frozen sections of liver cells and renal tubules lining cells.

2. When the tissue contains a PAS-positive lipid which is insoluble in acetic anhydride-pyridine, as appears to be the case in arterial walls and capillary basement membranes.

3. When the tissue contains sphingolipids.

The strongly positive PAS staining of sphingolipids can be attributed to the presence of the amino and hydroxyl groups on adjacent carbon atoms of the sphingosine moiety. In cerebrosides, the hexose group provides an oxidative point, while the double bond of the unsaturated fatty acid serves further to strengthen the reaction. The reduction by acetylation of the staining ability of Niemann-Pick cells preserved in formalin may be atributed to the oxidation of this material during storage. Dihydroxy groups probably appear at the double bond sites as a result of the partial oxidation by atmospheric oxygen, and together with the sphingosine 1 amino 2 hydroxy groups can then be acetylated and thus protected from attack by periodate.§

Conclusions. It must be concluded that the PAS method cannot be used to differentiate Niemann-Pick from Gaucher cells, and that it is not strictly specific for carbohydrates. A positive reaction may be given also by sphingolipids, and possibly by other substances having an amino group and a hydroxyl group on 2 adjacent carbons, as well as by unsaturated lipids.

Summary. 1. Unsaturated lipids stain with Schiff's reagent after oxidation with periodate (PAS reaction). The reaction is not inhibited by previous acetylation according to the technic of McManus and Cason. 2. Sphingolipids stain with the PAS technic whether they contained a carbohydrate moiety or not. Their reactivity may be ascribed to the presence of a hydroxy and an amino group on vicinal carbon atoms of the sphingosine moiety, and to the double bond in the fatty acid radicle. Acetylation reduces but does not abolish the staining ability of sphingolipids, since the double bond is not affected by the procedure. 3. The PAS method is not suitable for the differentiation of Niemann-Pick from Gaucher cells. 4. The ability of liver cell granules and of some renal tubule cell granules in fresh frozen sections to stain with PAS was abolished by acetylation, and largely restored by deacetylation. The fact that the staining of reticulin fibers in walls of vessels was not completely prevented by acetylation is considered as indicative of the presence of unsaturated fatty acid radicles in these structures.

§ The light staining of the lecithin-cephalin mixture may be ascribed both to the double bond of the fatty radical and to the serine moiety of cephalin.

Received October 16, 1950. P.S.E.B.M., 1950, v75.

Arterial Volume Pulses and Peripheral Vascular Responses to Drugs. (18274)

RICHARD A. MCLEAN. (Introduced by Edwin J. Fellows)

From the Research Division, Smith, Kline and French Laboratories, Philadelphia, Pa.

The work of Nyboer(1), Bloom, Kaufman, Nims and Nyboer(2), and others has shown that peripheral vasoconstriction produces a decrease and vasodilation an increase in the blood volume pulse of a body segment or organ. Certain difficult problems of instrumentation arise in recording simultaneous volume pulses from a number of total organs.

1. Nyboer, J., (in) *Medical Physics,* Vol. 1, O. Glasser (Ed.), The Year Book Publishers, Chicago, 1944, 340; Nyboer, J. (in) *Medical Physics,* Vol. II, O. Glasser (Ed.), The Year Book Publishers, Chicago, 1950, 736.

2. Bloom, W. L., Kaufman, S. S., Nims, L. F., and Nyboer, J., Natl. Res. Council, Com. on Aviation Med., Rep. No. 246, January 1944.

Such measurements can readily be made on the arteries which supply the various vascular beds. If these arterial volume pulses reflect changes in the peripheral vascular network beyond them, information on the interrelated responses to physiological stimuli and vasoactive drugs may be obtained.

Method. In our laboratory, simultaneous measurements of this type have been made at a number of points in the circulation. Typical experiments include volume pulses from the renal, splenic, inferior mesenteric and femoral arteries in addition to the pressure pulse from the carotid artery. These have been obtained either by the impedance plethysmograph described by Nyboer(1) or by a photoelectric plethysmograph. In the first case the artery is placed in a lucite trough between two electrodes and uniform contact maintained with a conductile paste. A high frequency signal passed between the electrodes is modulated by the pulsatile electrolyte which represents the blood volume pulse. This is recorded by means of an impedance bridge. The circuit reported by Henny and Boone(3) and Oppenheimer and Chamberlain(4) for the Roentgen Electrokymograph was utilized in the photoelectric measurements. A small mirror is attached to the lower end of a lucite rod which is mounted to face an RCA 931-A photomultiplier. Another lucite rod conducts light from a small flashlight bulb and projects it onto the mirror at an angle suitable for reflection up the bar leading to the phototube. A segment of an isolated artery is interposed in this optical system by placing it on the mirror just below the converging ends of the lucite rods. Both diameter and opacity of the blood vessel modulate the light to yield a pulse tracing similar in form to that obtained with the impedance circuit or with a sensitive pressure plethysmograph. All blood pressure measurements were made from a carotid artery using a Sanborn Electromanometer. A modified Grass eight-channel electroencephalograph was the graphic recorder.

