OMICS Group Conferences
Accelerating Scientific Discovery

3rd World Congress on
Cell Science & Stem Cell Research
November 20-22, 2013 DoubleTree by Hilton Baltimore-BWI Airport, MD, USA

A simplified isolation method for epithelial stem cells and its use for fabrication of the chitosan based skin substitute

Mohd Hilmi Abu Bakar, Ahmad Sukari Halim and Asma Hassan
University Sains Malaysia, Malaysia

There are challenges among scientist as hair follicle stem cells (HFSCs) derived from a human hair follicle remain poorly expanded in culture medium and still requires a coating agent for proliferation in culture vessels. In the present study, the isolation, characterization and differentiation of HFSCs for chitosan based skin substitute application were performed. Scalp, 1 cm by 1 cm was collected from patients for neurosurgery or wound suturing procedure. Viability of HFSCs in defined keratinocytes serum free medium (KSFM) in coated plates and CnT07 medium in non-coated plates were compared. Differentiation of HFSCs into epidermis was carried out using CnT02-3D medium with confirmation by mouse monoclonal antibodies against human cytokeratin 6 and human involucrin. Co-culture between fibroblasts and HFSCs into chitosan were established at a density of $3x10^6/cm^2$ and $1x10^6/cm^2$ respectively. After 10 days of culture, the HFSCs became confluent and applicable for skin substitute application. Viability analyses using presto blue showed that the HFSCs culture in CnT07 with uncoated plates were significantly higher than HFSCs culture in KSFM with coated plates ($P<0.05$). Molecular characterization of HFSCs via flow cytometry analysis demonstrated that the percentage of HFSCs expressing CD200 and K15 were 65.20±3.16 and 72.07±6.62 respectively. The population doubling time of HFSCs was 21.48±0.44 hours in CnT07-uncoated plates and 30.73±0.75 hours in KSFM-coated plates. After three weeks, chitosan based skin substitute was harvested for treating impaired wound healing in animal model. This method is a simple technique for HFSCs isolation and raises hope for the *de novo* skin or epidermal substitute preparation.

hilmiab@yahoo.com