

PROTECTIVE EFFECTS OF ANTIBIOTICS ON COLON ANASTOMOSIS

(EXPERIMENTAL STUDIES)

by

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INTRODUCTION

In 1955, ISIDORE COHN performed an end-to-end anastomosis on the colon of the dog, in which he made a 5 cm incision on the mesentery on one side of the anastomosis, and then injected Achromycin solution directly into the lumen of the colon through a thin plastic tube. The result was that the injection of Achromycin proved to be of great help in healing the anastomosis. He emphasized the importance of administration of antibiotics, especially their intraluminal administration.

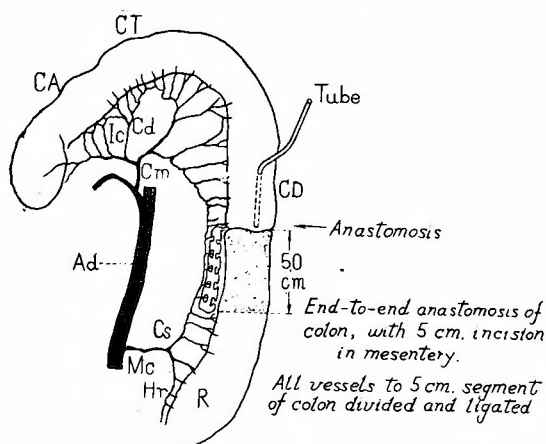
Following his example, I made the below experiments in order both to make a comparative study on the results of the operation varying according to the ways of administering antibiotics, and to examine the possibility of transplanting a completely devascularized segment of the bowel under administration of antibiotics.

METHOD

Healthy adult dogs, weighing 5.0 to 10 kg, were used. Except water, no diet was given to them for 24 to 48 hours before operation.

Operations were performed under aseptic conditions with 2.0 to 5.0 cc of 10% isomytal solution intravenously injected for anesthesia. A mid-line incision was used. After assuring that there was nothing abnormal in the peritoneal cavity, the omentum was resected as completely as possible, and as is shown in Figs. 1 and 2, a segment of the colon descendens was held between two intestinal clamps, and then cut with a scalpel. The lumen of the bowel was cleaned, and an end-to-end one-layer anastomosis was performed with an ALBERT'S continuous suture. A plastic tube (1.5 mm in diameter) was inserted into the lumen of the colon on the healthy oral side of the anastomosis, so that its tip might remain adjacent to the anastomosis. The tube was then fixed by the method of tobacco bag suture. Beginning at the anastomosis and proceeding 5 cm in the direction of the anus, all vessels to the colon were doubly clamped, divided, and ligated. The abdomen was then closed in three layers.

For sewing materials, #4 silk suture was used for the mesentery, fascia, and skin, and #2 silk suture for the anastomosis.

Fig. 1 Diagram of Operative Procedure.

CA	Colon ascendens.	Mc	A. mesenterica caudalis.
CT	Colon transversum.	Cm	A. colica media
CD	Colon descendens.	Ic	A. ileocolica,
R	Rectum.	Cd	A. colica dextra
		Cs	A. colica sinistra.
Ad	Aorta descendens.	Hr	A. rectalis cranialis.

**Fig. 2** # 49. Photo taken at operation. End-to-end colon anastomosis, with 5 cm incision in mesentery on one side of anastomosis.

The dogs were given nothing to eat or drink on the 1st postoperative day. On the 2nd day, however, they were given water, and from the 3rd through the 5th day they received a liquid diet, and then, finally, normal animal food.

An autopsy was performed on those animals which died, as soon as possible after death. Those which survived were sacrificed after the 14th postoperative day. At the operation, 10% isomytal solution was intravenously injected for anesthesia. It was checked with naked eye as to whether the wound on the abdominal wall had been supplicated, or if there was anything abnormal in the peritoneal cavity. The colon was then taken out, and cut lengthwise in order to examine the lumen. After that, it was fixed 10% neutral formalin solution so as to measure the length of the mesentery separated. Histological examinations were also made on paraffin section.

In the experiments there were used crystalline dihydrostreptomycin sulfate „Sankyo” and crystalline penicillin G “Banyu” (abbreviated to SM and P respectively in the following pages).

RESULTS

When all the vessels of the segment of the colon were divided in the operation, the segment contracted and gradually changed its color to dark red, as it received no blood supply except through the bowel wall. The subserosal veins were seen enlarged and congested.

In the present experiments, all the dogs were divided into 6 groups as is shown in Table 1. The amount of colon contents varies according to cases, so they were

Table 1; Groups of experiments.

