INTRODUCTION:

Porous scaffold materials with pore size between 300-500 µm are proven osteoconductive and widely used in orthopaedic applications. To develop porous scaffold materials with osteopromotive function is even desirable for treatment of difficult conditions. Recently, we identified a novel phytoestrogenic molecule Icaritin for effective prevention of osteoporosis and osteonecrosis [1, 2]. The present study aimed to produce an osteopromotive porous composite scaffold material by incorporating Icaritin into standard poly(L-lactide-co-glycolide)/tricalcium phosphate(PLGA/TCP) setting based on a computer-controlled production technology and fabricated at low-temperature to preserve the bioactivity of Icaritin. The osteopromotive effects were evaluated based on the structural and mechanical properties of PLGA/TCP/Icaritin composite scaffold and the slow release of Icaritin form the composite scaffold in vitro. The biocompatibility of scaffold will also be evaluated by using bone marrow mesenchymal stem cells (BMSCs).

METHODS: The pure PLGA/TCP scaffold was used for comparison (n = 6). Macro and micro structure of PLGA/TCP/Icaritin composite scaffold were evaluated by macroscopic observation and scanning electron microscope (SEM). Surface composition was analyzed by X-ray energy dispersive spectrometry (EDS), and the 3-D structure in terms of porosity and its interconnectivity was quantified by micro computer tomography (micro CT). Water absorption and the mechanical properties were also evaluated apart from quantification of Icaritin concentration in the composite scaffold by high-performance liquid chromatography (HPLC). After being isolated from iliac crest of rabbits, the BMSCs were seeded on the PLGA/TCP/Icaritin composite scaffold and the seeding efficiency, BMSCs proliferation, and alkaline phosphatase (ALP) activity were examined.

RESULTS: The result showed that there was no significant difference in porosity, water absorption and surface composition of the scaffolds after incorporating Icaritin into the PLGA/TCP composite scaffold as compared with pure PLGA/TCP scaffold. Both micro- and macropores were found interconnected with an average pore size of 420µm and 2-15µm respectively in both groups (Fig.1). The mechanical properties of PLGA/TCP/Icaritin scaffold, including Young's modulus (44.6±2.19MPa), compressive strength (2.3±0.51MPa), and energy to failure (29.6±3.30(10^2)J) in an axial compression mode were comparable to those of PLGA/TCP scaffold, yet, about 4.6, 2.3 and 9.0 times higher in axial compression than those of lateral compression in both groups (p<0.05 for all). The Icaritin content incorporated into the scaffold remained unchanged and showed a slow-release pattern tested in vitro, with about 35% release of Icaritin over a period of 7 weeks. The seeding efficiency and proliferation of BMSCs and ALP activity of PLGA/TCP/Icaritin group were superior to those of pure PLGA/TCP group.

DISCUSSION: In conclusion, this is the first time to incorporate a phytoestrogenic molecule Icaritin into a standard PLGA/TCP scaffold material to form a composite porous scaffold material with functional and interconnected pores. The osteopromotive nature of this novel composite scaffold material was demonstrated by slow release of phytoestrogenic Icaritin, suggesting favorable environment for attachment, proliferation and osteogenic differentiation of BMSCs in vivo. These novel composite porous scaffold materials implied its potential for a wide range of orthopaedic applications.

REFERENCES:

Fig.1. Comparison of PLGA/TCP(A) and PLGA/TCP/Icaritin(B) scaffolds. Macroscopic observation of scaffolds: (A1) White PLGA/TCP scaffold; (B1) Light yellowish PLGA/TCP/Icaritin scaffold due to homogenously distributed Icaritin (Icaritin powder is yellow in color before mixing into PLGA/TCP). 2D structure of the composite scaffolds by microCT: A2 and B2 (Arrows point to the maropores interconnected with neighbors). Surface structure of the composite scaffolds by SEM: A3 and B3 (50x, arrows point to macropores, frame magnified to A4 and B4); A4 and B4 (5000x, arrows point to micropores).

ACKNOWLEDGEMENT: The project was supported by Hong Kong Innovation and Technology Support Programme (Tie 2) GHP/001/08.