ULTRASONIC MEASUREMENT OF THE HEPATIC PORTAL BLOOD FLOW IN RATS BY CONTACT BIOMICROSCOPY

N. Ya. Kovalenko and D. D. Matsievskii

UDC 616.36-005+616.149-008.1]-073.432.19

KEY WORDS: ultrasonic transducer; hepatic portal blood flow; biomicroscopy; microcirculation.

In the last decade three types of biomicroscopy have been developed and used for the experimental study of the microcirculatory system of the internal organs of mammals: transillumination, luminescence biomicroscopy, and biomicroscopy in reflected light. These methods, based on different optical principles, can be used for the intravital study of microvessels under normal and pathological conditions over a period of time. However, methods of intravital study of the microcirculation, even if the diameter of the microvessels and the linear and volume velocities of the blood flow in them can be recorded automatically, reflect merely local hemodynamic situations which may arise at the level of the microcirculatory units of an organ, i.e., at the final level of integration of the cardiovascular system. Meanwhile, in order to understand the complex hemodynamic relations which exist in the whole body between the different levels of integration of the circulatory system, the study of the inflow of blood into an organ and its outflow from it, accompanied by simultaneous biomicroscopic study of terminal microcirculatory structures, is highly informative.

In practice, the conduct of a complex biomicroscopic investigation of the microcirculatory system accompanied by adequate quantitative evaluation of the hemodynamics of an organ encounters a number of technical difficulties, arising from the extremely small linear dimensions of visceral blood vessels in animals usually used for the study of the microcirculation (rats, mice, guinea pigs). Instrumental measurement of the linear and volume velocity of the blood flow in these vessels with existing standard measuring techniques, while preserving artificial homeostasis of the test organ and of the animal as a whole at the necessary physiological level, can be very difficult. Recent advances in ultrasonic measuring tech-

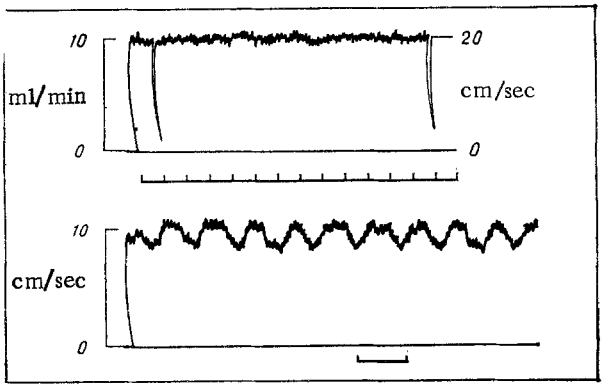

Fig. 1

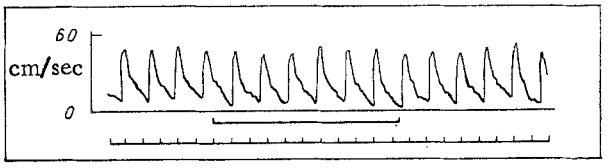

Fig. 2

Fig. 1. Types of blood flow in portal vein of rat liver. Above — uniform flow; below — wave-like flow. Time marker 10 sec.

Fig. 2. Curve of blood flow in hepatic artery of rat. Time marker 1 sec.

Laboratory of Pathophysiology of Extremal States and Bioengineering Laboratory, Institute of General Pathology and Pathological Physiology, Academy of Medical Sciences of the USSR, Moscow. (Presented by Academician of the Academy of Medical Sciences of the USSR P. D. Gorizontov.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 93, No. 2, pp. 66-68, February, 1982. Original article submitted September 24, 1981.