Vasoconstriction and vasodilation. A number of workers (1,2,5-9) have established the fact that the volume pulse of a segment or organ of the body is decreased by peripheral vasoconstriction and increased by vasodilation. Work in this laboratory indicates that the volume pulse in an artery supplying such a segment or organ responds in like manner. We have found that the volume pulse of the total leg as measured by electrical impedance and the volume pulse of the femoral artery measured photoelectrically respond in a comparable manner when cats anesthetized with pentobarbital sodium receive intravenous injections of vasoconstrictor or vasodilator agents.

Fig. 1 illustrates the relationship of mean arterial blood pressure to volume pulse in response to a constricting and dilating agent. It is to be noted that naphazoline hydrochloride (Privine) caused a reduction in femoral volume pulse which was accompanied by a rise in blood pressure, while N-isopropylarterenol (Isuprel) produced a marked increase in pulse height simultaneously with a reduction in mean blood pressure. The exact mechanism by which arterial volume pulses are reduced with peripheral constriction and increased with dilation is not entirely clear. Numerous factors are probably concerned. The volume of blood contained in the artery, as well as variations in the elasticity of the vessel which result from the volume changes, appear to influence the observed pulse height. In the case of the photoelectric pickup, a unit volume of blood added to a vessel already engorged with blood will produce relatively less change in opacity and in cross-sectional area under the photosensitive element than will the same volume of blood added to a less distended artery. Since the capacitative

3. Henny, G. C. and Boone, B. R., *Am. J. Roentgenol.*, 1945, v54, 217.

4. Oppenheimer, M. J. and Chamberlain, W. E., (in) *Methods in Medical Research*, Vol. I, V. R. Potter (Ed.), The Year Book Publishers, Chicago, 1948, 232.