- 1) Group 1; Cases given no antibiotics.
- 2) Group 2; Cases given P and SM intramuscularly.
200,000 units of P and 1.0g of SM intramuscularly given at operation, and after operation, 100,000 units of P and 0.5g of SM intramuscularly given every 6 hours for 7 days.
- 3) Group 3; Cases given P and SM intraluminally.
200,000 units of P and 1.0g of SM, both in powder, sprayed in the lumen of colon at operation, and immediately after operation, 100,000 units of P and 0.5g of SM, both dissolved in 10 cc of sterilized water, introduced into the lumen through a thin plastic tube every 6 hours for 7 days.
- 4) Group 4; Cases given P and SM first intraluminally and later intramuscularly.
P and SM, at first, given intraluminally, but later intramuscularly due to the plastic tube slipping out.
- 5) Group 5; Cases given P and SM intraluminally, and procaine penicillin G intramuscularly.
Immediately after operation, 600,000 units of procaine penicillin G suspension given once a day, together with intraluminal P and SM.
- 6) Group 6; Cases belonging to the above 5 groups, on which autopsy was performed 48 hours after operation.
- 7) Group 7; Cases on which transplantation of completely devascularized and freed segment of colon or ileum was performed.

divided into 4 classes, that is, from 'very little', through 'a little', 'much', to 'very much' (shown in the following as '+', '++', '++', and '+++', respectively).

(1) Group 1; cases given no antibiotics (Table 2.)

After operation, all three cases belonging to this group showed a very bad appetite, vomited, and were much weakened until they died 36 to 88 hours after the operation. The amounts of bowel contents were within the range of 'a little' to 'much', and had nothing in particular to do either with the general conditions the animals showed after the operation, nor with the local changes in the operated area.

As mentioned above, an autopsy was performed as soon after their death as possible. No case was found with a wound infection, but in the peritoneal cavities of them all there was a lot of foulsmelling, thick purulent fluid. The serosa of the devascularized portion was tinged with dark purple, and the colon wall near the suture line had become jellied, with perforation observed here and there.

In #12 and #50, perforation was so large that it may better be said that the colon had been divided at the suture line. Needless to say, fibrinous adhesion was observed in the loops of the small bowel adjoining the anastomosis, and edematous hyperemia in the serosal surface. The specimen disclosed that the length of the incision made in the mesentery was 5 cm in all cases by actual measurement; that in #49 and #12 the whole devascularized segment, while clearly distinguishable from the adjoining normal bowels showed a dark brown color all around the bowel, indicating a state of necrosis, with its folds disappearing; and that in #50 this color change was observed only in that part of the devascularized portion near the anastomosis site. These findings may be said to show that the amount of blood supplied to the area of devascularization through intramural vessels varied according to cases (Fig. 3).

The histological findings of the devascularized segment revealed that beginning from the intact bowel and proceeding in the direction of the anastomotic line, cha-

Table. 2 Group 1; Cases given no antibiotics.

Operative procedure : End-to-end anastomosis of colon, with 5 cm incision in mesentery on one side of anastomosis.

Dog No.	Weight (kg)	Sex	Survival time (Hours)	Colon contents at operation	Infection of abdominal wound	Autopsy findings			Findings of mucous surface in devascularized segment	Length of incision in mesentery by actual measurement (cm)
						Ascites	Perforation	Adhesion		
12	7.0	♂	44H dead	+	—	+++ foul —	+	+	dark brown belt encircling whole devascularized segment, folds disappearing, edematous.	5.0
49	5.0	♂	36H dead	++	—	smelling pus	+	+	// //	5.0
50	5.0	♀	88H dead	++	—	+++ slightly bloody pus	+	+	part of colon wall disappearing within range of 2 cm from suture line further within range of 1 cm, colon wall changing into gray, folds disappearing, edematous.	5.0

Table. 3 Group 2; Cases given P and SM intramuscularly.

Operative procedure : End-to-end anastomosis of colon, with 5 cm incision in mesentery on one side of anastomosis.

