## SEMICONDUCTOR THERMOSTAT FOR WORKING WITH BIOLOGICAL PREPARATIONS IN OPEN CUVETTES

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An apparatus for controlling and stabilizing temperature in an open cuvet of capacity 15-20 ml within the temperature range 4-30° is described. Fluctuations of temperature do not exceed  $\pm$  0.1°. A semiconductor thermocooling bench of type TOS-2, as used for freezing blocks of tissue in histological practice, was used for cooling and heating. The TOS-2 was connected into the automatic temperature regulating circuit. The detector unit is a contact mercury thermometer placed in the solution in the cuvet. The method of temperature stabilization used in the apparatus is based on maintenance of a constant temperature drop with height of the cuvet. The temperature drop is about 2°. Temperature in the plane of the preparation is controlled by a point thermistor. This apparatus was used for microelectrode recording of action potentials from a strip of the frog's ventricle within the specified temperature range.

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Control and stabilization of temperature in open cuvettes, especially at temperatures below the external environmental temperature, can be very difficult because clumsy and complex cooling devices are required [1]. It is interesting to utilize the thermoelectric cooling and heating effect in certain intermetallic alloys, possessing semiconductor properties [2], in instruments for controlling temperature. If a direct current is passed through such a semiconductor element, heat is absorbed on one layer and emitted on the other. With a change in direction of the current the opposite effect is observed, i.e., each surface can serve as cooler and also as heater. Such elements are widely used at the present time in cryogenic work and in some physical investigations [3]. An apparatus for stabilizing the temperature of perfused blood has been described [4].

The apparatus now proposed is intended for temperature stabilization in an open cuvet. A strip of frog myocardium was placed in a cuvet for experiments with microelectrode recording of its electrical potentials at temperatures of between 4 and 32°. The TOS-2 thermocooling bench as used for freezing blocks of tissue in histological practice was incorporated in the apparatus. The bench consists of a separate power unit, providing a direct current of up to 35 A. The working area of the bench surface is 1600 mm² and its weight 0.4 kg. Power consumption from the main supply is 15 W. The heat emitted is carried away by tap water circulating through the bench.

The assigned temperature of the preparation placed in physiological saline can be obtained in several ways. A perfusion method is widely used, when a solution brought up to the required temperature is circulated through the cuvet. However, this method cannot be used when it is undesirable to prepare a large volume of solution because certain of the substances investigated, e.g., toxins and enzymes, are in short supply.

Temperature stabilization can be achieved by placing the cuvet in a thermostatically controlled volume of liquid, in which case an identical temperature at different points within the cuvet is attained by stirring the solutions with a small mixer. The use of a mixer also is undesirable, because a larger cuvet must be used, vibrations are produced in the preparation, and its motor causes additional interference.

The method adopted in the proposed apparatus is stabilization of the temperature gradient by means of an inertial contact mercury thermometer placed in the solution in the cuvet and connected into the power

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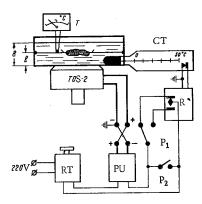


Fig. 1. Block diagram of apparatus for controlling and stabilizing temperature. TOS-2—thermocooling bench; PU—power unit of bench; CT—contact thermometer; T—thermistor with needle indicator for verifying temperature in plane of preparation; R—intermediate relay;  $P_1$ —switch for changing from "cooling" to "heating" operation;  $P_2$ —switch for changing from "automatic" to "programed" operation; l—distance from bottom of cuvet to plane of preparation. Diagram of apparatus illustrated operating on "cooling."

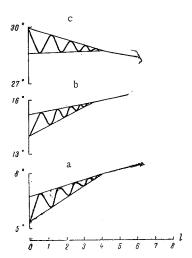


Fig. 2. Regulating curves of apparatus. a, b) Curves of temperature regulation at different heights of the cuvet when operating on "cooling" with mean temperatures of 7 and 15°, respectively, and input voltage of power unit of TOS-2190V; c) when operating on "heating" at mean temperature 28° and voltage 160 V. Smallest fluctuations of temperature are observed when  $l>4\,$  mm.

circuit of the TOS-2. In this case the thermometer shows the mean temperature of the solution, and in the solution itself there is a small temperature drop, about 2° over the height of the solution (0.8 mm). If the thickness of the preparation is small (<1 mm), the temperature drop in the preparation itself can be disregarded.

The cuvet, measuring  $40\times40\times15$  mm, is made of organic glass and glued with epoxide resin to the working surface of the TOS-2. At the side a contact thermometer is inserted into the cuvet through a hole. The instrument is fixed rigidly to a manipulator. The working part of the thermometer must be completely immersed in the solution and must touch the bottom of the cuvet. Tap water for cooling and the bus bars of the power circuit are conveyed to the TOS-2 through hoses.

The block diagram of the apparatus is illustrated in Fig. 1. When operating on "cooling" and the mean temperature of the solution reaches a certain level the contacts of the thermometer are closed, and an intermediate system, consisting of a semiconductor switch [1] and the relay MKU-48, is thereby made to switch on the power unit supplying the TOS-2. At the same time the temperature in the solution falls and the thermometer contacts are opened. The TOS-2 is switched on and off at the rate of about once per minute. With a change in polarity of the current feeding the TOS-2, the instrument operates on "heating". In this case the temperature in the cuvet is stabilized above the external environmental temperature. To convert the instrument to this mode of operation the switch P<sub>1</sub> is used, changing the polarity of the current and switching over the contacts of the MKU-48 relay in the intermediate system. When the thermometer contacts are closed, the power supply is switched off. The contacts of the switch P<sub>1</sub> in the power circuit of the TOS-2 are of knife type. Their resistance is usually low and they pass a current of not less than 50 A.

Since the TOS-2 has a greater heating power than is necessary for stabilizing temperature within this range, a regulating transformer (RT) of the LATR type is included in the circuit, lowering the input voltage of the power unit of the TOS-2. The apparatus is adjusted by choosing this voltage for both types of operation and by determining the smallest distance l (Fig. 1) from the working surface of the bench to the plane in which permissible fluctuations of temperature (±0.1°) are observed during regulation. Hooks for fixing the preparation and the lever of a tensometric sensor for recording muscle contractions are secured in this plane. Temperature in the plane of the preparation is controlled by means of a point thermistor and needle

indicator. The distance l is determined as follows. Solution is poured into the cuvet to a height of 0.8-1.2 mm, and fluctuations of temperature at different heights are measured when operating on "cooling." The measurements are made by a thermistor fixed to the manipulator in a vertical plane at temperatures of 7 and 15° (on the mercury thermometer) and also with different input voltages of the power unit. The time

of each measurement is 5 min. Curves of temperature regulation for optimum values of input voltage of the power unit are shown in Fig. 2. The distance *l* determined from curves (a, b) with a voltage of 190 V is 4 mm when operating on "cooling." When operating on "heating" the smallest fluctuations of temperature at this height within the temperature range 17-32° are obtained when the input voltage of the power unit is 160 V. The regulating transformer can be replaced by suitable voltage-dropping resistors.

Experience of work with the apparatus showed that the best characteristics of temperature regulation are obtained when the temperature of the cooling tap water is 17-20°. To overcome power-line interference the bus bar feeding the TOS-2 and the wires to the thermistor and contacts of the mercury thermometer must be grounded. The intermediate relay is powered by a battery or separate transformer with its secondary winding grounded.

If necessary the temperature in the cuvet can be varied in accordance with an assigned program, by regulating the input voltage of the TOS-2 power unit with a transformer. The switch  $P_2$  is provided for changing the instrument over to operating under these conditions.

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