Nicotiana tabacum L. and *N. sylvestris* Spegaz. and Comes were used as host plants for this study. They were inoculated with the two viruses either simultaneously or at intervals of one and four days. Controls consisted of plants inoculated with only one virus. The macroscopic symptoms of tobacco etch usually developed from one to three days before those of tobacco mosaic. Symptoms of both viruses were easily seen in doubly inoculated plants during onset and up to two or three weeks after inoculation. Later, symptoms were predominantly those of etch, doubly infected plants being distinguished from those infected with etch alone merely by a slight reduction in size.

Slides for microscopic examination were prepared by cutting thin slices of tissue, with trichomes attached, from the veins and edges of leaves, and mounting the tissue in 0.85% NaCl. Cytological examinations were made on the 11th and 40th days after inoculation.

The size and location of leaves wherein complete expression of cytological symptoms regularly occurred were determined for both viruses by examination of control plants. Corresponding leaves of doubly infected plants were then studied to determine the distribution of the two viruses in individual cells. The first cytological examination revealed the presence of crystalline inclusions of both viruses in every observed integumentary cell of such leaves from simultaneously inoculated plants. This was not entirely true for corresponding leaves of plants inoculated first with one virus and four days later with the other. In such plants, cells of leaves near the point of inoculation showed inclusions dominantly or entirely of the primary virus only. Cells of leaves higher up the plant, however, nearly always showed inclusions of both the primary and the challenge virus.

When the plants were reexamined 40 days after inoculation, again nearly every observed cell of appropriate leaves showed crystalline inclusions of both viruses. In many cases, leaves near the tops of the plants showed only tobacco etch inclusions while those lower down the plant showed both. This observation suggests that inclusions of etch develop sooner than those of mosaic in old plants and may help explain the dominance of etch symptoms.

The hexagonal plates of tobacco mosaic are known to be closely associated with tobacco mosaic virus. Likewise, the intranuclear crystalline inclusions of tobacco etch are closely associated with tobacco etch virus. Since it is very unlikely that the materials composing these inclusions were formed in distant cells and transferred in toto to the many cells where they were found occurring together, the evidence indicates not only that the two viruses were present in the same cells, but also that they multiplied in the same cells.

Experiments reported above gave no evidence of interference between tobacco mosaic and tobacco etch viruses, such as that reported between the severe strain of tobacco etch virus and potato Y virus (*1*). The interference phenomenon is well established for bacteriophages (*2*) and for certain viruses that cause disease in man and other animals (*3, 8*). It has been demonstrated for plant viruses only in the case cited above. However, the literature on interference occasionally confuses this phenomenon with *cross protection* such as was observed by Kunkel (*5*) between tobacco and aucuba mosaic viruses. It seems worth while to point out here that cross protection, in contrast with interference, consists of the protection afforded a plant by virtue of a previous infection with a closely related virus strain. Nonrelated viruses do not ordinarily give such protection.

References

1. Bawden, F. C., and Kassanis, B. *Ann. appl. Biol.*, 1945, **32**, 52.
2. Delbruck, M., and Luria, S. E. *Arch. Biochem.*, 1942, **1**, 111.
3. Findlay, G. M., and MacCallum, F. O. *J. Path. and Bact.*, 1937, **44**, 405.
4. Kassanis, B. *Ann. appl. Biol.*, 1939, **26**, 705.
5. Kunkel, L. O. *Phytopathology*, 1934, **24**, 437.
6. McWhorter, F. P. *Stain Tech.*, 1941, **16**, 143.
7. Vanterpool, T. C. *Phytopathology*, 1926, **16**, 311.
8. Vilches, A., and Hirst, G. K. *J. Immunol.*, 1947, **57**, 125.

Catheterization of the Coronary Sinus, Right Heart, and Other Viscera With A Modified Venous Catheter

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A technique of coronary sinus catheterization (*10*) has made it possible to study coronary blood flow and myocardial metabolism in intact dogs (*6, 7, 11*) and has been recently applied to similar studies in man (*2, 3*).[2] More recently a modified venous catheter has been devised to meet several of the special problems encountered in this procedure. In extensive trials in man, with Bing and his associates (*3*), as well as in dogs, the modified catheter described in this report appeared to facilitate catheterization of not only the coronary sinus, but also other regions in the heart and visceral veins.

