Up-regulation of Alkaline Phosphatase Expression in Human Osteoblasts on Polycaprolactone
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Introduction
Polycaprolactone (PCL) is widely investigated as a promising scaffold for tissue engineered products (TEP) including those designed for bone reconstruction (1). In such an application it must work not only as a substitute for bone but also as a support for osteogenic cells transplantation. While investigating human bone derived cells (HBDCs) behaviour in direct contact with PCL in various forms, we noticed alkaline phosphatase activity being repeatedly enhanced in culture on PCL comparing to the standard culture. The aim of this study was to confirm this observation and to verify if this phenomenon is accompanied by the activation of other markers of HBDCs differentiation.

Materials and Methods
HBDCs isolated from bone fragments amputated during orthopaedic surgery (2), were cultured on the PCL samples and on the tissue culture polystyrene (control) and in a differentiation medium. Cell morphology was observed by scanning electron and optical microscopes. Alkaline phosphatase activity (ALP) was determined after 1, 7, 14, 21 and 28 days of culture. Expression of the selected genes (ALP, collagen type I, osteocalcin) was investigated by a quantitative Real-Time PCR.

Results
HBDCs morphology on PCL was found to be similar as compared to the culture on tissue culture polystyrene (Fig.1).

![Fig. 1. Human osteoblasts morphology on control and PCL.](image1)

Enhanced ALP activity in the culture on PCL comparing to the control was confirmed after all observation periods (Fig. 2). The mRNA expression of ALP gene was the highest after 7 days in both the experimental and control culture, and was higher on PCL after 1 and 7 days.

![Fig. 2. Alkaline phosphatase activity per cell in cultured on PCL and control.](image2)

Fig. 2. Alkaline phosphatase activity per cell in cultured on PCL and control.

Surprisingly, osteocalcin as well as collagen type I demonstrated lower level of mRNA expression in cells cultured on PCL after all observation periods.

Discussion and Conclusions
Our results show that PCL has strong stimulated effect on gene expression and protein activity of the ALP. This is not accompanied by an enhanced activation of collagen I or osteocalcin. This finding is important for our understanding of the PCL influence on osteoblasts and it calls for further study aimed to analyze the mechanisms responsible for PCL up-regulation of ALP expression.

References

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Disclosures
Authors have nothing to disclose.