0007-4888/82/9302-0219$07.50

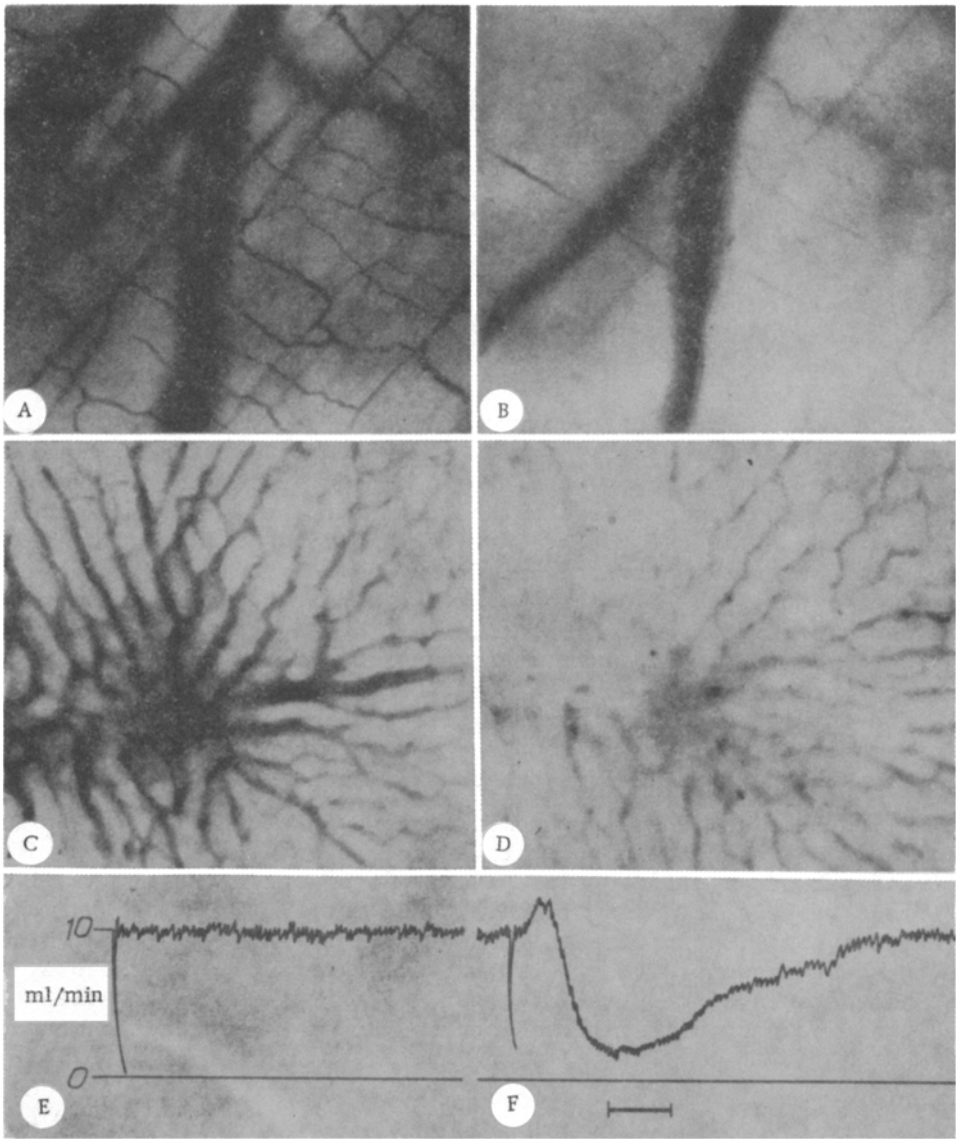

Fig. 3. Character of changes in intestinal and hepatic microcirculation and in portal blood flow of rats under the influence of acetylcholine. On left — before injection, on right — after injection of acetylcholine 0.1 mg/kg. A-D) Photomicrograph of surface microvessels in wall of small intestine and liver. Contact luminescence biomicroscopy, 30 ×; E, F) magnitude and character of blood flow in portal vein of liver. Time marker 10 sec.

niques, in the direction of miniaturization of transducers [3], have improved the prospects in this field.