Dog No.	Weight (kg)	Sex	Survival time (Days)	Colon contents at operation	Infection of abdominal wound	Autopsy findings			Findings of mucous surface	Length of incision in mesentery by actual measurement (cm)
						Ascites	Adhesion	Perforation		
33	7.0	♀	14D sacrificed	±	—	—	+++ with	—	smooth shallow groove about 1.5cm in width along anastomotic line	1.0
35	7.5	♀	// //	++	—	—	small bowel,	—	 1.0cm	4.2
42	5.0	♀	// //	+	—	—	spleen	—	 1.3cm	1.3

nges appeared first in the mucosa, next in the submucosa, and then in the muscular coat until complete necrosis spread over all layers. First, necrobiosis appeared in the epithelial cells of the mucosa, and cell infiltration in the propria mucosae, with the blood vessels enlarged. Then, the submucosa was seen edematous and congested, and as the edematous condition grew more marked and cell infiltration spread all over the muscular coat, necrosis came to show itself in all the layers. In the gangrenous area, however, a large number of vessels were seen enlarged and degenerated. (Fig. 4)

(2) Group 2; cases given intramuscular injection of both SM and P (Table. 3)

The cases belonging to this group received 1.0g of SM and 200,000 units of P intramuscularly when vessels were divided during the operation. After the operation, however, 0.5 g of SM and 100,000 units of P were injected every 6 hours for 7 consecutive days.

Compared with those of group 1, the cases of this group showed neither anorexia, nor vomiting, with only slight disorders in general conditions for a short period.

All three cases of group 2 survived more than 2 weeks and were then sacrificed. At autopsy, none of them had the abdominal wound suppurated, nor any free peritoneal fluid; all of them, however, had the operated portion of colon firmly adherent to the near-by loops of small bowel and the spleen. The findings of the specimen of the mucous surface were much the same in all cases, showing a shallow, smooth groove 1 to 1.5 cm in width encircling the lumen of the devascularized segment of the colon along the anastomotic line. In #42, a part of the segment was slightly stenotic, and the mucosa of its neighbouring area was edematous. No perforation was observed in any of the cases. The length of the incision made in the mesentery had become 4 to 4.5 cm by actual measurement. (Fig. 5)

The histological findings, which were much the same in all cases, showed that in the above-mentioned shallow smooth groove, mucous membrane had exfoliated to be replaced by ulcerous tissue developed; and that deep in the granulation tissues there were found the remains of muscular fibers. In the colon wall adjoining the ulcerous parts, the lamina propria mucosae and the submucosa suffered a slight degree of cell infiltration. (Fig. 6)

(3) Group 3; cases given both SM and P intraluminally (Table. 4)

1.0g of SM and 200,000 units of P, both in powder, were sprayed in the lumen of the colon at the time of anastomosis. Following the operation, 0.5 g of SM and 100,000 units of P, dissolved together in 10 cc of distilled water, were introduced into the colon through a thin plastic tube every 6 hours for 7 consecutive days.

In 5 cases, two survived and three died. The two cases that survived had 'a little' or 'very little' contents in the colon at the time of the operation. Afterwards, they recovered fairly well, with good appetite, and no vomiting at all.

They were sacrificed more than 2 weeks after the operation. Autopsy disclosed that in #32 there were small abscesses in the abdominal wound, and a little yellowish serous peritoneal fluid, while there was nothing wrong with #41. In both cases the operated segment of the colon was seen to be only slightly adhered to the nei-

Table. 4 Group 3 ; Cases given P and SM intraluminally.

Operative procedure : End-to-end anastomosis of colon, with 5 cm incision in mesentery on one side of anastomosis.

Dog No.	Weight (kg)	Sex	Survival time (Days, Hours)	Colon contents at operation	Cause of death	Autopsy findings				Findings of mucous surface	Length of incision in mesentery by actual measurement (cm)
						Infection of abdominal wound	Ascites	Adhesion	Perforation		
32	7.0	♂	14D sacrificed	+		(+) small abscess	(+) light yellow, transparent	(+) small bowel, spleen	-	slight degree of congestion and edema	3.8
41	8.5	♀	19D sacrificed	±		-	-	(+)	-	narrow smooth belt encircling colon along suture line	4.0
40	7.0	♀	120H dead	+	Perforative peritonitis	-	(++) purulent	(+) spleen	+	necrotic color change covering over 1/3 of devascularized area along suture line	4.3
25	8.0	♂	60H dead	+		-	(++) bloody purulent	(+)	+	dark reddish brown necrotic change spreading over all	7.0
39	6.0	♂	60H dead	±		-	(++) purulent	(++) small bowel	+	devascularized segment	6.0

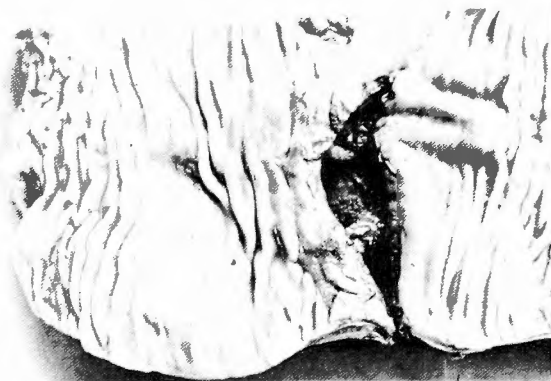


Fig. 3 #50. Appearance of mucosa at the site of anastomosis. The dog received no antibiotics.



