Radioactive Microsphere Distribution and Single Glomerular Blood Flow in the Normal Rabbit Kidney

Lise Bankir*, Nicolette Farman, Jean-Pierre Grünfeld, Edith Huet de la Tour and Jean-Louis Funck-Brentano

I.N.S.E.R.M. — Unité 90
Hôpital Necker, Paris, France

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Summary. Intracortical distribution of blood flow was studied in the rabbit kidney with 15 μm labelled microspheres (M) injected into the left ventricle. M injection did not alter renal function. Thanks to arterial filling of left kidney with silicone rubber, efferent vascular patterns of the glomeruli could be precisely identified. Glomeruli of different populations were sampled by microdissection and their radioactivity measured. Assuming that intracortical distribution of M reflected distribution of flow to the glomeruli, individual glomerular blood flows (GBF) were determined. In hydropenic rabbits, GBF was higher in deep glomeruli providing vasa recta (G4) (193 ± 14 nl·min⁻¹) than in most other superficial (G1 and G2) and deep glomeruli (G3) (190 ± 27, 113 ± 10 and 127 ± 9 nl·min⁻¹ respectively; $\bar{m} ± SEM, n = 6$). The exact significance of GBF found in superficial glomeruli with straight ascending efferent arterioles supplying agglomerular suscapsular cortex (G1) was questioned because of the possible axial streaming of spheres. Afferent medullary blood flow was calculated to represent 9.0 ± 0.9% of total renal blood flow.

Key words: Intrarenal Blood Flow Distribution — Glomerular Blood Flow — Nephron Heterogeneity — Radioactive Microspheres — Renal Cortex.

Morphologic heterogeneity of nephrons is well known [1, 6]. Heterogeneity of single nephron glomerular filtration rate (SNGFR) has been demonstrated in several works recently reviewed by Wright and Giebisch [24]. Individual glomerular blood flow (GBF) has also been estimated recently but heretofore no attempt was made to characterize the glomeruli according to their vascular pattern [8, 9, 21, 22] except for the most superficial glomeruli, the efferent arterioles of which are accessible for micropuncture [2, 12].

We propose a method for determining individual glomerular blood flow, based on the 15 μm microsphere technique combined with microdissection after arterial Microfil injection and maceration of the kidney.

* Attaché de Recherches I.N.S.E.R.M.
By this method, individual glomerular blood flow was determined in 4 populations of glomeruli distinguishable by their vascular pattern and their position in the cortex.

In preliminary experiments, we assessed that the microsphere distribution in the rabbit was similar to that found in dogs and rats [9, 11, 17, 19, 21] and identical in both kidneys of the same animal. The microsphere content of cortical slices was studied after injection of a small quantity of labelled spheres. Then, in another group of animals, we determined glomerular distribution of microspheres and individual glomerular blood flow. The number of spheres used in the latter experiments was high in order to allow accurate measurements. We have verified that such an amount of spheres did not impair renal function.

**Material and Methods**

"Fauve de Bourgogne" rabbits, males and females, weighing 1.8 to 3.0 kg, fed a normal pellet diet (Extralabo) and given free access to water, were used. They were anesthetized with Nembutal (Abbot, 30 mg/kg body weight intravenously) and tracheotomized. Arterial blood pressure (BP) was monitored through the right femoral artery with a strain gauge transducer (Telco). Left ventricle was catheterized with a Pe 50 catheter inserted through the left carotid artery. Microspheres (M) 15 ± 5 μm diameter, labelled with 85 Sr or 169 Yb (3M Company, St. Paul, Minn.) were suspended in 10% Dextran solution or in 60% sucrose solution (to avoid sedimentation) with 1% Tween 80. Ultrasonic dispersion of M was performed before each experiment with an ultrasound probe (M.S.E.) immersed into the suspension. Less than 2% of the spheres remained aggregated after a 3 min treatment; further ultrasonication did not improve dispersion. A measured amount of M suspension was injected into the left ventricle and the catheter content was flushed with 0.5 ml of isotonic saline. Position of the ventricular catheter was checked at the end of each experiment. Results presented were obtained in animals whose BP remained steady during injection of the tracer.

It is assumed that IV suspension is well mixed at the site of injection and that it behaves like whole blood in the arterial vascular tree. M are trapped in capillaries because of their diameter and the quantity of M found in an organ or a piece of tissue is proportional to its blood flow. In the kidney, they are trapped in glomerular capillaries and reflect afferent glomerular blood flow.

1. **Group A.** In 4 hydropenic rabbits, 0.3 × 10^6 M (0.7 mg, 5 to 10 μCi) suspended in 0.5 ml were injected in 20 to 30 sec. Animals were then sacrificed with a lethal dose of intravenous Nembutal. The kidneys were removed, decapsulated, weighed and frozen in a mixture of dry ice and acetone. From the frozen kidney, 2 or 3 small rectangular pieces, 2 to 3 mm thick and wide, including cortex and outer medulla, were taken. Each sample was subsequently sliced with a frozen section-microtome (I.E.C.) in 12 successive regular slices, 500 μm thick and parallel to the surface of the kidney (C1 to C12). Each slice was introduced in a pre-weighed counting tube containing 1 ml water. Tubes were weighed again and correction was made for evaporation. The remaining pieces of each kidney were weighed and placed in several counting tubes. Radioactivity of slices and remaining kidney was counted in a well-type scintillation detector (Mecaserto Mo 13). Results were expressed for each slice as q/Q where q is the radioactivity per gram of tissue slice (mean from the 2 or 3 samples) and Q the radioactivity per gram of total kidney.