The venous catheter now most generally used was designed by Cournand and Ranges (*4*), some years following Forssman's original description of right auricular catheterization in man (*8*). It is 100 cm long, of the ureteral type, with a woven shellacked nylon core covered with a heavy X-ray-opaque plastic coating and with a

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[2] The application of this technique, together with the nitrous oxide blood-flow method of Kety and Schmidt (*12*), to the measurement of coronary blood flow in man, has been a joint project undertaken by Bing and his co-workers at Johns Hopkins University Medical School, in cooperation with the authors and with Eckenhoff and his associates at the University of Pennsylvania Medical School (*2*). To all of these, the authors are deeply grateful for their collaboration and interest, and to Normal C. Jeckel and Edward W. Grant, of the U. S. Catheter and Instrument Corporation, for their help and advice.

curved tip terminating in a single straight opening or "eye." (A catheter 125 cm in length is usually necessary for Dexter's technique of sampling blood from the pulmonary capillary bed *5*.)

Using this standard catheter, it has often been difficult to obtain blood samples from the coronary sinus and cardiac veins. Probably a vessel wall or valve leaflet has occluded the single terminal eye as suction has been applied with the sampling syringe, even though fluid could always be freely injected through the catheter. This difficulty was sometimes encountered in drawing blood from other regions, such as the hepatic or renal veins or the right ventricle, and was greater with smaller catheters (No. 6 to No. 8F). However, for avoiding coronary venous obstruction and possible intimal damage (*11*), these smaller catheters have been desirable. Pressure recordings have also been damped sometimes, apparently by the single terminal eye impinging upon a closely confining vessel wall or even upon the endocardial surface.

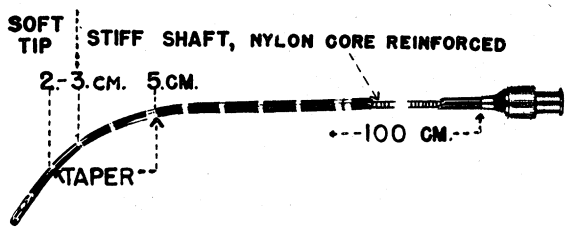

FIG. 1. Bird's-eye catheter (tapered).

The standard venous catheter of Cournand has, therefore, been modified by the addition of two small side eyes, or what we have termed "bird's-eyes," with shallow grooves in the outer surface of the catheter leading forward from the side eyes to the usual terminal eye (Fig. 1).[3] These features have prevented effective suction from being applied to a vessel wall, endocardium, or coronary-sinus valve leaflet during sampling, and no difficulties in withdrawing blood samples from any visceral vein have been encountered. Reasonable pressure recordings have been obtained which, if anything, have been less damped than those previously obtained in the right auricle and coronary venous system with the standard catheter. Any possible local trauma or cardiac arrhythmia arising from suction of intima or endocardium against the usual tip should also be minimized by the multiple eyes which break any suction against the modified catheter tip.

The "bird's-eyes" have been placed only 2 mm behind the tip, allowing accurate localization of blood samples and pressure recordings. The whole tip, including the grooves, has been given several extra coats of plastic finish to ensure a smooth atraumatic surface.

As a further modification, a tapered tip, also illustrated in Fig. 1, has proved valuable in catheterizing the hepatic and renal veins, as well as the coronary venous system of dogs and man (*9*). A No. 9F shaft with a gradual taper to a No. 7½F from a point 5 cm to 2 cm from the tip, or a No. 8F tapered to a No. 6½F tip, was found to be most advantageous. The nylon core has been doubly woven to a point 3 cm from the tip. This tapered catheter has combined the advantages of a relatively large, stiff, nonbuckling, easily manipulated, and easily visualized catheter shaft with a smaller, more flexible, less traumatic, and nonobstructing tip. This modification has facilitated controlled gentle insertion of the catheter into the coronary sinus. As an alternative, however, a No. 7 or No. 8F modified "bird's-eye" catheter without a taper has been used in man, often with a stylet inserted to within 3–4 cm of the tip to lend sufficient stiffness to the catheter shaft for controlled insertion (*3*). The stylet has then been removed as soon as the catheter had been successfully inserted into the coronary sinus.

