Magnesium Plasma Modified Bone Allograft for Large Bone Defect Treatment

Wenhao Wang (1)(2), Hoi Man Wong (1), Paul K. Chu (3), Frankie K.L. Leung (1)(2), Kenneth M.C. Cheung (1), Kelvin W.K. Yeung (1)(2)

- 1: Department of Orthopaedics and Traumatology, the University of Hong Kong, Pokfulam, Hong Kong.
- 2: Shenzhen Key Laboratory for Innovative Technology in Orthopaedic Trauma, the University of Hong Kong Shenzhen Hospital, Shenzhen, China.
- 3: Department of Physics and Materials Science, City University of Hong Kong, Kowloon, Hong Kong, China.

*Corresponding authors: wkkyeung@hku.hk

Keywords: Bone allograft, Magnesium ions, Plasma ions immersion implantation and deposition (PIII&D), Surface modification

Abstract: Bone allograft is the most widely accepted approach in treating patients suffering from large segmental bone defect regardless of the advancement of synthetic bone substitutes[1, 2]. However, the long-term complications of allograft application in term of delayed union or even nonunion were reported due to the stringent sterilization process prior to clinical implantation[3, 4]. Our previous studies demonstrated that the incorporation of magnesium ions (Mg²⁺) into biomaterials could significantly promote the gene up-regulation of osteoblasts and new bone formation in animal model[5, 6]. Hence, our group has proposed to implant Mg²⁺ into bone allograft by adopting plasma ions immersion implantation and deposition (PIII&D) approach.

The decellularization and gamma irradiation process were performed on bovine bone allograft prepared in 1 x 1 x 0.1cm. Subsequently, a thin layer of magnesium coating was prepared by using magnesium PIII&D technique. The surface morphology, elemental chemical and depth of the samples were examined by scanning electron microscopy (SEM), energy dispersive x-ray spectroscopy (EDS) and x-ray photoelectron spectroscopy (XPS), respectively. The cytotoxicity, cell morphology, proliferation and alkaline phosphatase (ALP) expression of magnesium-enriched bone allografts were evaluated by culturing human immortalized mesenchymal stem cells (hTMSC).

With the adjustment of implantation parameters e.g. voltage and time, the magnesium composite layer had been successfully established on the surface of allogenic bone. The thickness of the composite layer ranged from 500nm to ~800nm. The surface topography of allograft flattened and the microstructure of collagen fibers structure was also changed after plasma treatment. The cells elongated on the surfaces of plasma treated and untreated samples though. The cell viability in magnesium plasma modified allograft was significantly higher than that of the control. The ALP gene expression of hTMSCs in the group of PIII&D treated samples was highly up-regulated. However, there was no significant difference between plasma treated group and the control in terms of cell proliferation rate.

The magnesium plasma ion immersion implantation and deposition technique has been first applied to bone allograft. Indeed, this well-developed technology is able to alter the surface properties of materials, while the bulk mechanical properties maintained[7]. The *in vitro* results suggested that the modified allografts could up-regulate the ALP gene activity and cell viability of htMSCs. Hence, it is believed that the original mechanical properties of bone allograft should be maintained, when its bioactivity enhanced. Further experiments including in-depth molecular cell biological and animal studies should be initiated prior to clinical application.