Fig. 4 Microscopic appearance of #50, showing necrosis of all layers of colon wall. (H-E stain. $\times 40$)



Fig. 5 Appearance of mucosa at the site of anastomosis, in #35 which received intramuscular SM and P.



Fig. 6 Microscopic appearance of devascularized colon wall of #33 which was given intramuscular SM and P. Note ulcers developed and muscular fiber remaining deep in granulation tissues. (H-E stain $\times 40$)

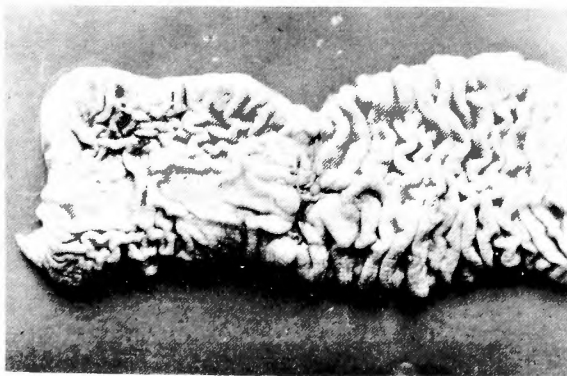


Fig. 7 Mucosa of #32 at site of anastomosis. The dog survived due to intramuscular administration of SM and P.

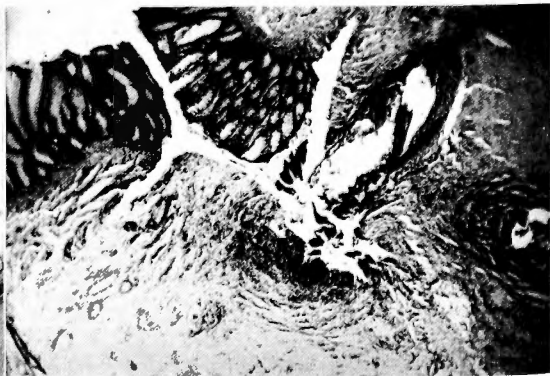


Fig. 8 Microscopic appearance of #32, showing little granulation tissue. (H-E stain $\times 40$)

hanging loops of small bowel and the spleen.

The specimen of the colon anastomosis showed that in #32 congestion and edematous swelling were apparent only to a slight degree, the former on the mucous surface at the anastomotic line, and the latter in the folds of mucosa. (Fig. 7) A case (#41) had a smooth belt about 0.5 cm in width running along the anastomotic line, in the vicinity of which the mucous surface had become swollen and edematous, and the anastomotic site was slightly stenotic.

By histological examinations of #32 it was found: that the mucosa of the devascularized area remained almost normal near the anastomosis; that both sides of anastomosis were connected together with a small amount of granulation tissues, which were ulcerous; that fibrocytes increased and gathered around the suture, where cell-infiltration was dominant; that connective tissues grew abundantly in the submucosa of the devascularized segment, with the deposition of hemosiderin.

In the case of #41, the histological findings were much the same, showing ulcers along the anastomotic line, and muscular layers interrupted here and there by the granulation tissues.

As mentioned above, 3 dogs died as a result of the operation. One of them (#25) died 60 hours after the operation without recovering its appetite. Another (#39) regained its appetite with other general conditions improving as time passed, but after the 1st evacuation that occurred at the 54th hour following the operation, it suddenly began to vomit and rapidly became weakened, until it died at the 60th hour. The 3rd one (#40) was recovering favourably after the operation until the 1st evacuation at the 72nd hour, when it began to vomit, became much weakened, and died at the 120th hour.

An autopsy was performed on each as soon after death as possible. None showed suppuration in the abdominal wound; however, there was a lot of foul-smelling, purulent fluid in the peritoneal cavity, and the serosal surface of the devascularized portion of the colon had become a dark purple color, with perforations ranging from the size of a red bean to the tip of the little finger—a state of perforative peritonitis; and that only a slight degree of adhesion was observed in those loops of the small bowel adjoining the anastomosis.

