Leonardo da Vinci, living in the 15th century, was, so far as we are able to determine, the first scientist to use injection methods to demonstrate the blood vessels. Garrison (1) illustrates a drawing from Leonardo's Quaderni in which the ramifications of the femoral artery give one the impression of an injection. Since this pioneer work anatomists have, from time to time, used this method for the demonstration of not only the larger blood vessels but also to demonstrate the capillary circulation. Among the more recent work should be mentioned that of, Huber (2) on the circulation of the kidney, Gross and Kugel (3) on the circulation of the heart valves, and that of Hinman and his coworkers (4) on the changes in the circulation in the hydronephrotic kidney. After all this most excellent work, we hesitate to add another paper on the dry details of technique, but because of the cordial reception which has been given to demonstrations which have been made at various scientific meetings of specimens prepared by celloidin injection of the human kidney, we feel that the technique may be of use to other workers in the same or allied fields of biology. The method as given here has been developed so as to give consistent results on the human kidney, but by slight modification of the strength of celloidin and the pressure will give equally good results on the kidney or other organs from the lower animals.

Method: The kidney is removed from the body as soon after death as possible, but good results may be secured as long as 30 hours after death. Particular care should be taken in sectioning the renal artery. It should by preference, be cut with a piece of aorta attached, so that a large canula may be inserted and also that it may be secured at a point at least one-half inch proximal to the point of primary division. (It has been our experience that a considerable number of human kidneys show an anomalous vessel arising from the main stem renal artery just distal to its origin from the aorta and entering the lower pole of the kidney directly). Further, one should be careful to remove

*Read before the Medical Science Section, Ohio Academy of Science, April 9, 1926.
the kidney with the perirenal tissue attached since the capsular branches of the renal artery in the human kidney are of considerable size and will give trouble in ligation later if a goodly portion of the tissue about the kidney is not included. As soon as possible after its removal from the body the kidney should be taken to the laboratory for the actual injection. A canula is tied into the renal artery and connected to the city water supply, in this manner allowing water to flow through the kidney from the water lines until 125 to 150 gallons of water are forced through the capillary bed of the organ. This in our experience is most satisfactorily accomplished under a head of 600 mm. of Hg. flowing for

24 hours. (Special clot dissolvers and vaso-dilators as used by some authors have not shown any advantage over this method in our hands). After this thorough washing the kidney is placed in a vessel and covered with water and the canula connected to the injection apparatus. A current of air is passed through the arterial tree and as the kidney is submerged in water, all leaks are readily detected and ligated. This is important since to maintain the required pressure of injection later and to avoid the loss of considerable quantities of celloidin, the arterial tree must be a closed system. When one is satisfied that all leaks are secured, a bottle of thin celloidin is placed in the circuit and a clamp placed on the rubber tube leading to the canula. By air pressure the pressure in the bottle of celloidin is raised to 600 mm. of Hg., at which time the clamp is suddenly released and the full pressure is allowed to enter the kidney vessels at once. This is important since a gradual raising of the pressure will not give a perfect injection. If the celloidin is of the
correct strength, it will pass into the glomeruli but will not pass beyond because of its viscosity. The thin celloidin injection is maintained for two to six hours when it is replaced by the thick celloidin which in injected under a constant head of 600 mm. of Hg. for twenty-four hours. During the entire procedure the organ is kept immersed is water. For a source of pressure we use tanks of compressed carbon dioxide to which a reducing valve is attached. In this manner a constant pressure may be maintained for hours. After twenty-four hours the canula is disconnected and the capsule and perirenal tissue stripped from the kidney. The kidney is then placed in a 75 per cent solution of hydrochloric acid which in 24 to 48 hours will so completely digest the soft parts that a gentle stream of water will remove them, leaving a celloidin cast of the blood vessels.

The celloidin solutions used in the injections are:

**Thin Celloidin.**
- Paraloidin: 2.75 gms.
- Camphor: 2.0 gms.
- Acetone: 100.0 cc.

**Thick Celloidin.**
- Paraloidin: 10.0 gms.
- Camphor: 8.0 gms.
- Acetone: 100.0 cc.

The celloidin may be colored by various fat soluble dyes. We have used Scharlet R for red and Methyl green for blue. If it is desired to inject the veins a similar technique is used except that 200 mm.
of Hg. is used instead of 600 mm. For the pelvis we use 20 per cent celloidin and 15 per cent camphor in acetone and inject under 60 to 75 mm. of Hg.

The accompanying figures illustrate both the completeness and the detail of injection that may be secured by this method.

BIBLIOGRAPHY.