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The Experimental Study on Lung Transplantation

—Preservation of the Canine Lung by Cooling and Perfusion—

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Previous studies in the laboratory have been shown that homografts of the lung in canine can survive extended period of time, if methotrexate is given to the recipient animal. Several homografts survive more than a month. A series of fifty unrelated mongrel pairs have been subjected to homotransplantation of the left lung. In each case the donor lung was stored in vitro 2 to 12 hours before placement in the recipient animal. The lung was perfusated with 5% dextrose, 3.6% PVP and plasma at 4°C by infusion pump and was ventilated with room air by respirator. It has been eluciated by histologic and electromicroscopic study including autopsy and survival rate that the safety storage period had been within 4 to 6 hours to prevent a fine change on donor lung due to storages.

INTRODUCTION

Successful clinical homotransplantation of the lung will require the solution of a variety of technical and physiological problems. The greatest one is the immune barrier and, as yet, relatively little progress has been achieved in the clinical suppression of this genetically determined reaction. However the survival of homografted organs has been prolonged by the use of drugs that delay immunologic responses of the host and it is necessary to preserve the donor lung for widely achieving homotransplantation, although the elaboration of technique and pulmonary function on autologous lung transplantation and the effects of methotrexate on suppression of immunological response had been reported at the previous meeting of Japanese surgical society[1] and

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Japanese association of thoracic surgery[2], the present series have evaluated the possibility of preservation of the canine lung by cooling and perfusion.

METHOD

In 50 young mongrel dogs, of medium size, a left pneumonectomy was performed through a lateral thoracotomy incision in the fifth intercostal space. The left pulmonary artery, the bronchus, and the left atrium, proximal to the insertion of the left pulmonary veins, were individually transected and the lung was completely removed from the chest. The donor lung was stored at 4°C, ventilated with 15 cm H_2O of intratracheal pressure by respirator and perfused with 5% dextrose, 3.6% PVP or plasma by infusion pump through the catheter into the pulmonary artery with 4.0 to 5.0 ml/min of perfusion flow rate in order to prevent from the occurence of lung edema (fig 1) and the histologic and electromicroscopic study had been appraised the change of the donor lung associated with the elapse of perfusion to evaluate the influence on the storage lung. Furthermore the life-time on homologous lung transplantation after the preservation within 4 to 6 hours, had been compared with the perfusate of 5% dextrose, 3.6% PVP or plasma, and with cooling alone without perfusion. The applica tion of the dye dilution method (cardiogreen) from the main pulmonary artery to the left atrium immediately after transplantation has been performed to evaluate the disparity of the survival rate between in group of cooling alone without perfusion and with perfusion.

RESULT

From histological finding, the swelling of the alveolar septum and alveolar epithel has been shown remarkably at the 20 th to 30 th minute after perfusion by 5% dextrose (fig 2) and the desquamation of alveolar epithelium, the rupturing of alveolar wall and elastic fiber and emphysematous change of alveolar structure had been certified at the 7 th hour after perfusion (fig 3). However the degree of inflicted damage on the lung perfused by PVP was less than by 5% dextrose at the 4 th to 6 th hour after perfusion (fig 4.), although pulmonary damage treated with PVP had been demonstrated in proportion to perfusion period. The lung treated with plasma had been inflicted with as much as with PVP (fig 5), but the erythrocyte in the pulmonary vessels has been replaced incompletely by plasma compared with other perfusate. From electromicroscopic finding from which it had been discerned the change of basement membrane or of alveolar cell and capillary endothelial cell related to gas exchange, the edematous

patterns on the lung had been presented as a rough and bright zone due to low electric density in the intercellular stroma and basement membrane at the 15 th minute after 5 % dextrose perfusion (fig 6) and these changes are more progressive with the passage of perfusion time (fig 7). At the 2 nd hour after perfusion the rupturing and destruction on alveolar cells and capillary endothelial cells have become manifest (fig 8), while the changes on the basement membrane and stroma inflicted by PVP or plasma perfusion have not been revealed remarkably (fig 9), that is the edematous appearance on perfused lung has not been predominant, even at the 12 th hour after perfusion (fig 10). However the cytoplasma on alveolar and capillary endothelial cells has become rough and its cells membrane has become irregular.

By the above findings, the perfusion with PVP and plasma has provoked slight edematous changes but it has been proven that cellular destruction had not occured in 4 to 6 hour duration.

The life-time on homologous lung transplantation after the preservation by perfusion within 4 to 6 hours had been compared with the perfusate of 5 % dextrose, PVP and plasma. The survival duration in group of 5 % dextrose perfusion has been much shorter than in group of PVP and plasma perfusion, although it has no significance between PVP and plasma perfusion. Furthermore the survival rate in group of cooling alone without perfusion was worse than in group of PVP and plasma perfusion (table 1). The pulmonary hemodynamics immediately

Table 1.
Survival rate after transplantation of storage lung during 4 and 6 hours

	Perfusate	Case No.	survival term		
			1–5 days	5–10 days	10–20 days
cooling with perfusion	5 % dextrose	13	13		
	3.6 % PVP	15	3	7	5
	Plasma	16	2	10	4
cooling alone		16	11	5	

after transplantation has been studied by the dye dilution method to approve the disparity of life-time between the group with perfusion and without perfusion. The elevated pressure of pulmonary artery, decreased cardiac output, prolonged circulation time and increased pulmonary blood volume in group of cooling alone have been manifest compared with in group with perfusion (table 2).