The suggested method of quantitative determination of the velocity of the hepatic portal blood flow, accompanied by simultaneous contact biomicroscopy of the abdominal organs, is based on the method of radiotelemetric measurement of the blood flow by ultrasound suggested previously by Matsievskii [2]. Further development of this method for the study of the coronary blood flow led to the perfecting of miniature ultrasonic transducers, by means of which the linear and volume velocity of the blood flow can be determined with a sufficient degree of accuracy in macrovessels about 1 mm in diameter [4].

The ultrasonic technique now used for quantitative determination of the portal blood flow has the following advantages over other measuring instruments:

1. Because of the minimal dimensions of the transducer, with a total weight of 1 g and

a binder 3 mm in length, the velocity of the blood flow can be measured in vessels with a diameter of 1-2 mm;

2. the transducer is equipped with a thin, flexible, unscreened connecting lead 0.4 mm in diameter which can be placed on thin-walled macrovessels without deforming them. This is a particularly important fact for vessels such as the portal vein of the liver, which has a high level of myogenic contractile activity and can be excited by rough external handling;

3. the absence of reaction of the vessel wall to ultrasound of the low intensity used to measure the blood flow;

4. the high sensitivity of measurement on vessels of small caliber because of the absence of drift of the zero line, making mechanical or hydraulic occlusion of the vessel unnecessary.

The ultrasonic transducer used consists of two miniature split half-cylinders, in the body of which are placed two piezoelectric transducers about 1 mm in diameter, made from TsTS piezoelectric ceramics. During measurement one of the transducers emits high-frequency ultrasonic waves which are focused on the flow of moving blood. Ultrasonic waves reflected from the blood cells are received by a pressure-sensitive element and amplified by an ultrasonic instrument. The frequency of the ultrasonic waves reflected by the cells in the moving flow of blood differs from the frequency of those applied by an amount proportional to the linear velocity of the blood flow (the Doppler effect). This difference is recorded by the ultrasonic instrument and displayed as a blood flow curve. The high sensitivity of the piezoelectric transducers used, which operate at a frequency of 8 MHz, and also the use of modern highly sensitive amplifiers with a low intrinsic noise level allow very weak signals reflected by the blood cells to be recorded with sufficiently high quality and the linear velocity of the blood flow to be estimated quantitatively in cm/sec in blood vessels about 1-2 mm in diameter. In the course of measurement the ultrasonic transducer lies firmly against the blood vessel so that the areas of cross section of the vessel and transducer are equivalent. Accordingly the transducer can be calibrated in units of volume velocity of blood flow (in ml/min). The transducers were calibrated on blood vessels under acute experimental conditions and also in special model experiments in which flows with a known volume velocity were created.

Quantitative measurements of the portal blood flow were made on rats weighing 200-300 g and accompanied by a simultaneous study of the microcirculation in the liver and intestine by contact luminescence biomicroscopy, as described previously [5]. In the anesthetized animal laparotomy was performed with a circular incision. The portal vein of the liver was carefully isolated from the neurovascular bundle and taken up on a ligature. An ultrasonic transducer of appropriate diameter was applied to the separated part of the vein. All manipulations required to perform contact luminescence biomicroscopy were then carried out. To measure the systemic arterial pressure (BP) a cannula was inserted into the carotid artery. During the investigation, signals of the blood flow were recorded on magnetic tape and on an N-338-6 instrument with transmission band up to 100 Hz. A mercury manometer was used to record the systemic BP.

Biomicroscopy with simultaneous measurement of the portal blood flow was started not earlier than 30-40 min after application of the transducer. The linear velocity of the hepatic portal blood flow recorded in these experiments varied under normal conditions from 14 to 26 cm/sec. The volume velocity of the blood flow varied correspondingly from 7 to 13 ml/min. Two types of portal blood flow were found to exist in rats under normal conditions: uniform and wave-like. In the latter case the number of waves was usually from 5 to 10 per second. The amplitude of the fluctuations of the portal blood flow did not exceed 30% of the amplitude of its constant component (Fig. 1). In some animals, additional pulsed waves connected with the phases of respiration were recorded superposed on the uniform or wave-like portal blood flow.