The findings of the mucous surface of the specimen disclosed that in all of the three cases, either the whole region of devascularization or the part near the anastomosis had become dark reddish brown and necrotic, with its folds disappearing. The length of the incision made in the mesentery had become 4.0 to 7.0 cm by actual measurement. (Fig. 9)

The histological findings revealed that all the layers were necrotic at the anastomotic line of the devascularized portion; that a little farther from that part, cell infiltration was observed on the muscular layer; and that thrombosis occurred in enlarged veins. Each layer of the bowel had been degenerated, and hemorrhage and congestion was observed throughout the mucosa. Hemolysis was observed, and veins were seen enlarged in the submucosa. (Fig. 10)

(4) Group 4; cases given SM and P first intraluminally, and later intramuscu-

Table. 5 Group 4 ; Cases given P and SM first intraluminally and later intramuscularly.
Operative procedure : End-to-end anastomosis of colon, with 5 cm incision in mesentery on one side of anastomosis.

Dog No.	Weight (kg)	Sex	Colon contents at operation	Period of administration of antibiotics		Survival time (Hours)	Autopsy findings			Length of incision in mesentery by actual measurement (cm)	
				intraluminal use (Hours)	intramuscular use (Hours)		Ascites	Adhesion	Perforation		Findings of mucous surface
37	9.0	♀	##	18H	150H	260H dead of peritonitis	(##) bloody purulent	+	+	necrotic color change wholly in devascularized area	5.0
34	7.5	♀	##	24H	144H	140H dead of peritonitis	(##) purulent	##	+	...locally...	4.0
31	6.2	♂	±	48H	(-)	160H dead of peritonitis	(##)	##	+	.. locally...	4.5
38	5.0	♀	+	60H	18H	78H dead of peritonitis	(##)	+	+	...wholly...	4.5
26	10.0	♂	##	84H	18H	100H dead of peritonitis	(##) slightly bloody purulent	+	+	...locally...	6.8

Table. 6 Group 5 ; Cases given P and SM intraluminally, and procaine penicilline G suspension intramuscularly.
Operative procedure : End-to-end anastomosis of colon, with 5 cm incision in mesentery on one side of anastomosis.

Dog No.	Weight (kg)	Sex	Colon contents at operation	Period of administration of antibiotics (Days, Hours)			Survival time (Days, Hours)	Autopsy findings		Length of incision in mesentery by actual measurement (cm)
				Procaine penicilline G suspension	intraluminal use of P and SM	intramuscular use of P and SM		Perforation	Findings of mucous surface of devascularized segment	
28	6.0	♂	+	4D	7D	(-)	31D sacrificed	-	annular smooth belt along suture line	3.5
24	9.5	♂	##	1D	102H	66H	61D sacrificed	-		4.0
27	9.0	♀	##	1D	108H	6H	113H dead of peritonitis	+	necrotic	5.0

larly (Table. 5)

After operation, the dogs of this group received SM and P through a thin plastic tube inserted into the lumen of their colons for some hours, until the tube slipped out of the bowel and intraluminal administration became impossible. In four dogs (#26, #34, #37, and #38,) however, administration of antibiotics was continued intramuscularly until the 150th hour, while in one dog (#31) it was discontinued.

All cases died between the 78th and 260th hour. Autopsy was performed as soon after death as possible. It disclosed that none of them had a purulent wound on the abdominal wall. Except #37 which had hemorrhagic purulent fluid in the peritoneal cavity, the rest suffered from perforative peritonitis, with foulsmelling pus in the peritoneal cavity. The findings of the specimen of the mucous surface showed that a dark brown necrotic belt encircled the bowel, either over all the devascularized segment or locally along the anastomotic line. Perforation was found in all of the cases, and in #34 and #26 an almost complete separation of the anastomosis was observed.

Histologically, vessels in the muscular coat or the subserosa had become enlarged, and thrombosis and perivascular infiltration was noticed. In the circular muscular layer or submucosa, a more or less high degree of stratiform hemorrhage was observed, and all layers or layers inner than the circular muscular layer were necrotic.

In this groups, the periods of intraluminal administration of antibiotics varied according to cases. The tendency was that the shorter the period, i. e., the sooner intramuscular injection was begun, the longer they survived.

In #31 which had a little contents in the colon, the administration of antibiotics was discontinued at an early stage. From histological examinations, however, the findings of anastomotic area in this case showed a state rather better healed, than in the other four cases.