The tapered catheter has a uniform No. 7 or No. 6F bore throughout its entire length, providing a minimal dead-space volume. Occurring at a known distance from the tip, the taper provided a landmark under fluoroscopy for direct estimation of the depth of insertion of the catheter into the coronary venous system.

Earlier workers were discouraged from using a catheter with side eyes or multiple eyes by the tendency for blood clots to form and obstruct the catheter lumen. With the present modified catheter this has been avoided by keeping the catheter thoroughly washed out with saline between samplings. A continuous saline infusion had to be maintained in addition to an injection of 5–10 cc of saline after each observation or sample. The grooves in the outer surface of the modified catheter tip prevented complete obstruction of either side eye by a vessel wall, so that the saline infusion constantly washed all 3 openings free of blood which might otherwise stagnate and clot. It was not necessary to add heparin to the continuous infusion, although several investigators add 20 mg/liter as an extra precaution against clotting. Others prefer to fill only the dead space of the catheter with the proper amount of heparin just before drawing each blood sample, so that the patient himself will receive no heparin. Traces of this heparin then appear to cling to the sides of the catheter lumen during the ensuing washout and sampling in sufficient quantity to prevent clotting until fresh saline infusion can be re-established. Tiny platelet thrombi were sometimes found in the side eyes after long procedures in man, but usually did not interfere with sampling.

In dogs, which have a more active clotting mechanism than man (*1*), an additional routine for preventing clotting in the catheter was often necessary (*11*). This might also be of value in man. The routine consisted of baking the catheter dry following the usual chemical sterilization and dipping the tip in full-strength heparin (Liquamine, Lilly) for an hour just before use. After proper drying the hydrophilic plastic coating absorbed enough heparin to act as an effective local surface anticoagulant.

Baking (at 110–120° C dry heat for 1–2 hrs) also stiffens the catheter, making it easier to manipulate ac-

[3] Available from the U. S. Catheter and Instrument Corporation, 334 Bay Street, Glens Falls, New York.

curately and probably less traumatically (*5*). Excessive heat (over 125° C) or too frequent baking shortens the life of the catheter. The only alternative to occasional baking, when the catheter finally becomes too limp to control, is the use of a stylet to lend sufficient stiffness for controlled insertion, as described above.

References

1. BANFIELD, W. G., JR., HACKEL, D. B., and GOODALE, W. T. (To be published.)
2. BING, R. J., GOODALE, W. T., ECKENHOFF, J. E., HANDELSMAN, J. C., CAMPBELL, J. A., GRISWOLD, H. E., VAN DAM, L. D., HARMEL, M. H., HAFKENSCHIEL, J. H., LUBIN, M., and KETY, S. S. *J. clin. Invest.*, 1948, **27**, 525.
3. BING, R. J., GOODALE, W. T., HANDLESMAN, J. C., and ECKENHOFF, J. E. (To be published.)
4. COURNAND, A., and RANGES, E. A. *Proc. Soc. exp. Biol. Med.*, 1941, **46**, 462.
5. DEXTER, L., HAYNES, F. W., BURWELL, C. S., EPPINGER, E. C., SEIBEL, R. E., and EVANS, J. M. *J. clin. Invest.*, 1947, **26**, 547.
6. ECKENHOFF, J. E., HAFKENSCHIEL, J. H., FOLTZ, E. L., and DRIVER, R. L. *Amer. J. Physiol.*, 1948, **152**, 545.
7. ECKENHOFF, J. E., HAFKENSCHIEL, J. H., HARMEL, M. H., GOODALE, W. T., LUBIN, M., BING, R. J., and KETY, S. S. *Amer. J. Physiol.*, 1948, **152**, 356
8. FORSSMAN, W. *Klin. Wschr.*, 1929, **8**, 2085.
9. GOODALE, W. T., Unpublished data.
10. GOODALE, W. T., LUBIN, M., and BANFIELD, W. G., JR. *Amer. J. med. Sci*, 1947, **214**, 695.
11. GOODALE, W. T., LUBIN, M., ECKENHOFF, J. E., HAFKENSCHIEL, J. H., and BANFIELD, W. G., JR. *Amer. J. Physiol.*, 1948, **152**, 340.
12. KETY, S. S., and SCHMIDT, C. F. *Amer. J. Physiol.*, 1945, **143**, 53.

