THE ISOLATED AIR PERFUSED RAT HEART

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Abstract: A simple method has been developed for continuous monitoring of metabolic activity of an isolated, perfused rat heart by O2/CO2 respirometer. Since respirometer provides vital data on oxygen consumption and carbon dioxide production of a preserved organ on a continuous basis over a long period of time, it will be possible to use this method to monitor viability of not only isolated heart but also any given donor organ under preservation.

Key words: air perfusion organ transplantation isolated heart

INTRODUCTION

Success of transplantation of organs depends on the ideal preservation of organs during transport from the donor to the recipient. Hypothermia during transport slow down ATP loss in the excised organs and thereby keep them viable for a short period of time. In recent years attempts have been made to develop methods for long-term preservation of hearts by hypothermia and cardioplegia (1). Although hypothermia and cardioplegia prolongs preservation time to 48 hrs, these conditions have always shown some evidence of myocardial damage. In order to mitigate such myocardial damage, Bailey has preserved guinea pig hearts by hypothermic gas perfusion (2). His study has demonstrated that the combination of hypothermia and 95% oxygen gas perfusion, a procedure known to increase oxygen tension in cardiac muscle, has retained contractile activity in guinea pig hearts after 24 hrs of preservation and has not caused any edema.

The present study has been performed to determine whether rat hearts can be preserved by air perfusion, instead of 95% O2, which can predispose organs to undergo lipid peroxidation, and whether their viability can be monitored by continuously measuring oxygen consumption and carbon dioxide production by a O2/CO2 Respirometer (Micro-Oxymax; Columbus Instruments International Corporation, Columbus, Ohio).

METHODS

Hearts are mounted on a fluid perfusion system and perfused through the aorta with Krebs solution being delivered at a rate of 5 ml/min at 37°C by means of a roller pump as shown Fig. 1. After 10 min the peristaltic pump is turned off and heart is opened towards air delivery system. Fig. 2 is a schematic illustration of the air perfusion system. The system consists of a water jacketed heart retention chamber and airtight lid with two inlet and two outlet tubes. One inlet tube is used for cannulation of the aorta to deliver air through coronary system. One outlet tube withdraws air from the chamber by means of a teflon pump. The second set of inlet and outlet tubes are connected to a Micro-Oxymax. The outlet tube is used for withdrawing air from the chamber every 60 min for oxygen and carbon dioxide measurements while inlet tube returns air sample to the chamber.

RESULTS AND DISCUSSION

A total of five hearts have been perfused

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Fig. 1: Schematic diagram illustrating the system for perfusion of physiological salt solution through aorta of an isolated rat heart.

Fig. 2: Diagrammatic illustration of the unit for air perfusion of a single rat heart and for measurements of oxygen consumption and carbon dioxide production by Micro-Oxygraph.
utilising the methods described above. Of these five experiments, results of three are taken into consideration. One took an unusually long time to transfer the heart from fluid to air system. Another experiment was discontinued halfway due to power failure.

After 2-3 hrs of equilibration period, oxygen consumption by these heart reaches a steady-state $6.25 \pm 0.43 \text{ l/min/gm wet weight (mean±SE)}$ between 3-9 hrs. The oxygen consumption by isolated rat heart at 36°C is reported to be $523 \pm 47 \mu\text{l/min/g dry weight or 105} \pm 9 \mu\text{l/min/g wet weight, assuming 80% as water contents of these hearts (3).}$ It is about 18.4% (the amount calculated for the fluid-perfused-heart) in isolated guinea pig heart perfused with 95% gaseous oxygen (4). Thus, low oxygen consumption by the air perfused rat heart, which is about 6% of the fluid perfused heart, is partly explained by their diminishing contractile activity. Also, it is possible that low oxygen (20%) used in this study has not been high enough to provide adequate tissue oxygen.

Oxygen consumption rises and reaches a peak value of $14 \pm 2 \mu\text{l/min/g wet weight at 15.25 hr. (Fig. 3)}$. This high consumption of oxygen may be due to bacterial growth and/or peroxidation of cellular elements of the dying heart. In the present study, hearts were not excised from the chest cavity under aseptic condition and perfusion medium did not contain any antibiotic. Both these precautions with purified air through bacterial filters will minimise or completely eliminate infection-induced oxygen consumption. Effects of lower temperature and higher oxygen tension on the quality of preserved organ can be assessed by comparing the oxygen consumption values.

![Diagram: O2 Consumption vs. Time (Hours)](image)

Fig. 3: Individual oxygen consumption records from three air perfused hearts during 20 hr preservation period.
obtained from the respirometer. The system will allow one to determine whether the preserved organ is deteriorating by lipid peroxidation or not by periodically measuring ethane and pentane contents in the chamber air by collecting samples from the Micro-Oxymax after it has completed its measurements of oxygen and carbon dioxide (5) (Fig. 4).

**Fig. 4:** Individual carbon dioxide production records from the same three hearts as shown in Fig. 3.

**REFERENCES**


