An Apparatus for Microscopic Study of Frog Heart in Situ

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It has been shown by Gramenitsky, in articles which have since been neglected, that the excised, beating frog heart can be stretched in such a way as to render its atria and sinus venosus translucent and amenable to microscopy (1, 2), and that with such a preparation, new discoveries are made possible concerning structure and functioning (3). This means of investigation was extended by Fedorov (4) and by Smitten (5) to a study of the cardiac autonomic synapses.

An apparatus which I have devised, and which I term a *cardiostat*, permits the beating heart to be investigated similarly but without its removal from the body (Fig. 1). A narrow platform serves for plac-



FIG. 1.

ing the frog supine, with its legs straddling. A transverse strip of spring metal on the under surface clips its feet. A shield of coke tin on the end of a strip of metal keeps its head bent under the forward edge of the platform. An inverted shallow cup on another strip of metal restrains the upturned viscera other than the heart. A frame proceeding from the forward edge of the platform serves for the attachment of ligatures through the apex of the heart and the tissues at its base. A vertical shaft under the back edge of the platform supports the latter as it rests on the microscope platform. A system of ways and carriages gives movement in two horizontal directions, and a plate of metal pierced by thumbscrews allows vertical adjustment.

By means of the preparation described, the beating heart can be studied under the microscope with its innervation and to some extent its circulation intact. For the former purpose, the frog is to be anesthetized intraperitoneally instead of being pithed.

A preliminary report on the appearance of the ¹Much appreciation is expressed to Don Hauk for the construction of the cardiostat model reported and for the accompanying photograph of it.

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vagal synapses in functional and deteriorated states, as investigated by this means, is presented elsewhere (6). It may here be noted that without staining reversible changes are visible during synaptic transmission, the end-feet glistening as if with clear exudate and the ganglion cells undergoing a definite enhancement of detail. Morphologic deterioration, it may be added, becomes visible in the form of a slight enlargement of the end-feet, and a permanent enhancement of the cellular detail, within an hour of interruption of the innervation and circulation, and passes after many hours into a second stage, associated with functional deterioration, characterized by a numerical reduction of the visible end-feet, and tenacity of stains on the part of the ganglion cells.

The further range of investigation to which the cardiostat preparation is amenable includes problems of physiology, pharmacology, histology, and cytology. Observations are in progress on these. Excellent visualization, with or without staining, is afforded by an oil-immersion objective used with Ringer's solution instead of oil. Photomicrography and cinematomicrography are eminently feasible.

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Perfect Demarcation of the Diamond Crystal Structure in Germanium

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In our studies of single crystal growth in germanium, careful consideration was given to the advantages of both radiant heating and RF induction heating. Analyses of the advantages pointed to the use of direct radiant heating to eliminate the turbulence caused by induction methods. After this choice was made, it was then necessary to establish a desirable temperature gradient of horizontal isotherms from the lowest temperature at the top to highest at the bottom.

The over-all arrangement of our equipment permits observations of the top surface of the molten germanium. Thus one can watch the germanium nucleate into a single crystal solid. Visible nucleation commences from the top and is associated with definite geometric patterns, which may be triangular, hexagonal, or diamond in nature, depending upon the plane of growth.

Nucleation starts in a very small area, and the pattern formed continues to grow in size toward the sides



FIG. 1. A: twin line across surface; B: clear lines indicative of crystallization.

of the crucible. As growth continues undisturbed, with no parasites (i.e., secondary centers of nucleation) being formed, the resulting pattern covers the entire surface. Associated with this horizontal growth is a vertical growth that starts with the very small nucleation area and grows down towards the bottom of the crucible.

In Figs. 1 A, 1 B, and 1 C can be seen the visible physical manifestations indicative of a crystal growth. The specimen shown in Fig. 1 A has a very sharp line across the center, representing a twinning plane that is very easily seen when the ingot is cut and retched.

Since it had been noted that growth from the initial nucleation area continued both in horizontal and vertical directions, an investigation was carried out to determine the actual physical pattern that existed in the vertical direction during growth. Visually, the pattern on the surface can be observed, but this pattern is slightly distorted by surface tensions and is not the growth truly indicative of the natural plane extension that exists when the solid face is grown directly in contact with the liquid from which it is being solidified.

To determine the actual physical appearance of the growth planes, a study was carried out by "going fishing." We actually had a double hooklike apparatus made from quartz. With this we were able to get under the nucleated area and lift it quickly from its surroundings. The single crystal formation of germanium was a completely developed octahedron. Clear planes of growth were present, and these had clear-cut



FIG. 2. A, B, and C: fished samples showing clearly the crystalline structure.

angles associated with them. Apart from the pure geometric beauty exhibited by these "fished" single crystals, the geometric patterns could be studied for crystal growth orientation.

