

## Blood flow rate in jejunal ischemia-reperfusion injury

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**Abstract.** We examined changes in the blood flow rate in the course of jejunal ischemic injury using a laser Doppler flowmeter. For this purpose, we designed a sensor holding system, which involved the fixation of the sensor to the jejunum and allowed their movements to be synchronized. When the jejunum was reperused after 10 min of ischemia, the blood flow rate markedly increased and then gradually decreased to the initial level. A 30-min period of ischemia produced the slowest recovery of the blood flow rate. This system could be useful for monitoring the blood flow rate in different anatomical regions.

**Key words.** Blood flow rate; laser Doppler flowmeter; jejunal ischemia-reperfusion injury; anesthetized rat.

Tissue injury caused by ischemia-reperfusion is an important pathophysiological process which can be responsible for severe organ damage in humans<sup>1-3</sup>. There is evidence that an imbalance in oxidant-antioxidant activity is involved in many pathological processes mediated by free radicals, which include ischemia-reperfusion injury in various organs like liver<sup>4</sup>, heart<sup>5</sup>, intestine<sup>6</sup>, and brain<sup>7</sup>. Although ischemia is characterized in part by low tissue oxygen tensions, the evidence from studies on the central nervous system<sup>8</sup> or the intestine<sup>6</sup> suggests that an imbalance in oxidant-antioxidant activity contributes to the ischemia-reperfusion injury in various organs<sup>4,5,7</sup>. Therefore, the estimation of blood flow rate is very important in studying the pathogenesis of disorders in several organs, including the intestine. The present study attempted to monitor the blood flow rate in the ischemic-reperused rat jejunum using a laser Doppler flowmeter.

### Methods

**Animals.** Wistar rats weighing 300–400 g were anesthetized with chloral hydrate administered intraperitoneally (400 mg/kg). The level of anesthesia was then maintained by continuous infusion of chloral hydrate (20 mg kg<sup>-1</sup> h<sup>-1</sup>) through a cannulated femoral vein. This study was approved by the Ethical Committee for Animal Experiments, Oita Medical University.

**Preparation of ischemic rats.** After shaving the abdominal fur, a 2–3 cm midline incision was performed to expose the abdominal viscera. A portion of the jejunum was cut and closed on both sides, and this portion of the intestine was then laid on the abdominal wall. The blood flow rate was routinely recorded for 10 min before starting any manoeuvre. Thereafter, the intestine

was subjected to ischemia for 10 min, 20 min and 30 min by the occlusion of mesenteric arteries, and the blood flow monitored.

**Sensor holding system.** The blood flow rate was continuously monitored and recorded using a laser Doppler flowmeter (PeriFlux 4001-2, Perimed, Sweden) and an acquisition system (HIOKI 8811, memory Hi corder, Japan). To monitor the blood flow rate in the jejunum constantly, we designed the sensor holding system. This involves the fixing of the jejunal serosa with the probe (PF 413; Perimed, Sweden). To keep a constantly uniform contact between sensor and jejunal serosa, and to synchronize their movements, the sensor holding system was attached to a lever system whose load could be adjusted to one gram (fig. 1). The results are expressed as means of three tests.

### Results

The constant blood flow rate in the jejunal serosa, expressed in Perfusion Units (PU) was obtained by holding the laser probe in place, using the sensor holding system. The blood flow rate was maintained at  $500 \pm 100$  PU. The intestine was subjected to regional ischemia by occlusion of the mesenteric arteries, followed by reperfusion for a total of 80 min. Ischemia changed the blood flow rate in the jejunal serosa. Immediately after occlusion, a marked decline in the blood flow rate was observed. When the jejunum was reperused after 10 min of ischemia, the flow value was markedly elevated, and then gradually decreased to the initial level. Trials were then performed with mesenteric artery occlusion, lasting for 20 and 30 min. The flow rate after a 20 min ischemia period did not rise, but returned to the initial level immediately, while after 30 minutes of ischemia recovery was much slower (fig. 2).

