hour periodicity determined by them. The combination of a circadian rhythm and "Zeitgebers" is established so firmly that it buffers changes and introduces a marked time lag in modification made by a shift of daily routine as a result of a change in local time.

Observations by previous investigators on the temperature rhythm during a voyage showed a satisfactory adjustment to day-to-day change of longitude, while the present observations revealed a time lag. The difference is accounted for a wide difference in the speed of transposition across the longitudes. Fig. 3 is a schematic representation of the relation of a change in local time to a shift of body temperature pattern. So long as the speed of transposition is slow enough(1,2,4) and the resulting change of actual length of a day is less than 25 minutes, the 2 curves coincide well (Group A in Fig. 3). An east-bound transpacific voyage at a faster speed which resulted in a reduction of a day by 32 minutes revealed that the diurnal rhythm could not follow the day-to-day change of local time (Group B in Fig. 3). A rapid trip leaves a great amount of time lag (Group C, Fig. 3). During the first few days the adjustment to the new local time took place slowly, then proceeded more rapidly. The rate of adjustment shown here is considered to be the upper limit man can attain. In the adult the average was 50 minutes per day in subject T and 39 minutes in subject S, while in the child the rate was much higher.

*Summary.* The diurnal pattern of body temperature was observed on 3 members of a family including a 6-year-old child for 2 weeks covering the period during and after an east-bound flight which resulted in an advance of daily routine as much as 10 hours. Adjustment of diurnal rhythm to a new local time was discussed with reference to the speed of transposition. With a rapid shift of local time zones the characteristics of the normal pattern were disturbed, but gradually approximated the new time pattern (approximately 40-50 minutes shift per day).

The author wishes to thank Dr. Loren D. Carlson for advice and support in this study.

1. Gibson, R. B., *Am. J. Med. Sci.,* 1905, v129, 1048.
2. Osborne, W. A., *J. Physiol.,* 1908, v36, 39 P.
3. Sasaki, T., *Bull. Res. Inst. Diath. Med.* (Kumamoto, Japan), 1953, v3, 262.
4. Ogata, K., Sasaki, T., *Jap. J. Physiol.,* 1963, v13, 84.
5. Halberg, F., *et al.,* Publ. No. 55, *Am. Assn. Adv. Sci.,* 1959, 803.
6. Aschoff, J., *Naturwiss.,* 1954, v41, 49.
7. Miyazaki, M., cited in (4).
8. Palmai, G., *Med. J. Austral.,* 1962, v2, 989.

Received December 4, 1963. P.S.E.B.M., 1964, v115.

## A Simple Method of Obtaining Venous Blood from Small Laboratory Animals. (29134)

DAVID A. SORG AND BEVERLY BUCKNER (Introduced by Walter D. Block)
*Caylor-Nickel Research Foundation, Bluffton, Ind.*

Stone(1) reported a technique by which venous blood could be obtained from the orbital sinus in a mouse or rat. As described, the method required the puncture of the venous sinous behind the eyeball with a glass tube which had been drawn out to a capillary tip. It was originally suggested that the capillary end be introduced into the anterior angle of the eye and it was claimed that over 1 ml of blood could be obtained.

Although puncture made into the anterior angle will produce blood flow, nose bleeds and eye trauma frequently result. The authors find it far more satisfactory to use the posterior or outer canthus. Also, the use of standard microhematocrit capillary tubes further simplifies the technique.

With the method described below, repeated punctures can be made with a high degree of safety on rats, mice, guinea pigs, rabbits

and hamsters, without the need of syringes, needles or transferring of blood.

*Materials and methods.* Standard heparinized microhematocrit capillary tubes are broken into thirds. The sharp (broken) end should not have too long a bevel. With the animal held firmly or anesthetized, the sharp end of the tube is introduced into the outer or posterior canthus of the eye, under the nictitating membrane. With a slight twist, it is directed straight in, approximately perpendicular to the surface plane of the eyeball until a slight "give" is noted. Usually, the blood will flow freely and may be collected directly into a test tube. This is easily done by turning the animal over and holding the head down; the tube will remain fixed in place. Occasionally, twisting, advancing, or slightly withdrawing the capillary tube will facilitate better flow. When the desired amount of blood is collected, the tube is withdrawn and slight pressure on the eyeball is used to prevent further bleeding.

*Discussion.* The only difficulty encountered in this technique is occasional clotting of the blood in the capillary tube, but, even then, blood will often flow freely around the outside of the tube, which is satisfactory for most procedures. Although the collection is more convenient with the animal anesthetized, it may be done readily without anesthesia. Exsanguination of a 115-130 g rat will yield 4-6 ml of blood and in large rats more than 8 ml can be obtained without sacrificing the animals.

Serum protein analysis performed by the Weichselbaum(2) modification of Kingsley's biuret method on samples of blood obtained by this procedure did not vary significantly from specimens obtained by cardiac puncture.

*Summary.* A modification of a previously reported method of obtaining venous blood from small laboratory animals is described. A short piece of heparinized capillary tubing is used to puncture the orbital sinus at the posterior angle of the eye. Sufficient quantities of blood for laboratory analysis are obtained from either anesthetized or nonanesthetized animals without ill effects.

1. Stone, S. H., *Science*, 1954, v119, 100.
2. Weichselbaum, T. E., *Am. J. Clin. Path.*, 1946, v16, (Tech. VII), 40.

Received December 6, 1963. P.S.E.B.M., 1964, v115.

## Type of Antibody Elevated by 5-Fluoro-2-deoxyuridine and Endotoxin During the Primary Response of the Mouse.* (29135)

KATHARINE MERRITT AND ARTHUR G. JOHNSON (Introduced by W. J. Nungester)
*Department of Microbiology, University of Michigan, Ann Arbor*

The observations that gamma globulins are a heterogeneous group of plasma proteins have led to the development of methods for characterization of the individual types. It has been found that the gamma globulins with a sedimentation constant of 19 are susceptible to cleavage by sulfhydryl containing compounds such as 2-mercaptoethanol(1). This has led to the use of these agents for a rapid and convenient differentiation of the molecular character of the antibody molecule.

Recent evidence has shown that the inbred mouse, Balb strain, will respond with antibody formation following a single injection of any of several purified protein antigens(2). In addition, it was shown that drugs capable of inhibiting nucleic acid synthesis can actually increase considerably the amount of anti-protein antibody synthesized by these mice (2,3). The conditions under which immunosuppressive drugs could act as adjuvants were compared with those characteristic of the adjuvant action of bacterial endotoxins. The

* This investigation was supported by research grant from U.S.P.H.S.