5. Atzler, E. and Lehmann, G., *Klin. Wchnschr.*, 1935, v14, 1581.

6. Mann, H., PROC. SOC. EXP. BIOL. AND MED., 1937, v36, 670.

7. Rosa, L., *Z. f. Kreislaufforsch.*, 1940, v32, 118.

8. Fenning, C., *Am. Heart J.*, 1943, v25, 522.

9. Holzer, W., Polzer, K., and Marko, A., RKG. Rheocardiography, Wilhelm Maudrich, Vienna, 1946.

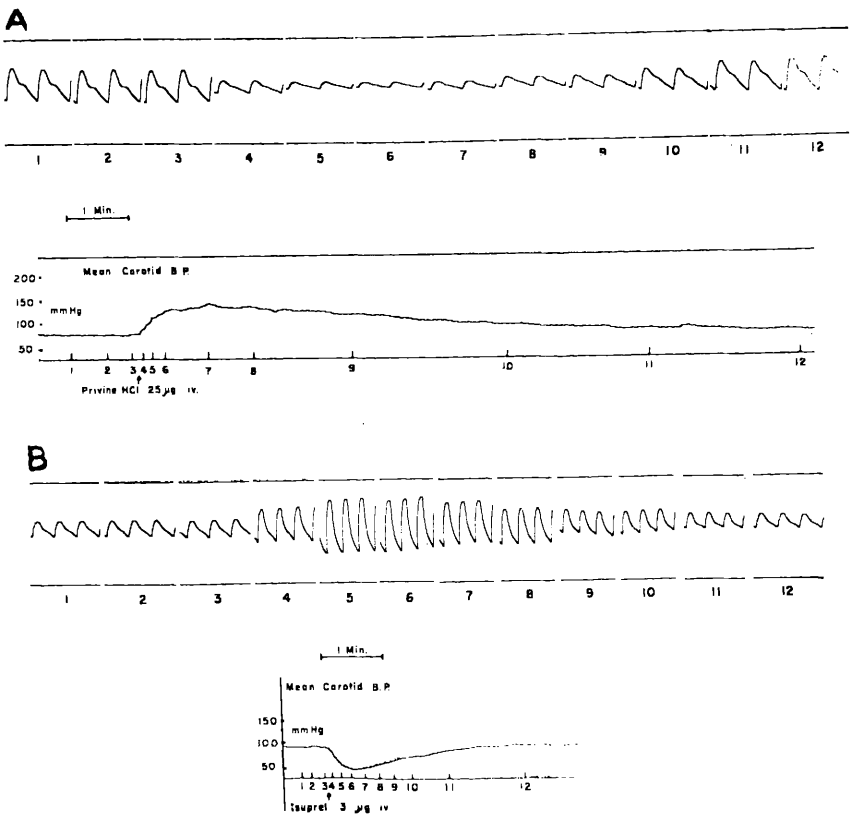

FIG. 1.

Simultaneous recordings of femoral arterial volume pulse and mean carotid blood pressure during circulatory responses to intravenous injection of a constricting (A) and a dilating (B) agent. The numbers under the pressure curves correspond to the samples from the electrical impedance pulse tracing which were taken at a faster paper speed.

coupling employed with the phototube circuit records change from a reference level but not total change, it will show the relative effect of each pulsation but will not indicate the degree of engorgement. To this influence of the geometric factor is added the loss of elasticity which occurs in a distended vessel. Therefore, increasing blood content will have a diminishing effect on the net distention produced by each pulse. Both of these factors probably influence the photoelectric measurements. In the electrical impedance determinations, the geometric relationship is not involved. In this procedure total electrolyte is balanced out by a series resistance between the bridge and the artery and the addition of a unit volume of blood electrolyte causes the same deflection of the galvanometer regardless of the degree of engorgement at the point of measurement. Therefore it is probably the decrease in elasticity of the distended vessel that is largely responsible for the reduction in recorded pulse height.

In a given vascular bed, vasomotor changes may occur as a result of mechanisms which involve the entire arterial system. Active constriction of the arteries and arterioles may of itself oppose the distention of the vessels by an increase in pressure. Conversely a decrease in tone may augment the effect of the pulse.

There is another factor which may contribute to a reduced volume pulse in an artery leading to a constricted peripheral vascular bed: the increased pressure in that vessel may limit the amount of blood entering from the aorta with each ventricular contraction. For example, if the vascular bed sup-

plied by the mesenteric vessels is constricted relatively more than other areas of the body, more resistance to flow is present and less blood (smaller volume pulse) may be expected to enter the mesenteric arteries from the aorta. At a uniform stroke volume, the magnitude of the central pressure pulse in the aorta is controlled by the algebraic sum of the various peripheral factors which oppose blood flow. In the arterial branches we believe these factors are specific for the vascular bed which a given vessel serves. With the extreme peripheral constriction, venous return may be considerably reduced. If this occurs, a reduction in the central pressure pulse width is seen as a result of a smaller stroke volume. This will reduce the volume pulse in the systemic arteries.

An increased volume pulse does not in itself indicate peripheral vasodilation. The effect of the heart must be taken into consideration. If a marked change in stroke volume or a positive inotropic effect does not take place, the peripheral response will occur in a consistent manner. In other words, the arterial volume pulse will increase with dilation and diminish with constriction. With a positive inotropic action such as is produced by epinephrine, an increase in aortic pressure pulse width takes place. This may be attributed to an augmented strength of ventricular contraction and perhaps also an increased venous return. Under these conditions, arterial volume pulses may be increased. Simultaneous records made in this laboratory with a volume pulse recorder and a bubble flow meter show that the increased arterial pulses are associated with an increase in blood flow. Perfusion experiments carried out in the same vascular beds indicate that in certain organs, epinephrine produces vasoconstriction. This apparent discrepancy is clarified if the drug is given intra-arterially or, for the multichannel experiments, into the aorta. Under these conditions, the drug affects the peripheral vessels before it reaches the heart. For somewhat more than one circulation time, the peripheral constriction is apparent and the arterial volume pulses are reduced. Subsequent cardiac stimulation often overshadows a peripheral constrictor action, since arterial volume pulses are increased as an increase in blood flow takes place.

