Introduction: An ideal bone graft should have osteoconductive, osteoinductive properties to offer the framework for the formation of new bone. In addition, biodegradable materials have been widely used as artificial bone graft, such as poly(lactic) acid (PLA). To develop bone graft materials used for different surgical applications, it is proposed to prepare bone grafts materials with various mechanical strengths Preparation of polymer/ceramic composite materials could be a solution. Hydroxyapatite (HAp) or β-tricalcium phosphate (TCP) based PLA composites have shown osteoconductivity for bone growth. It is anticipated that the independent control of PLA based bone graft properties can be achieved by appropriately adjusting the composition of composite materials. Among which, adjustment of the formula for PLA composites is the simplest method to manufacture different mechanical properties. Several techniques of the scaffold have been used to fabricate polymers into porous matrices for tissue engineering applications, including solvent-casting/particulate leaching, phase separation, fiber extrusion and fabric forming processing [1]. The solvent-casting/particulate leaching and phase separation approaches require the use of organic solvents. Residues of organic solvents, which remain in these polymers after processing, may damage transplanted cells and nearby tissue and inactivate many biologically active factors that one might wish to incorporate into the polymer matrix for controlled release. Our approach is to use salt-leaching method [1] to produce PLA/TCP porous 3D scaffold, and different mechanical strengths with different PLA/TCP ratios. An rabbit animal model was carried out to prove the application of PLA/TCP porous scaffold.

Materials and Methods: PLA with an inherent viscosity of 0.55 dl/g, obtained from Bio Invigor Corporation, Taipei, Taiwan was mixed with tri-calcium phosphate (TCP) particles (obtained from Sigma (NO. 04238)) at room temperature. The ratios of PLA/TCP were 10/0, 9/1, 8/2, 7/3, and 6/4 by weight. NaCl particles (104-250 μm) were added into PLA/TCP mixtures. The ratio of (PLA/TCP)/NaCl was 2/8 by weight. Subsequently, the composite was hot-pressed uniaxially for 10 min at 50 degree C under a pressure of 13.79 MPa. Cylindrical specimens (5 mm diameter, 6 mm length) of the (PLA/TCP)/NaCl composites were prepared for the following tests. The composite materials were immerced into dH2O 72 h, in order to leach NaCl. After 72 h later, 80% porosity of PLA/TCP porous scaffold was obtained. Compressive strength of the composite were measured with the guidance of ASTM D695-02a by MTS at a loading rate of 0.5 mm/min [2]. For animal study, six female New Zealand white rabbits (2.5 kg) were anesthetized with 0.2-0.3 ml/kg Zotil (the mixer of ketamin and comblene, Virbac, Taipei, Taiwan) and 0.25 ml/kg xylazine by intramuscular injection. The rabbit was placed in dorsal recumbency and a midline incision was made from the femur with a #10 scalpel blade. Then the electric drill (5 mm diameter) was applied to create a cavity (5 mm diameter, 6 mm length) on the femur near the knee joint. The rabbits were sacrificed after 4 weeks later. The femoral bone of the injured site was removed and fixed in 10% formalin solution. The thin sections of the bone tissues were examined after staining with hematoxylin-eosin.

Results: Fig. 1 shows the SEM images of the cross-section of the scaffold. It is seen that the average size of the hole is about 200 μm. Fig. 2 shows the H&E stained of the tissues surrounding the operated area of rabbits. The ultimate compressive strength of the PLA/TCP composite materials were measured. For example, the compressive strength of PLA/TCP=7/3 composite scaffold is 2.5 MPa. It is observed that the ultimate compressive strength increases with the increasing amount of TCP.

Discussion: Organic solvents have been widely used to dissolve PLA to prepare PLA composites or scaffold. However, the cytotoxicity of the solvent prohibits being used in medical applications. By a salt-leaching procedure used in this study to manufacture the PLA/TCP porous scaffold, the possibility of residue solvent can be eliminated.

It is observed that the size of the scaffold can be controlled by the particle size of NaCl powders added in the manufacturing process. The ratio of (PLA/TCP)/NaCl=2/8 by weight demonstrated that the scaffold's porosity is 80%. In Fig.3, it is seen that a few osteoblasts grew around the hole of scaffold. The osteoblast grew more fast form the PLA/TCP porous scaffold than control. We could see the circle shape from the section. It is observed that with higher PLA/TCP ratio, the higher ultimate compressive strength. It is believed that the high mechanical strength of tricalcium phosphate ceramics provide the reinforcement effect in the composite materials. Thus, the mechanical properties such as strength and elastic modulus of PLA can be controlled by the amount of mixed TCP particles. On the other hand, the modulus of elasticity of the composites was improved effectively with increasing TCP content. Further increase of TCP leads to the decrease of elastic modulus at the PLA/TCP ratio of 7/3. The effect of TCP particle size on the mechanical properties of the scaffold should be also discussed in the future. After the relationships between composite dosage and material properties of PLA scaffolds are established, PLA composite materials with optimum mechanical strength and degradation rate can be designed and synthesized for specific medical implants.