Histological Study of an Experimentally Constructed Decellularized Rat Lung as a Model of Whole Organ Three-Dimensional Natural Scaffold

Thesis

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List of Abbreviations

Abb.	Full term
3D	Three Dimensional
Arg	Arginine
bFGFs	Basic Fibroblast Growth Factors
CHAPS	Cholamidopropyl dimethyl Ammonia pro Panesulfonate
CHOH-CHOH	Carbon to Carbon bond
dECM	Decellularized Extracellular Matrix
dH2O	Deionized Water
ECM	Extracellular Matrix
EDTA	Ethylenediaminetetraacetic acid
EGTA	Ethylene Glycol Tetra-acetic Acid
GAGs	Glycoseaminoglycans
HCL	Hydrochloric Acid
HUVECs	Human Umbilical Cord Endothelial Cells
Lys	Lysine
MHC	Major histocompatibility complex
MVECs	Micro Vascular Endothelial Cells
PAS	Periodic acid–Schiff
PBS	Phosphate-buffered saline
PCO	Pressure of Carbon monoxide
PO	Pressure of Oxygen
SB-10	Sulfobetaine-10
SB-16	Sulfobetaine-16
SD	Standard Deviation
SDS	Sodium Dodecyl Sulfate
SEM	Scanning Electron Microscope
TEM	Transmission Electron Microscope

Abstract

Introduction: With the increase of end stage lung diseases and the great problems facing lung transplantation tissue engineering become a promising solution. The first step in lung engineering is to obtain a 3D Extracellular matrix lung scaffold via decellularization. Decellularization aims to remove cells from tissue ultrastructure while preserving the mechanical and biological properties of the tissue. Intact ECM provides critical cues for differentiation and migration of cells that are seeded onto the organ scaffold.

Objectives: This study aimed to obtain an intact and well-preserved ECM lung scaffold by decellularization of rat lungs.

Methods: Decellularization of lungs of seven Wistar rats was achieved by perfusing detergents through the pulmonary artery. The resultant scaffolds were fixed and analyzed histologically.

Results: It was found that the decellularization process effectively removed the cellular and nuclear material while retaining native the 3D ECM of lung tissue. The architecture of the collagen and elastic fibers networks were preserved as comparable to the native lungs. Furthermore, the basement membranes of the bronchiolar and interalveolar septa were intact.

Conclusions: This methodology is expected to allow decellularization of human lung tissues and permits future scientific exploration in tissue engineering.

Keywords: extracellular matrix , lung, decellularization, rat.

INTRODUCTION

Recently, end stage pediatric and adult lung diseases are increased dramatically. This represent one of the great problems facing healthcare systems worldwide as human lungs do not generally repair or regenerate. These diseases include chronic lung diseases of prematurity (bronchopulmonary and alveolar capillary dysplasia), chronic obstructive pulmonary disease which includes emphysema and chronic obstructive bronchitis, idiopathic pulmonary fibrosis, cystic fibrosis and idiopathic pulmonary arterial hypertension **(Minino et al., 2011; Robert and Gabriel, 2011).**

The only current viable option for patients with end stage diseases is lung transplantation as pharmacological intervention helps only to decrease the symptoms at the various disease stages **(Mark et al., 2014)**.

But unfortunately, lung transplantation still has many problems, for example to choose a suitable candidate you must exclude those with recent malignancy, active infection with virus B or C with proved liver damage, proved extrapulmonary vital organ dysfunction and some HIV positive recipient **(Robert and Gabriel., 2011)**.

Another problem facing lung transplantation is waiting for a suitable donor. The most common and available donor is brain-dead organ donor, but for sorrow usually there is insults

that can happen to the lung during the hospital admission of the donor. Such as volume overload, acute lung injury, contusion, aspiration and pneumonia. So, the gap between the number of patients who have received a transplant and those who are in the waiting list has become wider than ever **(Orens and Garrity, 2009).**

Also, there are many complications after lung transplantation. This include primary graft dysfunction (acute graft rejection with noncardiogenic pulmonary edema in the first 72 hours post transplantation), air way ischemia, infections that may be viral or bacterial and considered a leading cause of early and late deaths, acute rejection and chronic graft dysfunction **(King-Biggs et al., 2003).**

Over the years, the researchers kept thinking in alternative sources of organs, they have investigated xenotransplantation and tissue engineering.

"Xenotransplantation is the transplantation of living cells, tissues or organs from one species to another; thus far, organ transplant from animals to humans has been impossible because of the overwhelming rejection and the risk of transmitting animal viral diseases to humans" **(Baptista et al., 2009).**

Tissue engineering is a relatively new field that uses living cells, biocompatible materials, and suitable biochemical