Figs. 2 A, 2 B, and 2 C show quite clear crystal structures which have been "fished" out from their growth environment very quickly. There is some disorder associated with the faces shown, but this is due to adherence of small amounts of germanium to the growing planes. When the growth is "fished" out these small amounts of germanium quickly freeze in a disordered fashion. They are not found when surfaces grow naturally without being subjected to rapid dislocation and extreme changes in temperature.

We regret that we cannot supply samples of these "fished" crystals, for our supply is limited.

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The Pirani Gauge

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The wide application of the Pirani gauge (1) as a heat conductivity manometer is evident from the excellent review of the literature by Dushman (2). Chief among the advantages and disadvantages of such a device for measuring pressures in the molecular and transmolecular regions are those given by DuMond and Pickels (3), von Ubisch (4), and others (2, p). 368), summarized in the following statements: (1) It is most suitable at very low pressures; (2) thin filaments insure a quick response at low pressures and a greater sensitivity at higher pressures; (3) materials available for filaments are sufficient so that pressures of many gases, corrosive and otherwise, may be measured; (4) by special design the instrument may be used to measure pressures over a wide range. The main disadvantages are (1) The shift of the "zero" point and (2) the increase of the ratio of radiation loss at pressures below 10⁻³ mm Hg, since at these low pressures the molecules of gas travel in straight lines between the wire and the walls, just as the photons of radiation do.

The range of pressures for which the Pirani gauge operates satisfactorily is from 10^{-4} mm to 10^{-1} mm of Hg (2, p. 367). Some experimenters, however, have extended this range to a lower limit of 10^{-5} mm (5) and to an upper limit of 50 mm (6) by modifying the design of the gauge.

The main disadvantage of the Pirani gauge, which has heretofore restricted its lower limit to 10^{-4} mm because of the so-called zero drift, has been studied by Kersten and Brinkman (5). Their solution to the problem is the construction of three identical gauges, two of which are exhausted, outgassed, and sealed off, and the third is connected to the vacuum system. By inserting one of the sealed-off gauges, called the ref-

erence gauge, and the measuring gauge in adjacent arms of a Wheatstone bridge, these authors obtained a balance point of the bridge which is indifferent to fluctuations of the bridge potential or to small changes in the temperature of the surroundings of the gauge. The second sealed-off gauge, called the zero gauge, is mounted in a separate branch of the circuit and heated to about the same temperature as the reference gauge. With such an arrangement the time required to attain equilibrium was very small. Since the heating of the measuring gauge was continued, it was possible to start the pressure measurements immediately after finishing the zero point control. With this arrangement and an applied heating current at the ultimate vacuum of 6.4 ma the deflection of the bridge meter was directly proportional to the gas pressure from 0 up to about 0.02 mm Hg. Beyond this region the sensitivity of the gauge-bridge combination decreased rapidly with increasing pressure.

In order to extend the use of the Pirani gauge to regions of higher pressures Rittner (7) applied the controversial point of the influence of wire-to-wall distance as a determining factor in the pressure measuring range. He used a convenient length of 1 mil tungsten wire and suspended it coaxially inside a piece of 2 mm capillary Pyrex, fitted with a glass side tube. The gauge was operated at constant resistance in a Wheatstone bridge by varying the applied voltage and measuring the total bridge current required to establish balance. The gauge was immersed in an ice bath. The author claims a precision of measurement of $\pm 2.5 \times 10^{-4}$ mm at low pressures, where the sensitivity was greatest, and ± 0.35 mm at 15 mm, where the gauge sensitivity was falling rapidly. This is equivalent to a precision of $\pm 2.5\%$ or better over the pressure range 10μ to 15 mm.

Von Ubisch (8, 9) modified the electrical circuit of the Pirani and thereby measured pressures up to 50 mm Hg. The gauge is an extended spiral of 0.01 mm tungsten wire of approximtaely 5 cm length mounted inside a capillary. A regenerative feedback is provided through the bridge containing the hot wire. The amplitude of oscillations adjusted itself to keep the temperature of the wire approximately constant at all pressures. The range of heating current extended from 4 to 35 ma for a corresponding pressure range of 10^{-3} mm to 50 mm Hg.

The effect of wire-to-wall distance in the measurement of pressures of rarefield gases is a controversial problem (10). Kersten and Brinkman (5) quote Knudsen's theory that the use of the hot wire gauge is based upon the principle that the thermal conductivity of a gas is directly proportional to the pressure as long as the mean free path of the molecules is larger than certain distances within the measuring system. Von Ubisch (11) confirmed the fact that in order to insure proportionality between pressure and heat conductivity, it is sufficient that the diameter of the wire be small compared with the mean free path of the molecule. He observed a deviation of 10% from