fluorocarbon emulsion: Fluosol-DA. Thus, it is evident that short-term survival of animals following perfusion to very low hematocrits does not depend on the intrinsic capability of the replacement medium to transport oxygen other than in simple solution.

The present finding of hypernatremia in response to blood substitution with Haemaccel was unexpected since the sodium concentration in this solution is similar to that found normally in rat plasma. However, since the mean plasma sodium concentration in animals immediately prior to exchange-transfusion (table 2) was slightly lower than that found in uncatheterized control animals, it might be expected that extensive perfusion with an essentially isonatric fluid would serve to restore plasma concentrations back to normal levels. Support for this conclusion is provided by previous observations that in the rat, a 3% reduction of plasma sodium concentration occurred in response to repeated blood sampling. Thus, removal of blood for measurement of baseline parameters prior to exchange-transfusion coupled with the slight but unavoidable loss which inevitably occurs during catheterization, would help to explain the reduced initial plasma sodium concentrations seen in the present experiments.

In conclusion, the present finding that a relatively simple, gelatin-based, colloidal plasma volume expander can be used for near total blood replacement in conscious rats provides a convenient model system for assessing the acute responses to blood substitution while avoiding potentially disturbing influences from components of more complex preparations.

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On the pathway of the rectosphincteric reflex

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Summary. In several rat models, including those with circular and semicircular rectal aganglionosis, the rectosphincteric reflex was examined. The reflex was confirmed to be essentially an intramural one and its route is considered to run mainly in the longitudinal and partly in the oblique directions.

The presence of an intact rectosigmoid canal is known to be required for maintenance of the internal sphincter relaxation reflex, or the rectosphincteric reflex, which is widely used for examining Hirschsprung and other chronic constipation diseases. Since the reflex is known to be preserved in cases of lumbosacral meningocele and spinal cord injury, it is considered to be an essentially intramural reflex. However, the details of its pathway are obscure. In the present study, we confirmed experimentally that the reflex was mediated by intramural ganglia and made some interesting observations on its pathway.

Materials and methods. Wistar rats of either sex, weighing 200-400 g, were used. The rat, kept fasted for 24 h and treated with a glycerol enema 2 h before the experiment, was fixed supine under nembutal anesthesia, and a small rubber balloon with a polyethylene catheter, filled with 0.15 ml of water and connected to a pressure transducer, was inserted and fixed within the anal canal. The rat was laparotomized and a rubber balloon with a polyethylene catheter having a content of 2.0 ml, was inserted into the rectal lumen through an incision made at the descending colon and was fixed at a predetermined level. Stimulation was applied by the inflation of the latter balloon with 1.0 ml of air. Square wave electrical stimulation, 0.5 msec in duration, 10 Hz, 30 V and continuing for 3 sec, was applied on the rectal serosa with a bipolar electrode which had an inter-electrode distance of 3 mm. A total or an anterior half rectal transaction was done at the level 3 cm oral to the anal orifice. A total circular or an anterior half rectal aganglionosis, at the same level and about 1.5 cm wide, was produced by serosal application of 0.1% benzalkonium chloride solution 6 weeks before experimental observation, and aganglionosis was later confirmed histologically (fig. 1). A total freeing of the recto-anal canal from the surrounding structures, keeping the internal sphincter muscle intact, was done under nembutal anesthesia; the symphysis pubis and the urinary bladder were removed, and experimental observations were made about 20 min later, when sphincter tonus recovered after a transient drop.

Results (fig. 2). In all 5 normal rats, balloon and electrical stimulations at the rectum up to 6 cm oral to the anal orifice induced a positive rectosphincteric reflex (a transient drop in intra-anal pressure), while balloon and electrical stimulations 8 cm oral to the anus failed to cause the reflex in 1 of 5 rats and in all 5 rats, respectively. In the rats in which the rectum had been totally transected or made circularly aganglionic, both kinds of stimulation of the anal portion always induced a posi-
tive reflex, while stimulation of the oral portion always failed to induce it. In the rats in which the anterior half of the rectum had been transected or made aganglionic, both modes of stimulation of the oral and of the anal portion, except for electrical stimulation at the oral anterior wall, invariably caused a positive reflex. Positivity of the reflex on electrical stimulation of the oral anterior wall depended on stimulation sites (fig. 2): 0 of 8 cases at point 1 (middle point on the oral rim of incision), 1 of 8 at point 2 (1 cm oral to point 1), 4 of 8 at point 3 (1 cm oral to point 2), 6 of 8 at point 4 (corner point on the oral rim of incision) and all 8 at points 5 (1 cm oral to point 4) and 6 (1 cm oral to point 5); 0 of 8 at points 1' and 2', 2 of 8 at point 3', 3 of 8 at point 4' and all 8 at points 5' and 6'. (Points 1' through 6' in the aganglionic model correspond to points 1 through 6 in the transection model, respectively.) It was also confirmed in the rats with an anterior half rectal transection that mucosal and serosal stimulations at the anal anterior wall caused the same response. The reflex was always positive in the rats in which the recto-anal canal was totally freed from the surrounding structures.

Discussion. The rectosphincteric reflex is routinely induced by distention of the rectal wall and the receptor is considered to exist in the mucosal layer. Several authors noticed that the same reflex was induced by electrical stimulation of the rectal mucosa. In the present study, we observed that a rectal serosal electrical stimulation caused the same reflex as rectal mucosal electrical stimulation and also balloon stimulation did. This result is reasonable, supposing that the electrical stimulation causes excitation of intramural nerve processes, which are more excitable than the soma, and square wave pulses less than 1 msec in duration cause no direct excitation of intestinal smooth muscle cells. That an intact continuous rectosigmoid canal is required for maintenance of the reflex was reconfirmed by our observations in the rectal transection and recto-anal freeing experiments. Since the ultrastructural findings on the BC-induced aganglionic rectum demonstrate intact smooth muscle cells, it is obvious that the reflex is mediated by the intramural ganglia and not by longitudinal muscles. The pathway of nerve impulses within the intramural nervous system, leading to relaxation of the internal sphincter muscle, is not specified in detail, but the findings in the present cases of the anterior half rectal transection or aganglionosis suggest that the impulses are transmitted anally mainly in the longitudinal but partly in the oblique directions. In the case of oblique transmission, it may be that the impulses reaching the effector muscles, through a decrement in transsynaptic transmission, are above the threshold in some cases and below it in others, thus resulting in positivity of the reflex below 100%.

Figure 1. Photomicrograph of the BC-induced rectal aganglionosis. The anterior rectal wall, undergoing serosal application of BC solution 6 weeks before, is shown in longitudinal section. Nerve cells in myenteric ganglia have disappeared. HE, × 375.

Figure 2. Examples of the rectosphincteric reflex test in various experimental models, using electrical stimulation. 5 experimental models are shown from above to below: total rectal transection, circular aganglionosis (shaded), anterior half transection, anterior semicircular aganglionosis (shaded) and total freeing of the recto-anal canal (oral side is up). Letters and numbers on the left show stimulation sites. O, oral; A, anal; AM, mucosal surface of the anal anterior wall; 1, middle point on the oral rim of incision; 4, corner point on the oral rim of incision. Points 2 and 3 are distant from point 1 by 1 and 2 cm respectively. Points 5 and 6 are distant from point 4 by 1 and 2 cm respectively. Situation is the same for 1' to 6'. Arrows show stimulation time. Downward deflection shows reduction in intra-anal pressure. Numerators and denominators of the numbers shown at the right extreme are numbers of cases of positive response and of those examined.