The mechanism of iron absorption was studied using ligated intestinal loops of rats. Low molecular iron chelate, stable at neutral pH, was used to prevent factors such as the physicochemical properties of iron salts from influencing intestinal absorption of iron. Mucosal uptake of radioiron ceased within 1 hour of the iron injection into jejunum, while net transport of iron from lumen to carcass proceeded in proportion to time. Total iron absorption by duodenum, jejunum, ileum, and rectum measured 8.2, 6.0, 5.0, and 5.1, while net transport of iron to carcass was 6.0, 1.2, 0.7, and 2.3 %/cm of intestinal length in 2 hours. Net transport of iron to carcass via duodenum was higher than via other intestinal segments. The amount of mucosal uptake and the net transport of iron from lumen to carcass increased proportionally depending on the amount of iron injected into duodenum and jejunum in the range of 20 to 1000 µg. Cycloheximide significantly inhibited the mucosal uptake of iron, but had no effect on the net transport of iron from lumen to carcass by duodenum and jejunum.
The present paper describes the experimental results of the mechanism of iron transport by rat intestine using a low molecular iron chelate (p-15), which is stable at neutral pH in the solution, as the iron intake source.

**MATERIALS AND METHODS**

Male albino rats of Donryu strain, weighing 80 to 120 g, were used for the experiments. Young rats weighing 50 to 60 g (purchased from Nippon Rats Co) were fed an iron-deficient diet for 2 to 3 weeks prior to the experiment to ensure constant conditions for the present iron metabolism studies (15).

The following methods were employed: a) determination of iron absorption, including operative technique for making ligated intestinal loops in rats fasted for 24 hours (16). b) preparation of 59Fe-labeled p-15 solution, and c) radiation counting by well-type γ-ray scintillation counter (Riken Co). 59Fe-p-15 (1 μCi/mg Fe) was dissolved in physiologic saline adjusted to pH 7.5 by 0.1 M Tris-HCl buffer, and 0.5 ml of this solution, containing 100 μg Fe, was injected into the ligated intestinal loop of rats (14). The rats were then kept for 2 hours after stitching the abdominal wall. When the duodenal loop was used for the experiment, bile duct was ligated to prevent bile secretion into duodenum.

At the end of the experiment the rats, under ether anesthesia, were slaughtered and 2 ml of blood was withdrawn by heart puncture. The intestinal loop was removed immediately from the carcass, and the residual radioactivity in the whole loop was counted to determine the net transport of iron from lumen to carcass. The segment was then opened, washed three times with physiologic saline to remove the unabsorbed iron, and radioactive activity taken up by the intestine was counted. “Total iron absorption” was determined as the sum of “mucosal uptake” and “net transport of radioiron from lumen to carcass.” Total amount of 59Fe appearing in blood was calculated on the assumption that blood volume is 6% of body weight (17).

59Fe-p-15 was prepared from 59Fe-labeled tetrasodium monoferrous dicitrate, dissolved in warm water and adjusted to pH 7.5 by sodium hydroxide solution (14). About 60-80% of Fe2+ was rapidly oxidized to Fe3+, but it was observed to form a stable low molecular chelate at a molecular weight of about 1500, as determined by gel filtration with Biogel P2 and P6 as described in a previous paper (13).

**RESULTS**

**Time Course of Iron Transport**

Ligated jejunal loops were injected with 0.5 ml of 59Fe-p-15 solution containing 100 μg of Fe, and iron absorption by the intestine was determined 30, 60, 120, and 180 minutes after iron administration. The results were expressed as the percentage of total radioiron given which was absorbed per centimeter of intestinal length (Figure 1).

Although net transport of radioiron proceeded sharply in the initial 1 hour and then showed gradual increase with time, the total iron absorption (the sum of the iron transported to carcass plus the iron taken up into the washed gut segment) reached a plateau and showed cessation of mucosal uptake after 1 hour. These data suggested the presence of a control mechanism for iron absorption in the intestinal cells. After a certain amount of iron (7% of p-15 administered per centimeter of intestine in this experiment) permeated the wall of the ligated loop, further uptake was seen to have ceased, due to some saturation phenomena, as shown previously (14, 16).