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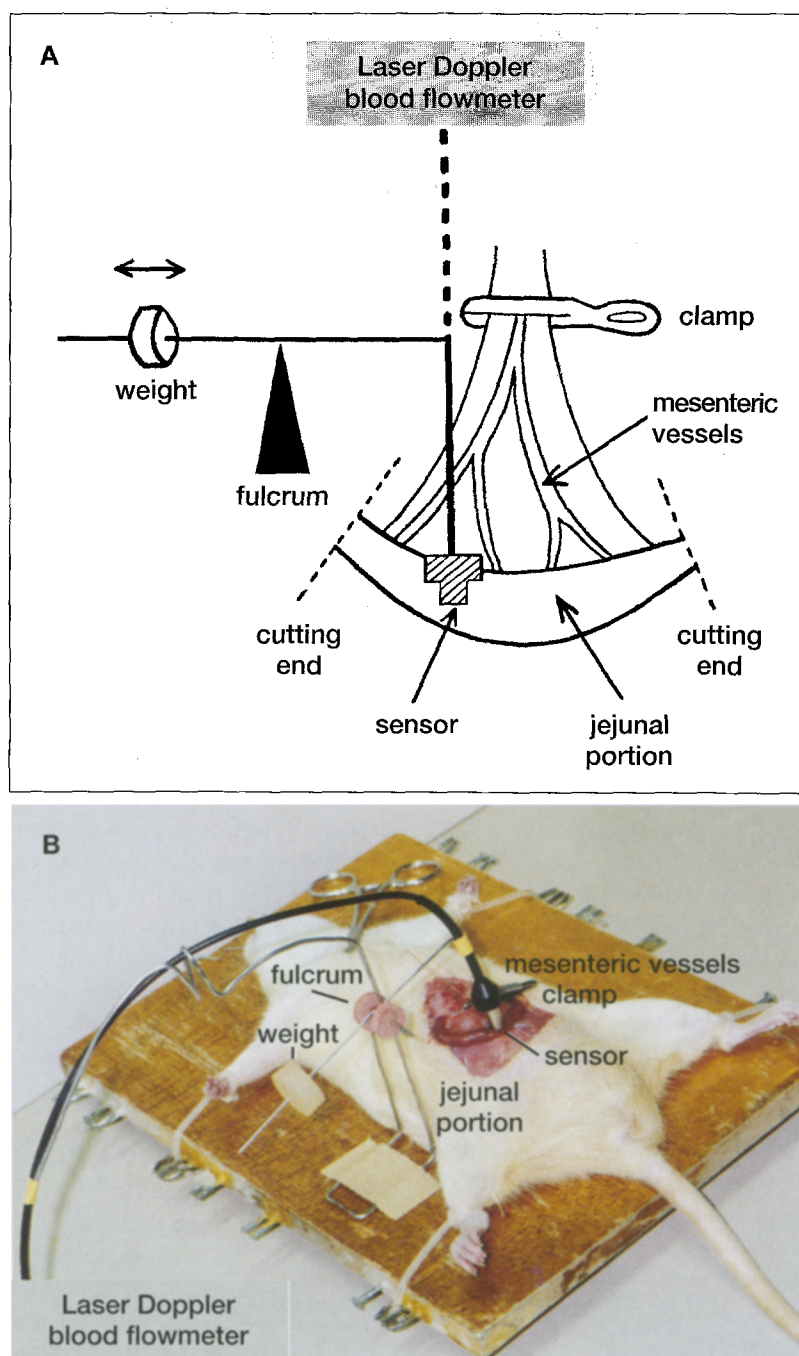


Figure 1. Sensor of laser Doppler flowmeter holding system in jejunal serosa. After the abdominal cavity had been opened through a midline incision, the jejunum was cut. The sensor holding system includes loose fixation of the system line and synchronization of the movements of the jejunum and the sensor, which was held by a fulcrum.

A total of three rats was investigated in each test, one rat being examined at each of the indicated times. The test was repeated three times on different days. The results showed no difference in the pattern of blood flow rate in the three rats examined after 10, 20 or 30 min of ischemia on different days.