In some experiments, in which transducers were applied concurrently to the portal vein and hepatic artery, portal and arterial fractions of the hepatic blood flow were recorded separately. Isolation of the arterial flow curve from the general signal was based on the frequency principle used previously by Matsievskii when using ultrasound to visualize the blood stream [4]. The shape of the blood flow curve in the hepatic artery was of a well-marked pulsating character with the typical steep rise at the beginning of systole, followed by a slower fall in diastole (Fig. 2).

Changes in the diameter and velocity of the blood flow in the terminal microvessels of the intestine and liver observed visually in these experiments correlated with the portal blood flow recorded instrumentally. This correlation was particularly clear after direct injection of physiologically vasoactive substances into the main blood vessels of the organs. The writers showed previously that acetylcholine, injected into the aorta at the level of origin of the superior mesenteric artery of a rat in a dose of 1.0-0.1 mg/kg causes transient generalized constriction of the microvessels of the intestine and stomach [1]. A fall in blood volume in the sinusoidal system of the liver was observed under these circumstances. Measurement of the linear and volume velocities of the portal blood flow after such an injection of acetylcholine in the above doses into the aorta showed a transient fall (Fig. 3), accompanied by a fall in the systemic BP.

The suggested method of combined evaluation of the portal circulation can be used to study relations between different levels of integration of the circulatory system in other regions of the body.

LITERATURE CITED

1. N. Ya. Kovalenko and A. M. Chernukh, Vestn. Akad. Med. Nauk SSSR, No. 11, 29 (1980).
2. D. D. Matsievskii, Byull. Éksp. Biol. Med., No. 9, 119 (1970).
3. D. D. Matsievskii, Kardiologiya, No. 1, 134 (1977).
4. D. D. Matsievskii, Kardiologiya, No. 4, 135 (1978).
5. A. M. Chernukh and N. Ya. Kovalenko, Byull. Éksp. Biol. Med., No. 9, 119 (1970).

MEASUREMENT OF THE GENERAL AND LOCAL BLOOD FLOW IN A TRANSPLANTED KIDNEY BY THE HYDROGEN CLEARANCE METHOD

I. T. Demchenko and A. B. Kabakov

UDC 616.61-089.843-07:616.61-005-073.7

KEY WORDS: transplantation of the kidney; renal blood flow; hydrogen clearance method.

Quantitative determination of the general and local blood supply to the kidney after transplantation has hitherto been a very difficult problem. One of the most suitable methods for this purpose is the hydrogen clearance method, which requires introduction of platinum electrodes into the kidney or into the lumen of the renal vein [3, 5]. Unfortunately such a procedure, which involves destruction of kidney tissue and opening of the blood vessel, imposes substantial limitation of the use of the hydrogen clearance method, especially under chronic experimental conditions.

One of us (I.T.D.) recently showed that hydrogen diffuses freely through the wall of the renal vein, so that the total renal blood flow can be determined without opening the vessel [2]. Hydrogen has also been shown to diffuse rapidly through the fibrous capsule covering the kidney, which means that the blood flow in the cortical layer can be measured by a flat electrode without damaging the tissues. On the basis of these investigations a combined method of quantitative determination of the general and local blood flow in the transplanted kidney was developed and has been used at the First Leningrad Medical Institute in chronic experiments on dogs.

The object of the present investigation was to examine this method and, in particular, to describe the construction of the electrodes, the technology for recording hydrogen clearance, and methods of calculating the blood flow.

Laboratory of the Cerebral Circulation, I. N. Sechenov Institute of Evolutionary Physiology and Biochemistry, Academy of Sciences of the USSR. Department of General Surgery, I. P. Pavlov First Medical Institute, Leningrad. (Presented by Academician of the Academy of Medical Sciences of the USSR S. V. Anichkov.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 93, No. 2, pp. 68-70, February, 1982. Original article submitted June 2, 1981.

 0007-4888/82/9302-0222$07.50