Project number 68

Evaluation of Vasculotide, an Angiopoietin-1 mimetic, to attenuate the development of pulmonary edema during clinical ex vivo lung perfusion

［1］Research group

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Expenditure report of research funds：
Consumables 20,000 YEN
Travel cost 180,000 YEN

［2］Research setup

Lung transplantation (LTx) is a lifesaving therapy for patients with end-stage lung diseases. However, the number of patients waiting for lung transplants greatly exceeds the number of donors available. A novel strategy to overcome the shortage of lungs available is ex vivo lung perfusion (EVLP). EVLP can increase the number of donor lungs available in 2 important ways: 1) better evaluation of marginal organs, and 2) treatment and repair of injured organs [Cypel M, et al. N Engl J Med. 2011; 364:1431-40, Cypel M, et al. J Heart Lung Transplant. 2008;27:1319-25].

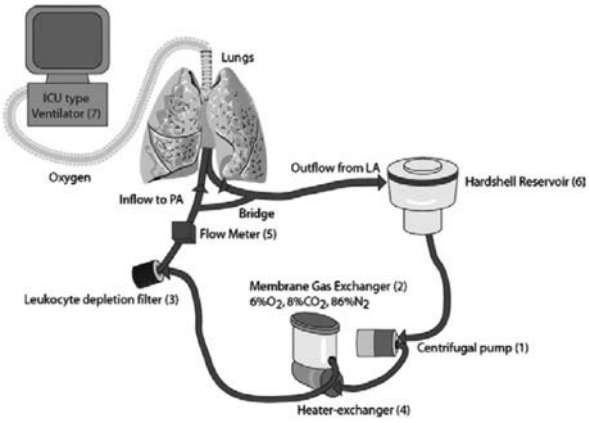

Figure 1. Ex vivo lung perfusion

The perfusion solution (STEEN solution) used during EVLP contains human serum albumin to provide an optimal colloid osmotic pressure and dextran to coat and protect the endothelium from excessive leucocyte interaction. Despite this optimization, injured lungs tend to develop pulmonary edema during the perfusion time. Pulmonary edema is one of the terminal conditions of lung failure, and also the most common reason for lungs to be declined for LTx after EVLP. Thus, strategies to mitigate the development of pulmonary edema are attractive as EVLP additives.

Angiopoietin-1 was isolated in the 1990's and has been known to have important roles in vascular development and angiogenesis [Davis S, et al. Cell. 1996;87:1161-9, Suri C, et al. Cell. 1996;87:1171-80]. Tie-2 is a tyrosine-protein kinase receptor for Angiopoietin-1, expressed almost exclusively in vascular endothelium. Angiopoietins and Tie families have primary roles in the late stages of vascular development, and also in adult vascular remodeling and stabilization. Binding of Angiopoietin-1 to Tie-2

regulates endothelial cell survival, vascular inflammation, vascular permeability, and angiogenesis. Vasculotide is an Angiopoietin-1 mimetic which is a Tie-2 receptor agonist which was initially developed to treat acute kidney injuries. Since then, it has also demonstrated positive therapeutic effects in many different disease settings associated with vascular dysfunction [Sugiyama MG, et al. Sci Rep. 2015;5:11030, Rübig E, et al. Sci Rep. 2016;6:22111, Thamm K, et al. World J Transplant. 2016;6:573-82]. The primary mechanism of Vasculotide is to activate the Tie-2 signalling pathway to exert anti-inflammation, anti-permeability, anti-permeability, and anti-apoptotic effects. We hypothesize the activation of this signaling pathway will reduce vascular leakage which leads to pulmonary edema.

The aims of the study are to evaluate the efficacy of Vaculotide to treat pulmonary edema formation, and to determine the optimal dosing of Vasculotide in the setting of an isolated organ.

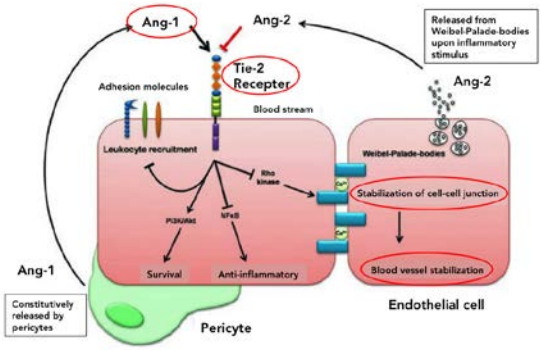

Figure 2. Ang/Tie-2 ligand-receptor system

［3］成果（Research outcomes）

（3－1）研究成果（Results）

First of all, human donor lungs declined for transplant after clinical EVLP were used for this study. After confirmation of research consent, lungs were divided in 2 groups. In the low dose group, 15 μg of Vasculotide were give into EVLP circuit (Concentration: 10 ng/mL). In the high dose group, 150 μg of Vasculotide were give into EVLP circuit (Concentration: 10 ng/mL). Afterwards, the lungs were perfused and ventilated for 4 hours according to the standard Toronto EVLP protocol. Physiological parameters such as peak airway pressure, lung compliance, and pulmonary vascular resistance were recorded every hour. Partial pressures of oxygen and carbon dioxide, glucose and lactate levels, and electrolytes were also measured hourly through blood gas analysis. Tissue and perfusate samples were collected hourly to perform biological and immunological cytokine analysis. In addition, tissue samples were fixed with formalin and used for hematoxylin-eosin staining and immuno-fluorescence staining for vascularization markers such as ZO-1. As a result of above human lung study, low dose of Vasculotide was considered to be optimal dosing in order to use for treatment during EVLP.

（3－2）Future perspectives

We expect Vasculotide to exert anti-inflammation, anti-permeability, and anti-apoptotic properties in lung grafts. Therefore, we anticipate Vasculotide administration during EVLP will ameliorate pulmonary edema and also expand lung utilization for transplant after EVLP. This study could have a significant impact in lung transplantation as it could open the donor pool to a significantly underutilized group of lung donors. Further, Vasculotide administration is cost-effective and easy, and positive results from its administration during EVLP could further increase the availability and success of lung transplantation. The findings from our proposed project will be crucial in establishing the best strategy for severe injured lungs during EVLP.

［4］List of Papers

(1) Watanabe Y, Galasso M, Watanabe T, Ali A, Qaqish R, Nakajima D, Taniguchi Y, Pipkin M, Caldarone L, Chen M, Kanou T, Summers C, Ramadan K, Zhang Y, Chan H, Waddell TK, Liu M, Keshavjee S, Del Sorbo L, Cypel M. Donor Prone Positioning Protects Lungs from Injury During Warm Ischemia. *Am J Transplant*. [Epub ahead of print]

(2) Galasso M, Feld JJ, Watanabe Y, Pipkin M, Summers C, Ali A, Qaqish R, Chen M, Ribeiro RVP, Ramadan K, Pires L, Bagnato VS, Kurachi C, Cherepanov V, Moonen G, Gazzalle A, Waddell TK, Liu M, Keshavjee S, Wilson BC, Humar A, Cypel M. Inactivating hepatitis C virus in donor lungs using light therapies during normothermic ex vivo lung perfusion. Nat Commun. 2019;10:48