may in some instances overcome the difficulties encountered in the usual histochemical procedures. By substantially decreasing the loss of enzyme, they may greatly shorten the time required in the incubation medium to bring out the activity of a given structure. The advantage of this in the azo dye method is apparent since the life of the diazonium compound is limited and the enzyme operates at suboptimal temperatures. That it may have advantages in the Gomori method, too, is suggested by recent work of Meath and Pope (28), who report that the most intense acid phosphatase staining occurs in the nuclei when the sections are of the customary fixed tissue. but in the cytoplasm when of unfixed frozen tissue.

It is too early to predict the extent to which omission of fixation and embedding can overcome the relative insensitivity of the azo dye method or the diffusion and adsorption difficulties complicating the Gomori method. These two methods at present are of little value for the localization of alkaline phosphatase on the intracellular level, except for structures with extremely high activity such as the brush borders of the kidney. For the moment it would appear that the actual isolation of intracellular constituents, although itself not free of pitfalls, offers the only means of localizing the enzyme inside the cell. The histochemical methods can, however, be used to advantage in indicating some pitfalls in the isolation method.

#### References

- 1. NOVIKOFF, A. B., PODBER, E., and RYAN, J. Federation
- Proc., 9, 210 (1950). GOMORI, G. Proc. Soc. Exptl. Biol. Med., 42, 23 (1939). TAKAMATSU, H. Trans. Soc. Path., Japon., 29, 492 3.
- (1939).NOVIKOFF, A. B., PODBER, E., and RYAN, J. Unpublished. 4.
- 5.
- 6.
- A. B., FOBER, E., and RARN, J. Unpublished.
  GOMORI, G. Am. J. Clin. Path., 16, 347 (1946).
  KRUGELIS, E. J. Biol. Bull., 90, 220 (1946).
  ROBERTSON, W. V. B., DUNIHUE, F. W., and NOVIKOFF,
  A. B. Brit. J. Expti. Path., 31, 545 (1950).
  DANIELLI, J. F., and CATCHESIDE, D. G. Nature, 156, 7.
- 8. 294 (1945).
- 9. SCHMIDT, G., and THANNHAUSER, S. J. J. Biol. Chem. 149, 369 (1943).
- 10.
- DANIELLI, J. F. J. Exptl. Biol., 22, 110 (1946). KURNICK, N. B., and MIRSKY, A. E., J. Gen. Physiol., 33, 11 265 (1950).
- 12. BRACHET, J., and SHAVER, J. R. Stain Technol., 23, 177 (1948).
- GOMORI, G. J. Lab. Clin. Med., 35, 802 (1950). 13.

- MANNHEIMER, L. H. and SELIGMAN, A. M. J. Natl. Cancer Inst., 9, 181 (1948). MENTEN, M. L., JUNGE, J., and GREEN, M. H. Proc. Soc. Exptl. Biol. Med., 57, 82 (1944). NEWMAN, W., et al. Am. J. Path., 26, 257 (1950). LISON, L. Bull. histol. appl. physiol. et path. et tech. 18.
- 19. 20.
- Microscop., 25, 23 (1948). MARTIN, B. F., and JACOBY, F. J. Anat., 83, 351 (1949). WILLMER, E. N. J. Exptl. Biol., 19, 11 (1942). 21.
- 22. 23. BRACHET, J., and JENEER, R. Biochim. et Biophys. Acta,
- 2, 423 (1948). MORSE, A., and GREEP, R. O. Anat. Record, 99, 379 24. (1947).
- 25. DEMPSEY, E. W., and DEANE, H. W. J. Cellular Comp. Physiol., 27, 159 (1946).
- BOURNE, G. Quart. J. Exptl. Physiol., 32, 1 (1943) 26.
- GOMORI, G. Proc. Soc. Exptl. Biol. Med., 69, 407 (1948) 27.
- MEATH, J. A., and POPE, A. Federation Proc., 9, 204 (1950).