## Discussion

Hemorrhagic disorders and ischemic states are the two major classes of circulatory disease<sup>9</sup>. We previously reported some work on the use of a microdialysis probe holding system<sup>5</sup>. We attempted this time to investigate

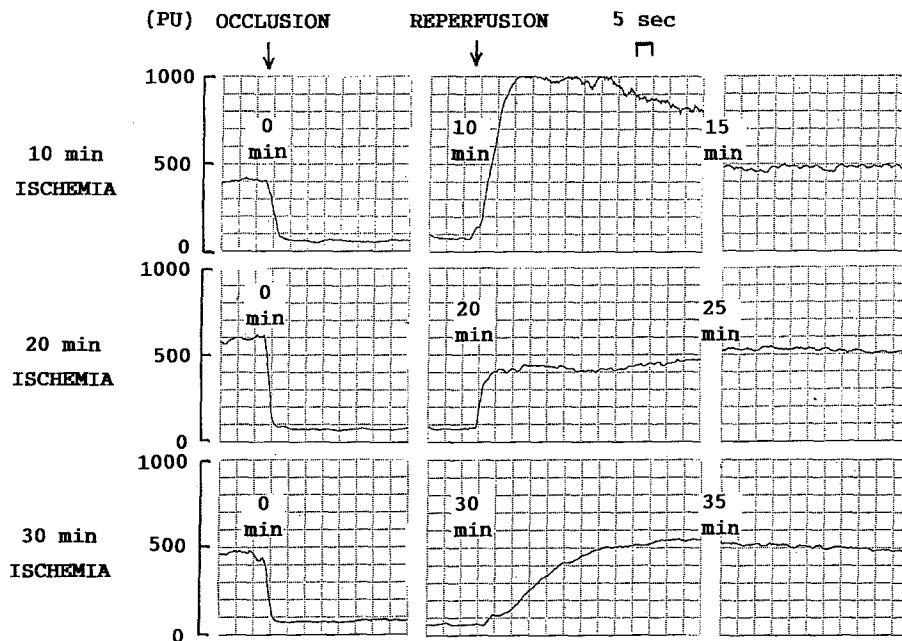


Figure 2. Changes in the blood flow rate in the course of ischemia-reperfusion. The intestine was subjected to jejunal ischemia for 10, 20 and 30 min by the occlusion of the rat mesenteric arteries. Immediately after occlusion, the blood flow rate markedly decreased. When the jejunum was reperfused, the flow rate after 10 minutes' ischemia increased remarkably, and then gradually decreased to the initial level. After 20 minutes' ischemia the flow rate immediately returned to the initial level, and it showed the slowest recovery after 30 minutes' ischemia. Signals are low-pass filtered: cut-off frequency = 25 kHz. No artifact filter was used.

the blood flow rate in the jejunum in the course of ischemic injury, using a holding system with a laser Doppler blood flowmeter. Because this type of flowmeter records microvascular blood flow from areas usually less than  $1 \text{ cm}^3$ , depending on the type of instrument used, the movements of the organ are the most common cause of artifacts. Even a few microns' change in probe-tissue distance produces unpredictable modifications in the output signal. To overcome this problem, and to keep a constantly uniform contact between probe and tissue, we designed a sensor holding system. It consists of a loose fixing of the system line, and a simple lever mechanism which allows the synchronization of the movements of the intestine and the sensor. A weight of 1 gram was needed to provide a constantly uniform contact between probe and jejunal serosa. This system allowed a constant blood flow to be measured in the undamaged jejunal serosa.

When the jejunum was reperfused, the flow rate after 10 minutes' ischemia was markedly elevated and then gradually decreased to the initial level. After a 30-minute ischaemic period the flow rate did not immediately reach the initial level. Ischemia-reperfusion injury caused morphological changes in the intestine (data not shown). In studies performed after 10 minutes' is-

chemia, light microscopic examination revealed conjugation of the blood capillaries and veins in the mucosal layer. However, after 30 minutes of ischemia the destruction of villi and epithelium was observed. These results indicate that the destruction of blood capillaries may change the blood flow pattern.

In conclusion, this sensor holding system, used for measuring the blood flow rate of jejunum in the course of ischemic injury, may also be useful for measuring the blood flow in other anatomical areas.

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