# Controlled acetylated chitosan scaffolds for tissue engineered artificial dermis

Sae-Hwan Lim, Chun Ho Kim\*, Young Ju Choi, Seung Jae Lee\*,
Hyun-Sook Park\*, and Young Sook Son\*

Modern Tissue Technologies Inc.,
Laboratory of Tissue Engineering Korea Cancer Center Hospital\*

#### Introduction

In the skin, wound repair is a complex process involving an integrated response by many different cell types controlled by a variety of growth factors. During the initial inflammatory phase fibroblasts start to enter the wound where they synthesize and later remodel new extracellular matrix material, of which collagen is the main component. Chitosan has known promise as one of structural biocompatible biomaterials for a number of tissue engineering applications. It has the wound healing effect through the induction of cytokine production and activation of inflammatory cells in animals. In the case of normal conditions of use, clinical tests carried out in order to promote chitosan-based biomaterials do not report any inflammatory or allergic reaction following implantation, injection, topical application or ingestion in the human body. However, It has been reported that fibroblasts might strongly adhere on chitosan film. As a result, fibroblasts do not proliferation on chitosan film although they remain alive, in comparing with keratinocytes. The limited fibroblast proliferation could hinder the wound repair.

### Materials and methods

Chitosan with 85 % degree of deacetylation were dissolved in acetic acid to give 1.5 w/v % solutions. Chitosonium acetate films were prepared by covering the cover glass in 24 culture plate, followed by solvent evaporation and drying at room temperature for 2 weeks. Chitosonium acetate scaffolds were prepared by freezing and lyophilizing chitosan solutions in pre-cooled, flat bottomed molds. To prepare the heat treated chitosan derivatives, crude chitosonium acetate films and scaffolds were heated in the oven at 70 -

110 °C under *in vacuo* at various time intervals. Heat treated chitosan derivative was produced, and the structural changes in chitosan derivatives were confirmed with FT-IR, and XRD. The resulting films and scaffolds were examined the effects of the heat treatment on the proliferation of human dermal fibroblasts (HDFs, derived from newborn foreskin and passage between 2 and 6) *in vitro*.

#### Results and discussion

Resulted derivatives did not dissolve in aqueous diluted acetic acid. Mechanical properties of heat treated chitosan derivatives were improved than chitosan especially in wet. Heat is often employed to facilitate polymer processing and sterilize pharmaceutical and medical products. But the rigid heat treatment could lead to crosslink of chitosonium acetate, reaction intermediate. This might induce reduced solubility and improved mechanical properties. The in vitro enzymatic degradation rates of the chitosan derivatives were faster than that of chemically acetylated chitosan scaffolds as well as chitosan itself. Besides HDFs proliferation in heat treated derivatives were improved than those of chitosan scaffold. In conclusion, heat treated chitosan scaffolds may be considered as very good biomaterials not only for cell cultivation but also for tissue engineered artificial dermis, guided tissue regeneration and soft tissue augmentation.

## References

- 1. Kozo Ogawa, Effect of heating an aqueous suspension of chitosan on the crystallinity and polymorphs , Agric. Biol. Chem., 55(9), 2375-2379 (1991)
- 2. Lim LY, Wan LSC, Heat treatment of chitosan films, Drug Development and Industrial Pharmacy, 21(7), 839-846 (1995)
- 3. Ackah Toffey. et. al., Chitin Derivatives. I. Kinetics of the heat-induced conversion of chitosan to chitin, J. App. Polymer Science, 60, 75-85 (1996)
- 4. Gampimol C, et. al., Moist heat treatment on physicochemical change of chitosan salt films, International Journal of Pharmaceutics, 232, 11-22 (2002)
- 5. Claire Chatelet. et. al., Influence of the degree of acetylation on some biological properties of chitosan films, Biomaterials, 22, 261-268 (2001)