(5) Group 5; cases given both procain penicillin G suspension intramuscularly, and SM and P intraluminally at the same time (Table 6)

To this group 600,000 units of procaine penicillin G suspension were intramuscularly injected together with intraluminal administration of SM and P.

In one case (#28) the intraluminal administration was continued for 7 days, but in the other two, it was discontinued and replaced by intramuscular injection because the plastic tube slipped out.

The case (#28) survived well for 31 days after the operation, and was then sacrificed. In the specimen, the mucous surface of the devascularized area had a smooth belt 0.7 cm in width encircling the bowel along the anastomotic line. (Fig. 11) Histologically, there were ulcers developed and some longitudinal muscular layer was observed deep in the granulation tissues. (Fig. 12)

The 2 cases, #24 and #27, were much the same in respect to weight, amount of contents of the colon, and other conditions. In both, the intraluminal administration was discontinued about four and a half days after the operation to be replaced by intramuscular injection because the tube slipped.



Fig. 9 Devascularized segment of #25 which died in spite of intraluminal P and SM. Not necrotic perforation over all devascularized portion.

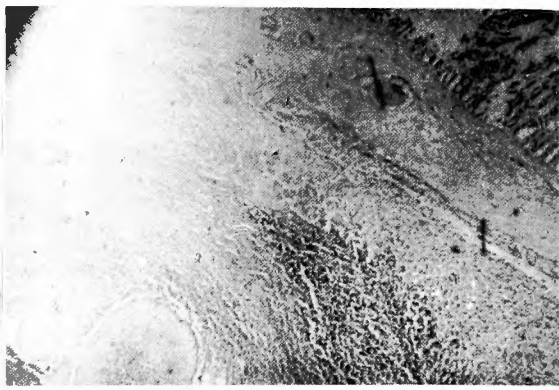


Fig. 10 Microscopic appearance of #25. Necrosis and hemorrhage are seen in all layers of colon wall. In muscular layer, cell infiltration, enlargement of veins, and thrombosis are marked.



Fig. 11 #28. Appearance of mucosa at the site of anastomosis. The dog survived under intramuscular procaine penicillin G and intraluminal P and SM.



Fig. 12 Microscopic appearance of #28. Ulcers developed, and some longitudinal muscular layer seen deep in granulation tissues. (H-E stain $\times 40$)



Fig. 13 Microscopic appearance of #1 (receiving no antibiotics), 48 hours after operation. Necrosis spreading over all layers of colon wall, with thrombosis and enlargement of vessels. (H-E stain $\times 40$)

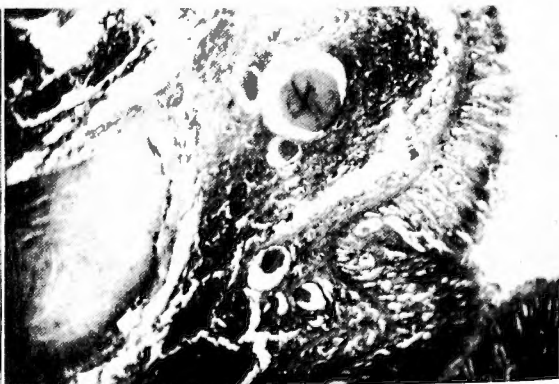


Fig. 14 Microscopic appearance of devascularized colon of #52, 48 hours after operation. The dog received intramuscular P and SM. Note only slight degeneration in lamina muscularis mucoae, although high degree of hemorrhage is seen and vessels are greatly enlarged. (H-E stain $\times 40$)

The dog (#27) died after 108 hours. The autopsy disclosed it had perforative peritonitis. The whole mucous surface of the devascularized portion had become dark brown and necrotic, with the folds of its mucosa missing, and two ulcers about the size of the tip of the little finger had developed at the anastomosis. On the other hand, #24 survived for 61 days, and then was sacrificed. The findings of the anastomosis were much the same as those of #28, both histologically and with the naked eye. In the former, the portion of anastomosis was observed to be much better healed than in the latter.

The histological examination of #27 revealed that there was a high degree of cell infiltration between the two layers of the muscular coat near the suture line, that the tissues of all layers were in a state of necrosis, and that the submucosa was hemorrhagic, with its veins greatly enlarged. A high degree of stratiform hemorrhage was found in the submucosa and circular muscular layer, in the outer layers of which was observed cell infiltration. Perivascular infiltration was also found in the submucosa, though to a slight degree.