# Average Body Temperature in Mice<sup>1</sup>

## I. S. Hart

#### Division of Applied Biology, National Research Laboratories, Ottawa, Canada

Estimates of average body temperature have been made for human beings (1) from skin and rectal temperatures. Few attempts appear to have been made to measure or estimate the average temperature of small mammals or birds, although determinations have been made of rectal, skin (2), esophageal (3), and subcutaneous temperatures (4, 5). In the present study a method was developed to investigate interrelations between the average body temperature of mice and environmental conditions, particularly during exposure to low temperatures.

The method<sup>2</sup> adopted was to place a mouse immediately after death in a Dewar flask calorimeter (218ml) containing 100.0 g of water at a known temperature, record the temperature change, and calculate the average temperature (method of mixtures) after applying the thermometer and "radiation" corrections. The temperature change was determined by thermometers divided into intervals of 0.1° C, but estimations to 0.01° C were made with the help of a lens. Each thermometer was fitted with a No. 11 rubber stopper that closed the Dewar flask, and the lower end of the thermometer bulb rested about 5 mm from the bottom of the flask. The best results were obtained by carrying out the tests in a constant-temperature room at approximately 20° C. The initial water temperature in the calorimeter was 20.5° to 21° C, and the final temperature 22° to 23° C for normal mice, or 19° to 20° C for chilled mice (body temperature lowered). "Radiation" corrections were made from the rate of change in final temperature by the Regnault-Pfaundler or the Dickinson methods (6). During the tests, stirring of the water in the calorimeter was accomplished by means of a mechanical shaker. It took about 30 min to reach the final temperature after introducing the mouse. After mice were killed and equilibrated to a constant temperature in an air bath, the average body temperature did not differ significantly from the temperature of the bath when 0.83 was used as the specific heat of the mice. The standard deviation of determinations on 15 mice so tested was  $\pm 0.25^{\circ}$  C.

Tests to determine whether post-mortem heat production contributes appreciably to the results furnished evidence that heat production after death is small or negligible compared to the heat present in the body. The evidence is: first, the consistency of the average body temperatures, which generally fell between skin and rectal temperatures; second, killing mice by KCN injections gave similar results to killing by percussion; third, thermal equilibrium with the environment is established rapidly (1 to 2 hr) after

A more detailed description is in preparation.

<sup>&</sup>lt;sup>1</sup> Preliminary note.

killing; fourth, the rate of cooling of mice killed by percussion was not slower than that of the same mice after being reheated about 2 hr later.

A comparison of rectal and average temperatures for resting albino mice held 1 to 2 hr at various environmental temperatures (0°, 10°, 20°, 30°, and 40° C) is shown in Fig. 1. After each mouse had become

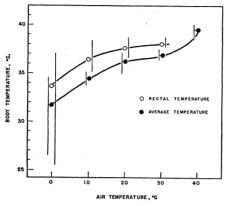


FIG. 1. The effect of environmental temperature on average body temperature and on rectal temperature.

quiet again, following removal of the thermocouple wires, it was killed by percussion and immediately placed in a calorimeter. These tests showed that both rectal temperatures and average body temperatures were relatively constant over only a small range of environmental temperatures. At each environmental temperature, rectal temperatures averaged 1° to 2° C higher than average temperatures, and the difference tended to be greater at lower air temperatures. The range of both average and rectal temperatures is shown by vertical lines in Fig. 1. This variation increased enormously at 0° C.