A Modified Photoelectric Apparatus for Permeability Studies[1]

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For a number of years the author has been using a photoelectric technique for measuring the permeability of erythrocytes similar to that described by Parpart (*3*). Most of the experiments have involved hemolysis studies of chicken erythrocytes. Since the rate of hemolysis may be influenced by a number of different factors (e.g., Jacobs, *2*) it was decided to use swelling measurements only as an indication of permeability changes. The author has experienced much more difficulty in obtaining measurable light changes when chicken erythrocytes swell, however, than when mammalian erythrocytes are used.

It was thought that perhaps greater sensitivity could be obtained using light of a wave length corresponding to the region of the spectrum which is maximally absorbed by hemoglobin. Consequently, the light was passed through a green filter[2] before going through the erythrocyte suspension. This, however, decreased the light transmission to such an extent that the current produced by the photronic cell was too small to give a measurable deflection of the galvanometer. A photoelectric call and amplifier were substituted for the photronic cell with satisfactory results. A photograph of the photocell-amplifier unit[3] is shown in Fig. 1 and a diagram of the circuit, in Fig. 2.

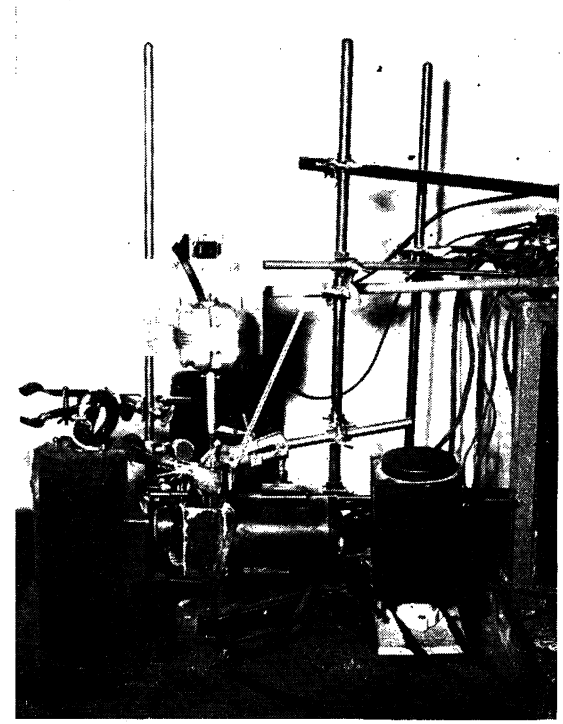

FIG. 1

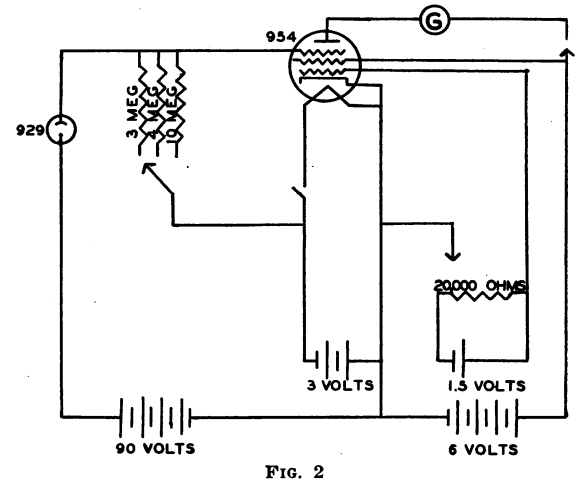

FIG. 2

[1] This work was made possible by grants from the U. S. Public Health Service and from the Faculty Research Fund of the University of Oklahoma.

[2] Corning glass filter #4010, which has maximum transmission at 525 mμ.

[3] The author is indebted to Hans Weltin, Department of Physics, University of California, Santa Barbara, for the design and construction of this unit.