component was observed in the ERG. In this respect the parietal eye resembles the eye of the scallop, *Pecten irradians,* which has a vigorous "off" response but lacks an "off" component in the ERG (*11*).

Further investigation of the parietal eye, which has only two neural cell types (photoreceptors and ganglion cells), may contribute to a clearer understanding of retinal processes. Techniques are now being developed to study the action spectra of the eyes and to isolate the responses of single units (*12*).

WILLIAM H. MILLER
Rockefeller Institute,
New York, New York

MYRON L. WOLBARSHT
Naval Medical Research Institute,
National Naval Medical Center,
Bethesda, Maryland

References and Notes

1. M. Nowikoff, *Z. wiss. Zool.* **96**, 118 (1910).
2. W. Steyn, *J. Roy. Microscop. Soc.* **79**, 47 (1960).
3. R. M. Eakin and J. A. Westfall, *J. Biophys. Biochem. Cytol.* **8**, 483 (1960).
4. W. E. Ritter, *Bull. Mus. Comp. Zool. Harvard* **20**, 209 (1890).
5. A. Dendy, *Phil. Trans. Roy. Soc. London* **201B**, 227 (1911).
6. R. C. Stebbins and R. M. Eakin, *Am. Museum Novitates* **No. 1870**, 1 (1958).
7. J. J. Walsh, *J. Cellular Comp. Physiol.* **4**, 379 (1934).
8. R. M. Eakin, W. B. Quay, J. A. Westfall, *Z. Zellforsch. u. mikroskop. Anat.* **53**, 449 (1961).
9. M. L. Wolbarsht, E. F. MacNichol, Jr., H. G. Wagner, *Science* **132**, 1309 (1960).
10. H. K. Hartline, *Am. J. Physiol.* **121**, 400 (1938).
11. F. Ratliff, *Biol. Bull.* **111**, 310 (1956).
12. This investigation was supported in part by a research grant (B-864) from the National Institute of Neurological Diseases and Blindness, U.S. Public Health Service, and by contract Nonr 1442(00) with the Office of Naval Research (H. K. Hartline, principal investigator).

14 November 1961

Thermodilution Method for Measuring Cardiac Output of Rats by Using a Transistor Bridge

Abstract. Featuring an interlocking bridge amplifier, our new method measures cardiac output in animals from man to the rat, and one can perform many multiple assays with safety in the same animal with accuracy and reliability as reported, using room-temperature saline, whether the animal is in the normal or hyperthermic condition.

Numerous workers (*1, 2*) have established the thermodilution method to be sufficiently accurate and reliable for measuring the cardiac output of dogs, it being comparable to the Fick and dye-dilutions methods. However, the limited sensitivity of these methods has required a relatively large quantity of very cold saline for injection, which limits their use to larger animals for study. Our method features significantly greater sensitivity, requires only 0.05 ml of room-temperature (about 25°C) saline for injection, and can be used with animals as small as a rat or even a mouse or hamster. Although the principle has been utilized for recording dilution curves in dogs and human beings (*3*), this report describes the instrumentation and more exacting procedure required for the study of the albino rat.

The circuit employed is a transistor-amplified dual interlocking bridge as shown in Fig. 1, with an amplification of 20 to 90, depending on the transistor selected. Both interlocking bridges are balanced simultaneously by the 1000-ohm, ten-turn potentiometer. The output connects to a medium- or high-gain commercial d-c amplifier and recorder of almost any type.

The switch S-1 can be turned to either the calibrating or recording position, so that by placing the thermistor in a solution of measured temperature one can adjust the 5000-ohm potentiometer to give an identical pen position on the recorder for that selected temperature, such as 37.4°C, for example. The switch S-4 momentarily shorts the output to null to verify accurate bridge balance, and the other switches, S-2 and S-3, are the contacts of a multiple switch to simultaneously connect the power supplies.

