Magnesium-Yttrium Alloys for Treating Anterior Cruciate Ligament Injuries

Huinan Liu, Daniel Perchy, Savio Woo.
Musculoskeletal Research Center, Department of Bioengineering, University of Pittsburgh.

Statement of Purpose: The anterior cruciate ligament (ACL) is the most frequently injured knee ligament. ACL ruptures have a limited capacity for healing because its fibroblasts have a low capability to initiate a strong healing response [1]. As a result, surgical repair or reconstructions are often necessary to restore knee joint stability. Following ACL reconstruction, the natural soft tissue-to-bone transition, consisting of distinct zones, often fails to develop. This can lead to altered loading patterns in the tissue/insertion site, thus, increasing the risk of re-injury. Recent studies demonstrated that porcine small intestine submucosa (SIS) could provide active growth factors to enhance ligament healing [2]. However, the mechanical properties of SIS-based scaffolds are several orders of magnitude lower than natural bone and ligaments, which could not provide initial mechanical stability and thus impede proper tissue remodeling and healing due to the lack of loading. Magnesium (Mg) alloys have greater mechanical strength than polymer-based biomaterials and can be metabolized in the body. Therefore, Mg alloys may provide initial mechanical support to allow early loading of healing tissues and subsequently improve tissue quality. However, the degradation properties of current Mg alloys are far from satisfactory for ligament-bone tissue engineering due to the fast early-stage degradation and the release of a large amount of hydrogen gas in a short time, which may have adverse effects on tissue healing, particularly in intra-articular environment. To address these issues, Mg-Yttrium (Mg-Y) alloy was developed. Its degradation properties and cytocompatibilities with bone marrow derived stem cells were investigated in this study.

Methods: Mg-4 wt.%Y alloys were cast at GKSS research center (Germany). The cast ingot was then cut into 10x10x0.25 mm discs for in vitro degradation and cell culture study. The discs were sterilized under UV radiation for 24 hours. Immersion method was used to evaluate in vitro degradation. The pH of the media and the weight change of Mg-Y alloys were monitored. Bone marrow derived stem cells (BMSCs) were harvested from a skeletally mature goat, purified and cultured in Dulbecco's Modified Eagle Medium (DMEM) supplemented with 10% fetal bovine serum (FBS) and 1% penicillin/streptomycin (P/S) under standard cell culture conditions (37 °C, 5% CO_2/95% air, humidified environment). The BMSCs (PN=1) were seeded onto Mg-Y alloys at a density of 40,000 cells/cm^2 and incubated for 24 hours. The cells were then washed using phosphate buffered saline (PBS) to remove non-adherent cells, fixed using 10% formaldehyde, stained using DAPI and counted under a fluorescence microscope (Nikon T200). The percentage of BMSCs adhered on Mg-Y alloys and tissue culture polystyrene (TCPS) were calculated.

Results: The pH of Mg-Y alloy culture was measured at the prescribed time points (Figure 1a) and the mass of Mg-Y samples was measured after culturing for the prescribed time periods and dried (Figure 1b). Mg-Y alloys degraded much slower in DMEM and DI water compared to PBS. High Cl^- concentration in PBS may contribute to faster degradation. Figure 2 showed the percentage of BMSCs adhered directly on Mg-Y alloys and on the surrounding tissue culture polystyrene (TCPS). The percentage of BMSC adhesion on Mg-Y alloy is slightly lower than TCPS.

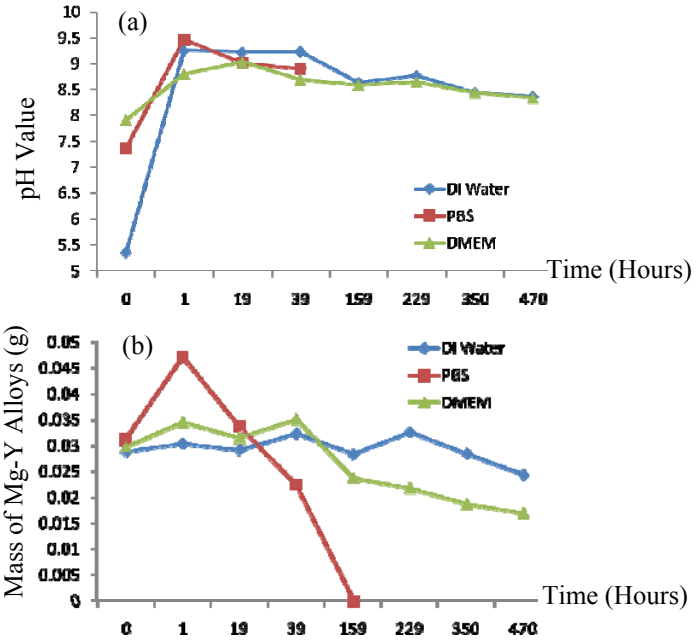

Figure 1: (a) the change of pH value of Mg-Y alloy culture with time. (b) the mass of Mg-Y alloys after the prescribed culture time.

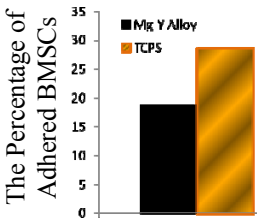

Figure 2: The percentage of BMSC adhered on Mg-Y alloy and on the surrounding TCPS after 24 hours of cell culture.

Conclusions: The in vitro degradation of Mg-Y alloys is dependent on the culture solutions. The high pH in cell culture induced by Mg degradation has negative effects on BMSC adhesion. Degradation and biocompatibility properties of Mg alloys need to be further improved for orthopedic applications.

Acknowledgment: We would like to thank NSF ERC for funding and Drs. Hort and Kumta for Mg-Y alloys.

References: [1] S. L. Woo, T. M. Vogrin, S. D. Abramowitch, The Journal of the American Academy of Orthopaedic Surgeons 2000, 8, 364. [2] R. Liang, S. L. Woo, Y. Takakura, D. K. Moon, F. Jia, S. D. Abramowitch, J Orthop Res 2006, 24, 811.