by smell in 3 instances, although in case (1) they definitely antedated the ether end-point and corresponded more closely to the shortened arm to tongue time.

Ordinarily when ether is injected it volatilizes and escapes almost completely from the lung during the first circuit of the blood. Only a negligible amount may remain and eventually reach the systemic capillaries. However, in the presence of a venous-arterial communication, ether in variable amount depending upon the anatomic defect, is shunted into the left side of the heart from where it is propelled into the systemic circuit setting up the above described irritative phenomena in the capillaries of the skin. The rapid saccharin time is similarly explained.

Circulatory studies with saccharin and ether have been performed by us in over 2,000 patients including 2 cases with acquired septal defect due to coronary thrombosis and other congenital cardiacs with and without cyanosis. In no other case have we encountered the above sensory and circulatory disturbances. The ether method is of definite value, therefore, in establishing with certainty the existence of venous-arterial shunt.

9812 P

Xylose, Inulin, and Creatinine Clearance in the Normal Frog.

ROY P. FORSTER. (Introduced by M. F. Guyer.)

From the Laboratories of Zoology and Physiological Chemistry, University of Wisconsin.

Hendrix, Westfall, and Richards[1] have shown from microanalyses of glomerular urine obtained directly from the exposed kidney of the frog, after intravenous injection of inulin, that it contained the same concentration of inulin as the plasma. Also, Richards and his coworkers[2] demonstrated in a double perfusion experiment that the frog's kidney was unable to excrete inulin when the fluid flowing through the intertubular capillaries contained inulin and that of the glomerular capillaries did not. Creatinine was found by them to undergo insufficient concentration in the frog renal tubules to suggest secretion.

1 Hendrix, J. P., Westfall, B. B., and Richards, A. N., *J. Biol. Chem.*, 1936, **116**, 735.

2 Richards, A. N., and Walker, A. M., *Am. J. Med. Sci.*, 1935, **190**, 727.

The following experiments on normal unanesthetized bullfrogs *(Rana catesbiana)* were designed to determine the relation of xylose to inulin clearance, the relationship between plasma concentration of inulin and the amount excreted per unit time, and the relationship between creatinine and inulin clearances. A total of 16 normal bullfrogs, varying in weight from 280 to 600 gm. were used. Between and during collections they were kept individually in large battery jars almost completely immersed in tap water. Exogenous creatinine, inulin, xylose, and water were injected into the dorsal lymph sac. Urine was collected by catheter at half-hour or hour intervals and blood was withdrawn directly from the heart before and after each experiment.

In 9 individuals xylose was found to have a lower clearance than inulin, the average xylose to inulin ratio being 0.80. In 27 individual collection periods on 7 animals with the plasma level of exogenous creatinine varying from 11.7 to 260 mg. %, and with the urine flow ranging from 3.1 to 27.8 cc. per kg. per hour the creatinine clearance remained practically identical with that of

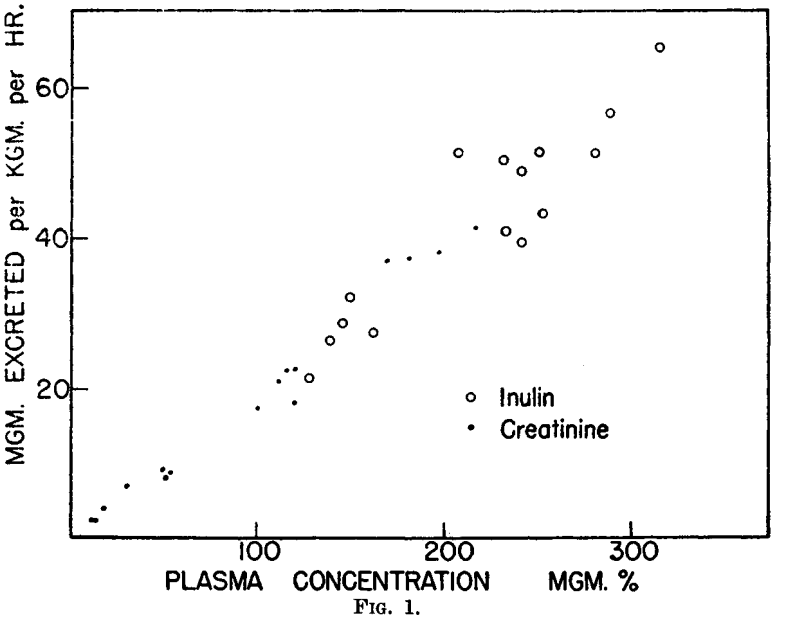

FIG. 1.

Data on 6 normal unanesthetized animals showing that the rate of inulin and creatinine excretion is proportional to the plasma level. As renal clearance in frogs varies with the urine flow these data represent all collections within the limited range of 10 to 18 cc./kg./hr. The absence of a curvilinear relationship between plasma level and amount excreted per unit time indicates that tubular secretion is not a factor in the renal elimination of these two substances. Scattering of the data is due to variations in urine flow, which in these animals is almost entirely a glomerular phenomenon.

inulin. The average creatinine/inulin clearance ratio of all collections was 0.996, with a standard deviation of 0.0643.

The clearance of both inulin and creatinine is not constant for any one animal, but varies with the urine flow. The average clearance at a urine flow of 1 cc. per kg. per hour is 3.5 cc. per kg. per hour, and this rises to an average of 20 at a urine flow of 15. When collection periods are selected within a limited range of urine flow, and plasma concentration of inulin and creatinine then plotted against the amount excreted per unit time the data describes a straight line, extrapolating to the zero axis, as illustrated in Fig. 1. This direct proportion to plasma level of inulin and creatinine, and the identical clearances of these substances under all circumstances investigated, is interpreted as indicating that they are excreted solely by glomerular filtration, and that tubular secretion is not a factor in their elimination.

9813 P

Partial Reactivation of Formolized Tobacco Mosaic Virus Protein.*

A. FRANK ROSS AND W. M. STANLEY. (Introduced by Richard E. Shope.)

From the Rockefeller Institute for Medical Research, Princeton, N. J.

A marked reactivation of tobacco mosaic virus protein that has been partially or completely inactivated by formaldehyde[1] has been obtained by dialysis at pH 3. In the experiments to be reported, inactivation was allowed to proceed at room temperature in reaction mixtures containing 2% purified virus protein and 2% formaldehyde in M/10 phosphate at pH 7. Samples were removed after different periods of time and immediately dialyzed against cold distilled water for about 6 hours in order to stop the reaction by the removal of excess formaldehyde. Preliminary experiments indicated that such inactivated virus protein could be partially reactivated by incubation with dimethyldihydroresorcinol[2] or with his-

* The details of the work are contained in a paper to be submitted to *The Journal of Biological Chemistry.*

1 Stanley, W. M., *Science*, 1936, **83**, 626.

2 Wadsworth, A., and Pangborn, M. C., *J. Biol. Chem.*, 1936, **116**, 423.