COMMENT

In the above experiments, an end-to-end one-layer anastomosis was performed on the colon of the dogs, in which a 5 cm incision was made in the mesentery on the anal side of the anastomosis in order to produce almost complete devascularization in the segment of anastomosis. All cases of untreated group died of peritonitis caused by necrotic perforations on the devascularized portion. 16 cases received antibiotics in various ways after the operation, and 7 of them survived, while the other 9 died of perforative peritonitis (Table 7). These 9 cases, however, survived longer than those of group 1 did, which proved the effectiveness of antibiotics in protecting colon anastomosis (Table 8).

Table. 7 Effect of antibiotics

Operative procedure : End-to-end anastomosis of colon, with 5 cm incision on one side of anastomosis

		Number of cases	Dead	Survived
Cases given antibiotics	Intramuscular P and SM	3		3
	Intraluminal P and SM	5	3	2
	First intraluminal, later intramuscular P and SM	5	5	
	First intraluminal, later intramuscular P and SM with intramuscular procaine penicillin G	3	1	2
	Cases given no antibiotics	3	3	

How did the results of the experiments vary according to the different way of administering antibiotics? All cases which received intramuscular administration survived, showing a much higher rate of survival than those which received intraluminal administration. The former all had ulcers developed at the anastomosis, while the devascularized segments of the latter indicated varying stages according to

Fig. 25 Relation between different methods of administering antibiotics and amounts of large bowel contents at operation.

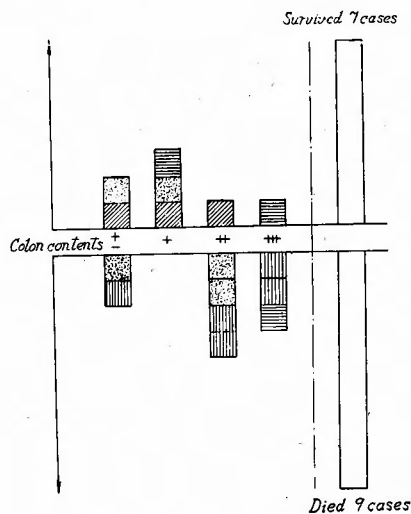


Fig. 26 Amounts of colon contents in cases which received antibiotics intraluminally.

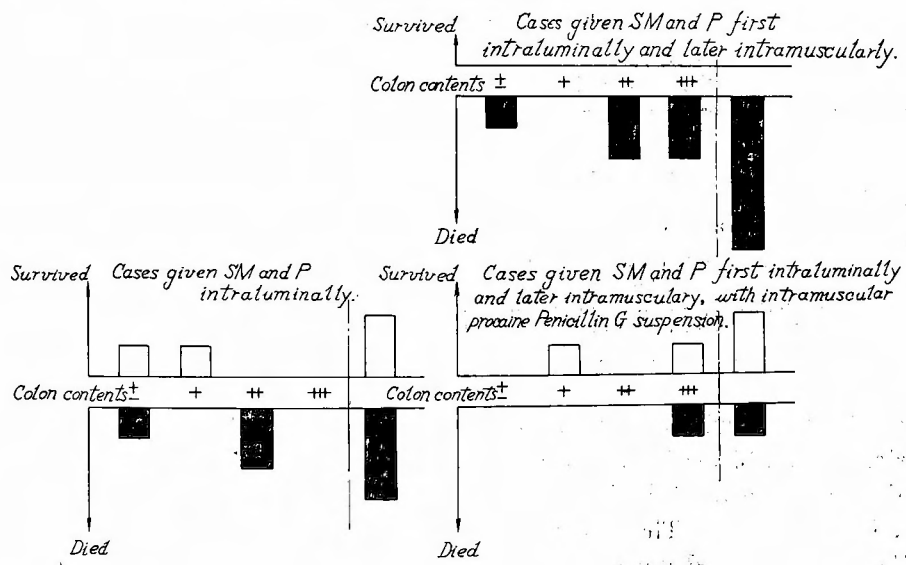


Table. 9 Changes 48 hours after operation.

	Dog No.	Sex	Weight (kg)	Findings 48 hours after operation			Histological findings	
				Ascites	Perforation	Findings of mucous surface of devascularized segment		
Cases given no antibiotics	1	♀	8.0	(#)	(+)	dark brown, necrotic,	Changes in tissues Hemorrhage Vessels Red cells Cell infiltration	necrosis in all layers only a trace necrotic enlarged hemolytic slightly
	13	♂	7.0	purulent	separated	jellied		
Cases given intraluminally	10	♀	9.5			dark reddish,	Changes in tissues Hemorrhage	slightly degenerated high degree
	11	♀	8.5			edematous	Vessels Red cells Cell infiltration	full of blood cells, slightly degenerated no hemolysis high degree
Cases given intramuscularly	52	♂	10.0					
	56	♀	9.0	(+) serous		folds disappeared, dark reddish brown		

cases.....some with the suture line almost completely healed, some with ulcers, and others with perforations or slough of a anastomosis.