It has been ascertained that the extended period of the canine lung preservation has been within 4 to 6 hours from autopsy that one dog

Table 2.
The pulmonary hemodynamic changes immediately after delayed transplantation (by the dye dilution method)

	Case No.	The pressure of pulmonary artery (mmHg)	cardiac output (cc/min/kg)	mean circulation time (sec)	pulmonary blood volume (cc/kg)
before transplantation	9	14.0	216	3.9	13.6
after transplantation perfusion group by PVP	5	17.3	273	3.7	15.6
after transplantation non-perfusion group	4	20.5	157	4.8	37.2

survived up to 15 days after transplantation had elucidated the remarkable necrosis on transplanted lung with 6 hours of perfusion time.

DISCUSSION

The preservation method of the canine lung by cooling and perfusion has been evaluated by histologic, electromicroscopic and hemodynamic study to obtain the extended period of storage lung.

It has been certified that perfusion flow rate with various perfusate should be low to prevent from provoked lung edema and isomolar and isotonic perfusate such as plasma and PVP had been available for adequate perfusion. However it seems that the limitation of the extended preservation period of the canine lung would be within 4 to 6 hours from the histologic and electromicroscopic finding in which it had not been shown severe parenchymatous changes on storage lung yet and from tolerance test which had observed survival period after transplantation.

Recently autologous and homologous lung transplantation have been achieved by some investigators[3)4)], but there are a few literatures[5)6)7)] related to delayed reimplantation including storage. The cooling and perfusion method as the preservation of the canine lung have been disputed and the method employed has been evaluated by means of dye dilution method, which is an excellent method to resume and improve pulmonary function on transplanted lung immediately after transplantation.

CONCLUSION

Cooling and perfusion method for the canine lung have been discussed

to extend the storage period. It has been elucidated by histologic and electromicroscopic study including autopsy that the safety storage period had been within 4 to 6 hours to prevent a fine change on donor lung due to storage. However further study to improve the storage method should be conducted although a functioning lung had been occasionally achieved.

It is certified by the dye dilution method that the recovery of pulmonary function on transplanted lung with perfused preservation has been much easier than with a nonperfused one immediately after transplantation.

REFERENCES

1) Tsuji Y., et al. : Presented on the meeting of annual Japanese surgical association 1963.
2) Tsuji Y., et al. : Presented on the meeting of Japanese association of thoracic surgery 1963.
3) Christiansen, K.H. et al. : Homologous Transplantation of Canine Lungs : Technique With Contralateral Pulmonary Artery Ligation, A. M. A. Arch. Surg. 86 : 495, 1963.
4) Neptune, W. B., et al. : Experimental Lung Transplantation. J. Thoracic Surg. 26 : 275, 1953.
5) Blumenstock, D. A., : Replantation and Transplantation of the Canine Lung. J. Surg. Res. 1 : 40, 1961.
6) Bogardus, G. M., : An Evaluation in Dogs of the Relationship of Pulmonary, Bronchial, and Hilar Adventitial Circulation to the Problem of Lung Transplantation. Surgery 43 : 849, 1958.
7) Hardy, J. D. et al. : Autotransplantation and Homotransplantation of the Lung. : Further Studies. J. Thoracic Cardiovascul.. Surg. 46 : 606, 1963.

Fig. 1. Cooling and Perfusion Method

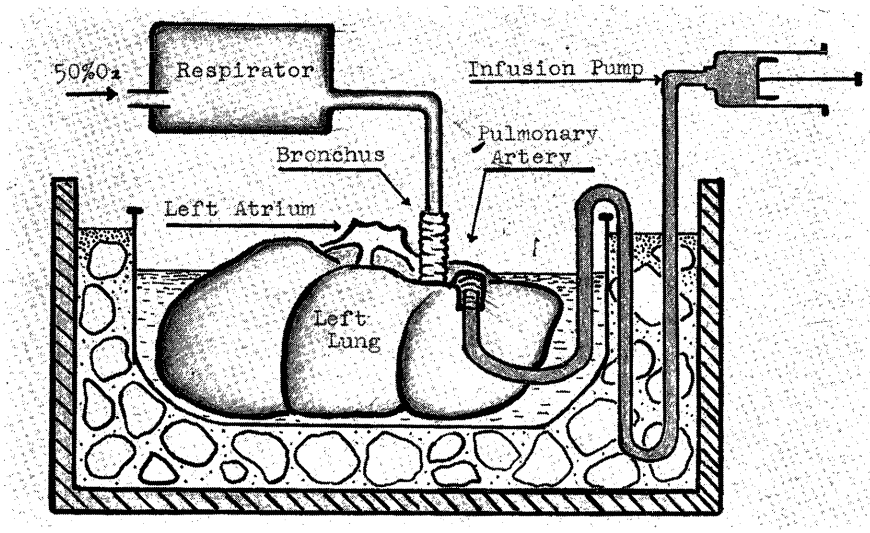
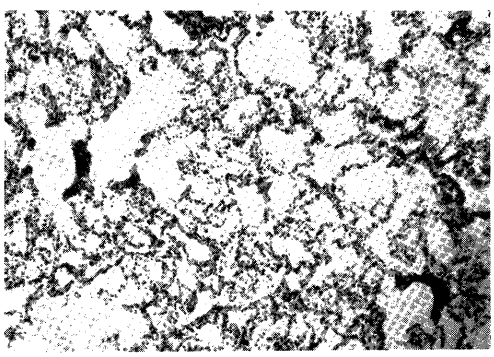