Any of several transistors can be used for Q-1 if they are PNP types of relatively high gain. For example, CK722, CK721, 2N107, 2N190, 2N1265, and 2N109 have been found satisfactory by test, especially the 2N109. Exhaustive experimentation has shown the circuit to be remarkably stable and linear if a reasonable precaution is used to select a transistor with low noise and a low open-circuit current. Transistor testing devices are useful, but trial tests in the circuit are more practical and pertinent. It should be especially observed that the transistor input connection is not, and should not be, grounded. There should be a single ground connection at the output as indicated. It may be desirable to adhere the case of the transistor to a metal plate, or embed it in plastic within a small metal receptacle attached to a metal plate or the chassis. Though not necessary for ordinary environmental fluctuations, this added heat sink

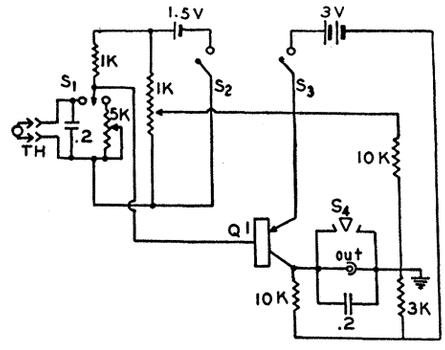

Fig. 1. Diagram of the preamplifier and interlocking bridge used with the measurement of cardiac output by thermodilution.

tends to damp and integrate any temperature fluctuations of the transistor over long measurement periods.

When a Sanborn Twin-viso d-c amplifier and recorder was used, at full gain setting (X1), a temperature change of 0.05°C gave a 40-mm deflection (80 percent of full scale). Other ranges can be used for calibration or measurements; a 1.0°C temperature change gave a 40-mm deflection with a gain attenuation of X20. Our temperatures were calibrated with a Bureau of Standards thermometer. With assays of different thermistors, we found that thermistors rated at about 2000 ohms (500 to 5000) at 25°C were most effective and efficient in this particular circuit, when used in the tip of a PE-10 polyethylene plastic tubing. The time constant of a well-constructed 2000-ohm thermistor was 0.4 sec, well beyond the required response speed for cardiac output measurements.

Fifty albino rats, weighing 350 to 550 g and anesthetized with pentobarbital sodium (24 mg/kg) injected intraperitoneally, were used in the animal studies. With each animal a 6-cm-long PE-50 catheter was inserted in the jugular vein to the right atrium, and the PE-10 thermistor-tipped tubing was passed down the right carotid artery to the aorta. For each assay, 0.05 ml of normal saline at room temperature was injected into the right atrium. This venous catheter was allowed to fill with blood before each injection, and a volume correction was made for this dead space. Between injections, the circulating blood was allowed to equilibrate thermally. Calculations of cardiac output were made by using the modified and corrected formula of Fegler (*1*).

The cardiac outputs of the total group of animals ranged from 196 to 299 ml/min per kilogram, and averaged

238 ml/min per kilogram. In a series of 14 animals where 15 assays of cardiac output in each animal at 2-minute intervals were measured in serial succession, the animal of widest variation was ±6 percent from the mean output of 133 ml/min, ranging from 126 to 141 ml/min, which demonstrates considerable reliability for the method.

In a more critical series of serial assays, 63 successive measurements of cardiac output were made at 2-minute intervals in each rat. The average cardiac output was 249 ml/min per kilogram, and the maximum range from the mean for each animal was ±9 percent, with a standard deviation of 6 percent. The last seven assays per animal showed a cardiac output per animal only 4 percent greater than the output on the initial seven assays; the difference was not significant. With our method, only about 3 ml of saline were added to the circulating blood during more than 2 hours. The values compare favorably with those of our other series and with other techniques (*4*).

A practical feature of this circuit is that it can be balanced to any temperature within physiologic range, including hyperthermia. To demonstrate the ability to measure under normal and hyperthermic conditions, we studied 28 rats before and after exposure to 2450-Mcy microwaves. Individual exposures were maintained until rectal temperatures were increased to 40°, 40.5°, and 41°C. Each animal served as his own control and three measurements were made before and after each thermogenic irradiation. The data are summarized in Table 1. The average cardiac output increased 26 percent at 40°C, 65 percent at 40.5°C (demonstrating the highest elevation of output), and 47 percent at 41°C. These three separate conditions were significantly different at a 1-percent level of confidence or better. The heart rate and arterial blood pulse pressures were also elevated above the control level in the three hyperthermic conditions.

Table 1. Effects of microwave hyperthermia on cardiac output of albino rats. The rats weighed 370 to 530 g. Cardiac output is expressed in ml/min per kilogram of body weight.

