basic research*, *22*, 95.
2. Sözen, T., Özışık, L., and Başaran, N. Ç. (2017). An overview and management of osteoporosis. *European journal of rheumatology*, *4*(1), 46.
3. Wang, L., You, X., Lotinun, S., Zhang, L., Wu, N., & Zou, W. (2020). Mechanical sensing protein PIEZO1 regulates bone homeostasis via osteoblast-osteoclast crosstalk. *Nature Communications*, *11*(1), 282.

Acknowledgement

- Turku Collegium for Science, Medicine and Technology
- Erasmus+ program
- Japan Society for the Promotion of Science (JSPS)
- Murata Science Foundation