Fig. 2.



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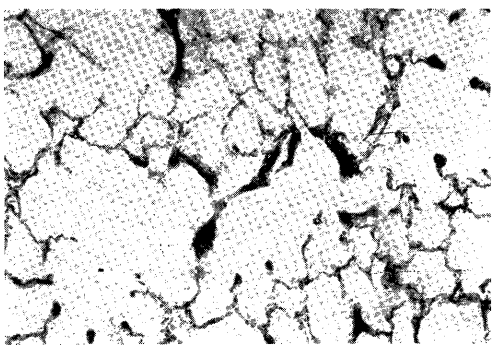


Fig. 4.

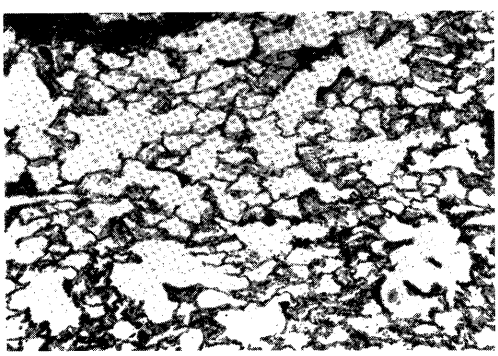
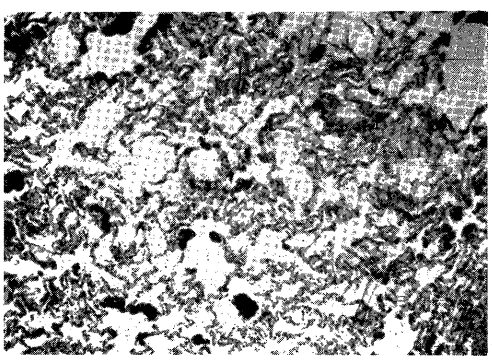


Fig. 5.



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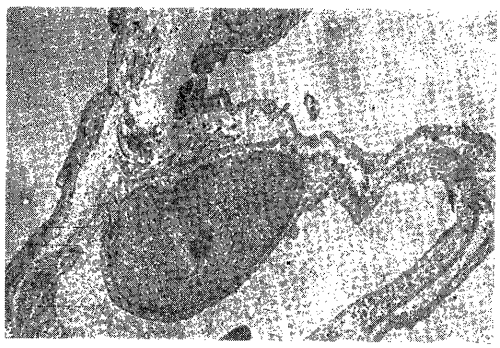

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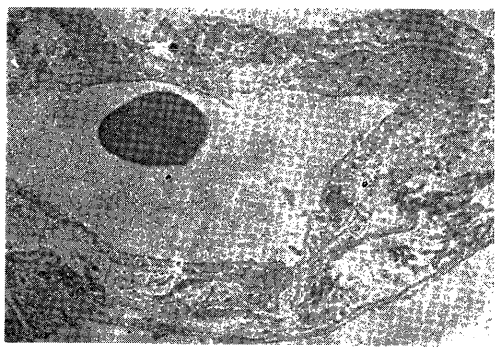

Fig. 8.

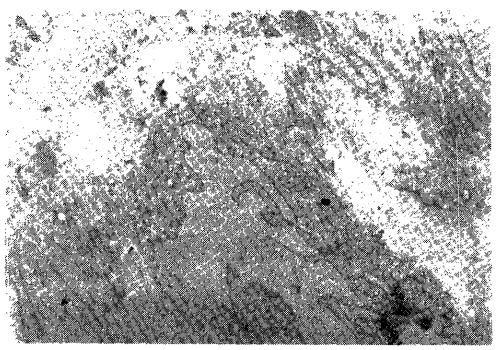

Fig. 9.

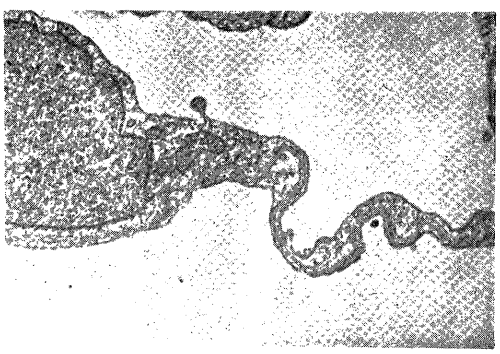

Fig. 10.