Determinations of rectal and average body temperatures have also been made on mice running in a treadmill at  $10^{\circ}$  and  $30^{\circ}$  C. At  $10^{\circ}$  C rectal temperatures ranged from  $34.2^{\circ}$  to  $38.0^{\circ}$  (averaging  $36.2^{\circ}$  C), whereas average temperatures of the same mice ranged from 31.1° to 35.5° (averaging 33.8° C). At 30° C rectal temperatures ranged between 40.7° and 44.0° (averaging  $42.1^{\circ}$  C), whereas average temperatures varied from 39.4° to 42.5° (averaging 40.1° C). It is evident that at 10° C there was very little difference in body temperatures during rest and activity; but at  $30^{\circ}$  C rectal temperatures averaged  $4.0^{\circ}$  and average body temperatures 3.5° C higher than corresponding values during rest. This behavior of body temperature during activity differed from the behavior of metabolism (7), which has been found to increase by approximately the same amount at different temperatures for the same degree of activity.

For mice chilled to the point of death, rectal temperatures gave no indication of average body temperatures. In 8 mice rectal temperatures at death varied from 5.0° to 14.5° C, but average temperatures were always higher than rectal temperatures by amounts ranging from  $0.4^{\circ}$  to  $4.3^{\circ}$  C. Evidently the rectal region cools to a greater extent than other regions of the body under these conditions.

Average body temperature determination is useful as a research tool for supplementing other temperature measurements in mice and possibly other animals. A major disadvantage is that a series of measurements cannot be made on the same animal. For this reason, the method is perhaps most applicable to lethal temperature studies, but it can be applied in other studies when large numbers of animals are available.

#### References

- BURTON, A. C. J. Nutrition, 9, 261 (1935). 1.
- 2
- 3.
- BURTON, A. C. J. Nutrition, 9, 261 (1935).
   ADOLPH, E. F. Am, J. Physiol., 161, 359 (1950).
   KENDEIGH, S. C. J. Mammal., 26, 86 (1945).
   BULLOUGH, W. A. J. Expit. Biol., 26, 76 (1949).
   VERNET, S., and METCALF, K. F. Science, 107, 655 (1948).
   DICKINSON, H. C. Natl. Bur. Standards (U. S.), Tech.
   News Bull., 11, 189 (1915).
   HART, J. S. (an. J. Research, D28, 293 (1950). 4 5.

7. HART. J. S. Can. J. Research, D 28, 293 (1950).

## Cow Feces and Chick Development

### H. W. Marlow<sup>1, 2</sup>

### Department of Biochemistry, Southwestern Medical College, Dallas, Texas

An extract of pregnant cow feces gave Rubin and co-workers (1) a chick growth factor. Hot water, 50%alcohol, or 95% alcohol extracted appreciable quantities of this factor. These workers also found (2) that their factor was soluble in 50% acetone and that it could be extracted with ammoniacal alcohol. It is stable to autoclaving at neutral pH, but is rapidly destroyed by autoclaving with 2N HCl. It may be precipitated by adjusting the pH to 3. Whitson and others (3) also extracted from cow manure a chick growth factor not identical with any known growth vitamins. Turner (4) and Riley et al. (5) not only demonstrated the presence of a chick growth factor in pregnant cow feces but of an orally effective androgen as well. McGinnis et al. (6) believe that the growth factor can be synthesized in hen feces upon incubation at  $30^{\circ}$  C for 72 hr.

Agreement as to the existence of a chick growth factor appears to be universal. It seemed desirable to explore the field further by studying the effects of an alcoholic extract of the feces on the comb size and body weight of young chicks of both sexes and on the testis size of male chicks.

Feces from pregnant cows were extracted 3 times with 95% alcohol in the cold by agitation. The alcohol was filtered off, and the alcohol-insoluble fraction was dried in a warm place and ground fine. Twenty per cent weight of the extracted feces was added to 80%of Purina growing mash. A preliminary experiment was devised as follows: One lot of single-comb White Leghorn male chicks was started on Purina growing mash as a control, and another lot was started on the

<sup>&</sup>lt;sup>1</sup> Present address: Veterans Administration Hospital, Mc-Kinney, Texas,

<sup>&</sup>lt;sup>2</sup> Grateful acknowledgments are made to Marion Bucklin and Betty Little for their aid in obtaining the data and in the preparation of the paper.