	Control rats	Experimental rats	Increase (%)
	Rectal temperature 40.0°C		
	221	273	23
	226	283	25
	263	343	30
	240	304	27
	244	295	21
	208	268	29
	214	269	27
	242	302	25
Av.	232	292	26
	Rectal temperature 40.5°C		
	228	400	75
	196	332	68
	226	336	49
	227	422	86
	224	346	55
	229	393	71
	267	420	57
	268	429	60
	275	459	67
	245	418	71
	268	434	62
	269	426	58
Av.	242	401	65
	Rectal temperature 41.0°C		
	249	357	43
	232	381	64
	247	362	46
	248	344	39
	254	360	42
	224	362	61
	268	386	44
	234	326	39
Av.	244	359	47

To further test accuracy and reliability, we constructed a physical system so that saline at 37°C flowed by an injection site for room-temperature saline, with a sensing thermistor downstream to measure flow rate by thermodilution, the latter being calibrated by a graduated cylinder receptacle. With an average fluid flow of 34.9 ml/min, the measured flow by our thermodilution was 35.1 ml/min, a difference of 0.2 ml/min, or 0.6 percent, which was not significant.

The described thermodilution method is practical to measure cardiac outflow in very small animals, being sensitive, accurate, and reliable. It offers the new possibility of numerous successive assays in the same animal, is practical for larger animals such as dog and man, and is inherently safe to employ (*5*).

ALFRED W. RICHARDSON
THEODORE COOPER
TERESA PINAKATT
Departments of Physiology and *Surgery, St. Louis University School of Medicine, St. Louis, Missouri*

References and Notes

1. G. Fegler, *Quart. J. Exptl. Physiol.* **42**, 254 (1957).
2. A. V. N. Goodyer, A. Huvos, W. F. Eckhardt, R. Ostberg, *Circulation Research* **7**, 432 (1959); A. Fronek and V. Ganz, *ibid.* **8**, 175 (1960); E. Rapaport and S. G. Ketterer, *ibid.* **6**, 214 (1958).
3. T. Cooper, E Braunwald, G. C. Riggle, A. G. Morrow, *Am. J. Cardiology* **6**, 1065 (1960).
4. F. R. Blood, D. L. Smith, F. E. D'Amour, *Am. J. Physiol.* **163**, 268 (1950); R. W. Bullard, *Federation Proc.* **15**, 28 (1956); L. A. Sapirstein, *Am. J. Physiol.* **193**, 161 (1958).
5. This work was supported by contract NR 102-362, from the Office of Naval Research.

31 August 1961

All-or-None versus Incremental Learning of Errorless Shock Escapes by the Rat

Abstract. Investigation showed that in a two-choice correction procedure the probability of an errorless response in those rats which have consistently made errors is not constant, as required by the all-or-none model, but increases with trials. Even an all-or-none model which provides for accidental error and accidental success is inadequate.

The growth function of Hullian and more recent linear models of learning assumes that successive reinforcements add increments of probability of response which are a constant fraction of what potentially remains to be learned, that is,

$$P_{n+1} - P_n = \theta(a - P_n),$$

where θ is a constant and a is the asymptotic value approached by P_n. The solution of this difference equation is given by

$$P_{n+1} = a - a(1 - \theta)^n$$

for $i = 0, 1, 2, \ldots, n$. Such a function adequately describes group data, for example, proportions of subjects succeeding on each successive trial (for instance, see Table 1), but the fit to individual data has been questioned. The behavior of individual subjects may be more consistent with a discontinuous all-or-none principle, each successive trial affording the same, constant probability of a subject's forming the appropriate habit. The proportion of subjects with their first conditioned eyelid response, and first correct paired associate, remains constant from trial to trial (*1*). However, these data came from human subjects. It is possible that this flip-flop nature of the probability of success might be peculiar to thinking and talking subjects: eyelid and verbal responses may develop with positive feedback, similar to the feedback provided for the galvanic skin response (*2*). To test this possibility we examined the conditioning of rats, which appear to have at most only one postural symbol or idea at any given time (*3*).

In experiment 1, 112 Sprague Dawley and hooded rats were individually restrained with their heads extending over photoelectric cells (*4*). Shock current (a-c) through the tail was increased toward a maximum of 5 ma (*5*). As soon as the head position became more than 50 deg to the left or the right of center, the photocells