In order to demonstrate the peripheral response in a discrete vascular bed, electrodes were placed on each side of a dog's nose in the region of high vascularity in the nasal passages. An impedance pulse was taken in the manner just described for the isolated artery. Topical application in the nasal passages of amphetamine sulfate or amyl nitrate caused responses of the vascular bed comparable to those noted in the femoral artery after intravenous injection of constricting or dilating agents. This method is routinely used in our laboratory for the evaluation of local vasoconstrictor compounds.

Blood flow. Hamilton and Remington(10) published a study relating cardiac output to the pressure pulse contour in the aorta. This method was elaborated in a further publication by the same authors and their associates (11) and their results substantiated by Huggins, Smith and Sinclair(12). This technic links the pressure pulse with blood flow. Studies in our laboratory show that an arterial volume plus also can indicate relative changes in blood flow. The question of back flow or retrograde flow appears to complicate this mechanism. However, in a peripheral artery, a relatively large backflow is unlikely. The relative volume of the arterial tree increases peripherally. Actual flow measurements made by Gregg and Green(13) on the coronary circulation, where the compression of the vascular bed by the contraction of the myocardium markedly accentuates resistance to flow during the systole, showed only about 10% backflow during the systolic period and none during diastole. Thus it appears that a volume pulse of an artery when related to heart rate can give an indication of blood

10. Hamilton, W. F., and Remington, J. W., *Am. J. Physiol.*, 1947, v148, 14.

11. Remington, J. W., Hamilton, W. F., Wheeler, N. C., and Hamilton, W. F., Jr., *Am. J. Physiol.*, 1949, v159, 379.

12. Huggins, R. A., Smith, E. L., and Sinclair, M. A., *Am. J. Physiol.*, 1949, v159, 385.

13. Gregg, D. E. and Green, H. D., *Am. J. Physiol.*, 1940, v130, 114.

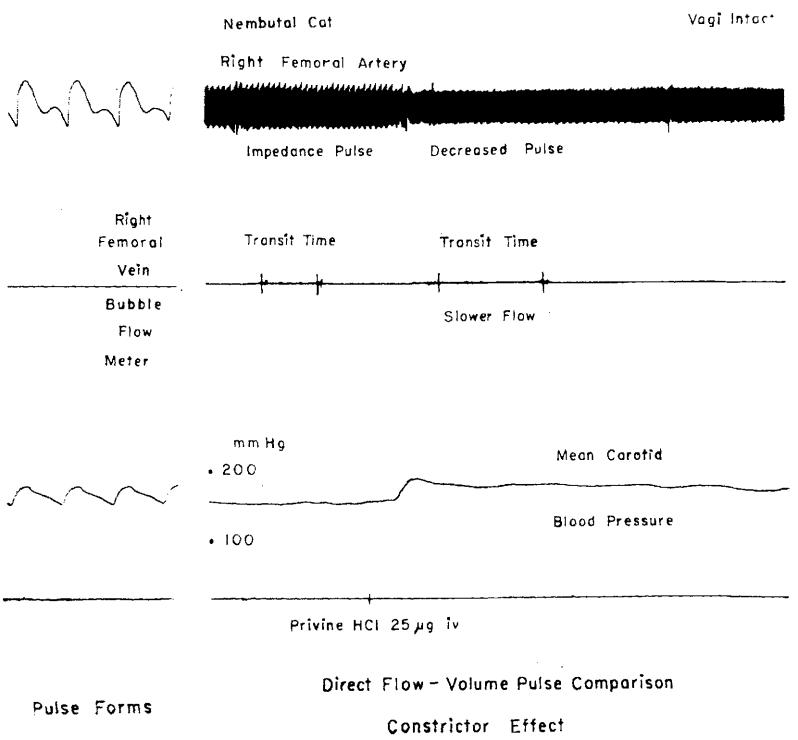

FIG. 2.

Direct Flow—Volume Pulse Comparison, Constrictor Effect. A small segment of record taken at a fast paper speed earlier in the experiment is included on the left to show the form of the impedance volume pulse which is condensed into an envelope by the slower paper speed.

flow through that vessel.

Bloom, Kaufman, Nims and Nyboer(2) published a method for calculating a flow index from a volume pulse tracing. The pulse height is considered proportional to the actual volume pulse. To include the diastolic factor a perpendicular is erected at the beginning of systole and a line drawn to represent the mean diastolic slope. This slope is proportional to the rate at which blood leaves the segment. This line is projected to intersect the perpendicular marking the beginning of systole and an expression of blood flow is obtained. The authors point out that this method does not distinguish between normal and backflow.

