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# DESIGN OF A BATH FOR PERFUSING ISOLATED LYMPH VESSELS

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The design of a bath for perfusing the isolated thoracid duct is described. The method enables changes in the lumen of the duct resulting from the action of various agents *in vitro* to be assessed.

KEY WORDS: *Isolated thoracic duct; perfusion.*

Perfusion of isolated lymph vessels can conveniently be carried out in a bath of the suggested design (Fig. 1). The bath is made of transparent plastic. It measures 30×8×4 cm and the capacity of its central part is 150 ml.

The inlet cannula (1) is inserted into the thoracic duct (TD) below the arch of the aorta and the outlet cannula (2) into the mouth of the duct in the neck. The TD is carefully isolated from the surrounding tissues and placed in the bath.

The cannulas are connected by short pieces of rubber tube (3) to glass tubes (4, 5), on which are fitted rubber sleeves (6) which fit snugly in grooves on the walls of the bath. These sleeves prevent the physiological saline from spilling when the bath is filled and they serve to arrange the glass tubes (4, 5) at a small angle to the floor of the bath so that the TD is immersed in the physiological saline. The tubes can be moved along their long axis in order to apply known tension to the duct, which can be verified by the length of a ligature which is equal to the length of the segment of TD *in situ*.

The bath is filled with Tyrode solution through tube (7) and the solution is drained through tube (8). To maintain a constant temperature, a polyethylene tube is provided on the floor of the bath and its ends protrude outside the bath (9, 10). Warm (37°C) water circulates through this tube.

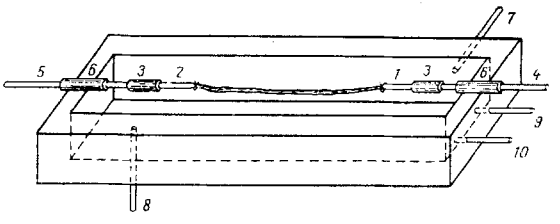


Fig. 1. Bath for perfusing isolated lymph vessels (explanation in text).

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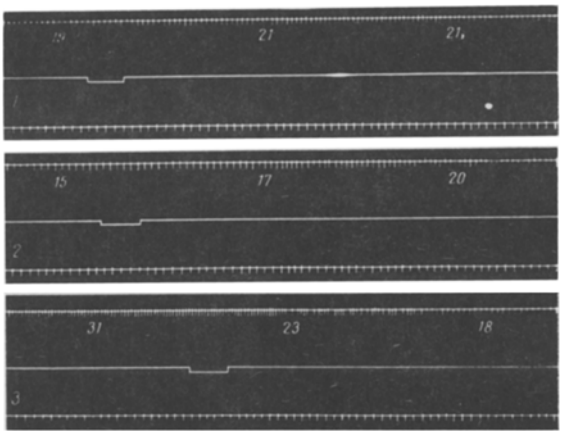


Fig. 2. Changes in lumen of isolated segments of thoracic ducts of dogs under the influence of catecholamines. From top to bottom: flow of perfusion fluid through thoracic duct (numbers show number of drops per minute before and 2 and 4 min after stimulation of adrenoreceptors); market of stimulation; time marker 5 sec. 1) noradrenaline ($12 \cdot 10^{-8}$ g/ml), perfusion pressure 27 mm water; 2) adrenalin ($88 \cdot 10^{-9}$ g/ml), perfusion pressure 25 mm water; 3) adrenalin ($44 \cdot 10^{-8}$ g/ml), perfusion pressure 27 mm water.

The isolated segment of TD was perfused with Tyrode solution with the addition of blood plasma (1:15). For this purpose the inlet cannula was connected through a thermostat to the delivery vessel and the outlet cannula was connected to a drop counter. On the addition of various substances to the bath an increase in the number of drops of perfusion fluid flowing through the isolated segment of TD in unit time indicates dilatation of TD whereas a decrease indicates constriction. The same bath can be used to study the effect of various agents on the length of an isolated segment of TD, reflecting the state of the longitudinal muscles in its wall. For this purpose only one end of TD is fixed to a cannula whose tip is lowered to the bottom of the bath. The bath is placed at an angle of 45° to the surface of the laboratory bench, sloping toward the drainage tube. A strip of paper with divisions is glued to the transparent floor of the bath.

By means of the method described it was shown that adrenalin hydrochloride ($44 \cdot 10^{-9}$-$11 \cdot 10^{-8}$ g/ml) and noradrenaline hydrotartrate ($12 \cdot 10^{-8}$-$24 \cdot 10^{-8}$ g/ml), if added to the surrounding solution, cause dilatation of the perfused segment of the cervical portion of the duct in dogs (Fig. 2). Measurement of its length showed that the segment was shortened. Presumably dilatation of TD in response to the action of alpha-adrenomimetics in the above-mentioned doses is active in nature and is due to an increase in tome, chiefly of the longitudinal muscle bundles in its wall. An increase in the concentration of adrenalin ($33 \cdot 10^{-8}$-$44 \cdot 10^{-8}$ g/ml) and noradrenalin ($36 \cdot 10^{-8}$-$60 \cdot 10^{-8}$ g/ml) causes constriction of TD, probably because of a simultaneous increase in the tone of the circular muscles. In some cases constriction of the duct was preceded by the appearance of marked motor activity of the vessel. The action of adrenalin and noradrenalin was blocked by phentolamine. Novodrin (isoprenaline sulfate) in a concentration of $22 \cdot 10^{-7}$-$66 \cdot 10^{-7}$ g/ml dilated the segment of TD a little but did not affect its length. In this case dilatation of TD was evidently due to relaxation of the circular muscles of the vessel wall. The addition of acetylcholine chloride ($66 \cdot 10^{-6}$-$132 \cdot 10^{-6}$ g/ml) to the bath led to constriction of TD. This effect was blocked by atropine.

The bath of the design described above can be used also to study responses of blood vessels.