2. Langendorff Heart

2.1. Principle

Langendorff heart is one type of isolated perfused heart which is widely used for biochemical, physiological, morphological and pharmacological researches. It provides detailed analysis of intrinsic ventricular mechanics, metabolism and coronary vascular responses.

Langendorff heart (Fig. 2.1) is the isolated perfused heart of which the coronary arteries are perfused by retrograde flow from aorta. For maintaining cardiac activity, the basic principle of Langendorff isolated heart is to perfuse oxygenated perfusate via coronary arteries using a cannula inserted in the ascending aorta. Then, during diastole when the aortic valves are closed, the perfusate flows through the coronary arteries. After passing through the coronary vascular system, the perfusate enters the right atrium via coronary sinus and finally flows out via the right ventricle and pulmonary artery. The perfusate might be blood, Tyrode, Locke or Krebs-Henseleit bubbled with carbogen at physiological temperature. The perfusate flow rate is controlled by hydrostatic pressure for constant pressure model, or by roller pump for constant flow model. The latter model was used for our experiments.

Like other isolated organs, the certain advantage of the Langendorff heart is that measurements can be obtained without many complications such as feedback effects and hormonal and neural controls that originate outside of the heart. This allows an investigation of more specific response to the cardiac drug.
Figure 2.1  (a) Normal circulation (Opie, 1997). (b) Circulation in Langendorff perfused heart (Dhein, 2005). In Langendorff perfused heart, perfusate fluid enters the heart via the aorta. RA, right atrium; RV, right ventricle; PA, pulmonary artery; LA, left atrium; LV, left ventricle; Ao, aorta; O$_2$, oxygen.
2.2. Langendorff Isolated Perfused Rat Heart System

For the Langendorff preparation, the model of isolated heart size3 (HSE-Harvard, March-Hugstetten, Germany), as shown in Fig. 2.2, was used in our experiments. The main part of Langendorff apparatus consists of a gear pump, an oxygenator, a roller pump, a heat exchanger, a thermostatic heart chamber and transducers. The gear pump is used for delivering the perfusate fluid from the reservoir to the oxygenator which produces thin film of the perfusate fluid in a carbogen (95% O₂ and 5% CO₂) atmosphere. Then, the saturated carbogen perfusate fluid is supplied to the heart by means of a roller pump, which generates a constant perfusion flow. This will produce a corresponding perfusion pressure, which is measured by transducer no.1 (Fig. 2.2). The perfusion solution is delivered to a heat-exchanger chamber (normally a double walled cylinder connected to the thermostat) with an aortic cannula at its bottom. When the heart is attached to the cannula, the thermostatic heart chamber is moved to cover the heart. By transducer no.2 (Fig. 2.2), left ventricular pressure is recorded isovolumetrically supplied via a balloon catheter.

Modified Krebs-Henseleit Buffer

The perfusate fluid used in this research was a modified Krebs-Henseleit buffer containing NaCl (118 mM), KCl (4.7 mM), CaCl₂ (1.5 mM), MgSO₄ (1.66 mM), NaHCO₃ (24.88 mM), KH₂PO₄ (1.18 mM), glucose (5.55 mM), Na-pyruvate (2 mM), and bovine albumin (0.1%w/v). The perfusate buffer was freshly prepared and filtered by 0.45 μm hydrophilic microfiltration membrane based on polyether sulfone (Sartorius AG, Gottingen).
Figure 2.2  Model of Langendorff heart at constant flow (modified from operating instructions for the experimental apparatus isolated heart size3 type 830, HSE-Harvard, March-Hugstetten, Germany)
2.3. **Langendorff Isolated Perfused Heart Preparation**

Rat was intraperitoneal (ip) injected with 500 IU heparin and anesthetized by ip injection of 50-80 mg/kg pentobarbital. After the rat became unconscious and lost pedal reflex activity, the heart surgery was started. First, its trachea was cannulated with an artificial ventilator, and then a midsternal thoracotomy was performed to open its chest. Second, the heart was hastened to excise and to transfer into oxygenated ice-cold modified Krebs-Henseleit buffer. Then, the cannula filled with oxygenated modified Krebs-Henseleit buffer was tied to its aorta. Finally, the heart attached cannula was rapidly switched to connect with the Langendorff perfusion apparatus. The apparatus had been previously set for constant flow rate (9.7 ± 0.5 ml/min) of the modified Krebs-Henseleit buffer saturated with carbogen (95% O\(_2\) and 5% CO\(_2\)), and its temperature was controlled at 37°C.

In order to measure pharmacodynamic response, a latex balloon tied to the end of a polyethylene tube, which was connected with pressure transducer, was carefully inserted into a left ventricle of the isolated heart. The balloon was inflated with 50% methanol to create a diastolic pressure of 5 to 6 mmHg. Four pharmacodynamic endpoints, left ventricular systolic pressure (LVSP), left ventricular enddiastolic pressure (LVEDP), heart rate (HR) and coronary perfusion pressure, were measured and monitored continuously by a physiological recording system (HSE Harvard, March-Hugstetten, Germany). Additionally, left ventricular developed pressure (LVDP) is defined as \( LVDP = LVSP - LVEDP \), and coronary vascular resistance (CVR) is calculated from perfusion pressure divided by coronary flow.