To demonstrate how pulse height recorded from an artery varies with peripheral resistance, simultaneous records of volume pulse from the femoral artery and blood flow in the femoral vein were taken along with mean carotid blood pressure. In Fig. 2 the response to an intravenous injection of 25 µg of Privine hydrochloride is illustrated. Here pressure increases while blood flow and pulse height decrease. In Fig. 3 is shown the effect of intravenous injection of 3 µg of Isuprel. The arterial volume pulse again varies inversely with peripheral resistance and reflects the dilation in the vascular bed which the artery serves. These pulses can be recorded from an unopened circulation without the use of an anticoagulant. This is often of considerable advantage in maintaining an experimental preparation in a physiologically responsive state.

Summary. Two methods are described for recording blood volume pulses from body segments and from isolated arteries. Evidence is presented to show that an arterial volume pulse reflects vasomotor change in the vascular bed which that artery supplies peripheral to the point of measurement. Peripheral vasoconstriction and vasodilation can

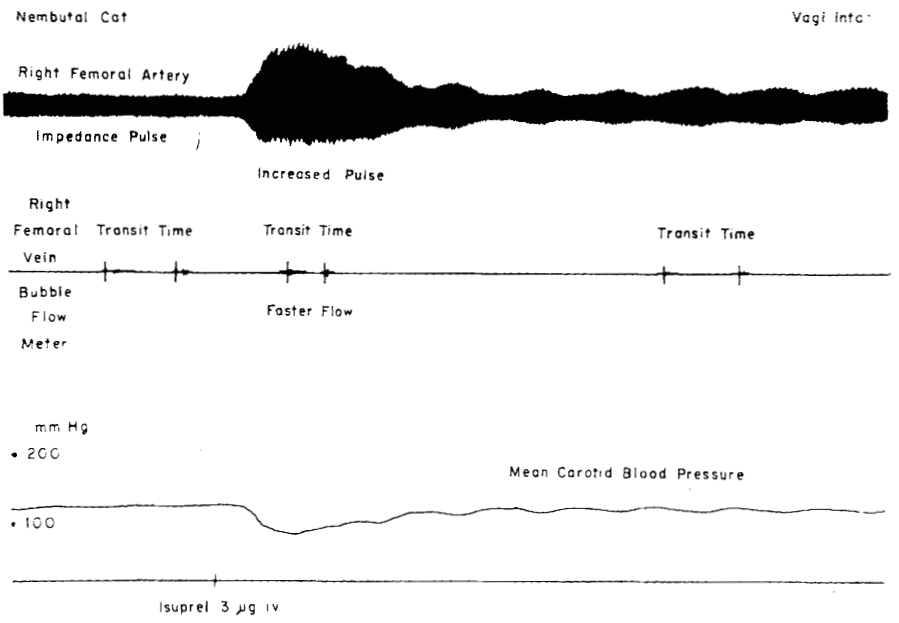

FIG. 3.
Direct Flow—Volume Pulse Comparison, Dilator Effect.

be demonstrated in this manner and indications of relative changes in blood flow can be obtained. With these methods hemodynamic responses to pharmacologic agents can be studied. The initial responses and subsequent adjustments can be followed.

Received August 21, 1950. P.S.E.B.M., 1950, v75.

Influence of Fasting and Adrenalectomy on Normal and Malignant Lymphoid Tissue Composition.* (18275)

ELIJAH ADAMS† AND ABRAHAM WHITE

From the Department of Physiological Chemistry, School of Medicine, University of California, Los Angeles, Calif.

Among considerations of interest in the study of a neoplastic tissue is its responsiveness to specific physiologic stimuli known to have characteristic influences on homologous normal tissue. The well-known involutional response of lymphoid tissue to pituitary-adrenal cortical secretions, and, through their mediation, to a variety of stresses(1), has prompted investigations of the sensitivity of lymphoid tumors to these influences. Such studies have already indicated that in laboratory animals(2,3), and in humans(4), strik-

* This investigation was aided by a grant from the Division of Research Grants and Fellowships of the National Institutes of Health, U. S. Public Health Service.

† American Cancer Society Post Doctoral Fellow. Present address: School of Medicine, University of Utah, Salt Lake City.

1. White, A., *Ann. Rev. Physiol.*, 1949, v11, 355.

2. Law, L. W., and Speirs, R., PROC. SOC. EXP. BIOL. AND MED., 1947, v66, 266.