Of the cases which were given antibiotics intraluminally, only those whose colon had 'very little' or 'a little' contents survived. This may mean that

whether or not antibiotics intraluminally given take effect depends upon the amount of contents in the colon, but at the same time, the fact that some cases which had only little contents in the colon died, shows that antibiotics given by the intraluminal route do not always pervade evenly inside the colon. (Figs. 25 and 26)

(6) Changes in the cases of the above groups observed at the same time, i. e., 48 hours after the operation (Table 9)

In order to see what changes took place in the cases belonging to the above-mentioned groups within the same hours, 6 cases were sacrificed 48 hours after the operation, including one which died at the 44th hour. The autopsy showed that 2 cases of group I had perforative peritonitis, with massive foul-smelling pus, and an almost complete separation of the anastomosis. In the other groups, 56 had 'a little' serous peritoneal fluid, and the mucous surface of the devascularized portion had become edematous and dark reddish brown, with its fold missing. On the contrary, the others had no peritoneal fluid though the mucous surface showed the same changes.

Histological examinations were made. In group 1, the devascularized portion of anastomosis had become necrotic from the mucosa through all the layers, and in the submucosa and tunica muscularis the vessels were observed enlarged and degenerated to quite a high degree, and a trace of hemorrhage was also recognized. (Fig. 13) The changes seen in the cases of group 2 and 3 were much the same. The mucosal epidermis had become necrotic over all the devascularized portion, although it remained normal here and there at the bottom of the crypt. The vessels were seen enlarged on the propria mucosae, which appeared hemorrhagic. The muscularis mucosae, though slightly degenerated, mostly remained normal, while the submucosa showed a high degree of hemorrhage. The vessels of the submucosa were full of erythrocytes, but their walls were only slightly degenerated, with no hemolysis. Perivascular infiltration was observed only to a slight degree. The inner layer of the muscular coat (circular muscular layer) had been degenerated and strongly hemorrhagic, while the outer layer (longitudinal muscular layer) showed no great changes, though the vessels were a little enlarged, with a slight degree of hemorrhage and perivascular infiltration (Fig. 14).

(7) Group 7; cases on which transplantation of a completely devascularized and freed segment of the bowel was performed (Table 10)

In the above experiments, the site of the anastomosis completely healed in some cases even though the blood supply was cut off either by the devascularization itself

Table. 8 Survival hour of dead cases

Methods of administration of antibiotics	Average survival hours
Cases given no antibiotics	56 H
Cases given antibiotics intraluminally	80 H
Cases given antibiotics in other ways	178 H

Table. 10 Cases where transplantation of completely devascularized segment of bowel was performed.

Methode of operation	Dog No.	Sex	Weight (kg)	Covering with omentum	Use of antibiotics	Survival time (Days, Hours)	Findings of specimen			
							Perforation	Length of transplanted segment (cm)	Shape of transplanted segment	Stenosis
Cases where a 5 cm segment of colon was once separated and then put back where it had been	29	♀	7.0	+	P and SM intraluminally	17 D sacrificed	—	1.5 ~ 1.0	belt shaped	+
	43	♂	5.0	+		25 D sacrificed	—	0.5 ~ 0.8	// //	+
	45	♀	6.0	+		25 D sacrificed	—	0.5	// //	±
Cases where a 5 cm segment of ileum was transplanted	36	♀	10.0	+	—	60 H dead of peritonitis	separation of suture	4.0	// //	—
	30	♀	7.0	+	P and SM intraluminally	29 D sacrificed	—	2.8	fan shaped	##
	44	♀	7.5	+		30 D sacrificed	—	2.0	// //	+
	46	♂	7.6	+		151 H dead of peritonitis	+	5.0	belt shaped	—
	48	♂	13.0	—	P and SM intraluminally	21 D sacrificed	—	1.5	fan shaped	##
	47	♂	12.0	+	P and SM intramuscularly	23 D sacrificed	—	1.5	// //	##

Fig. 15 Diagram of operative procedure of transplanting completely devascularized and freed segment of bowel.

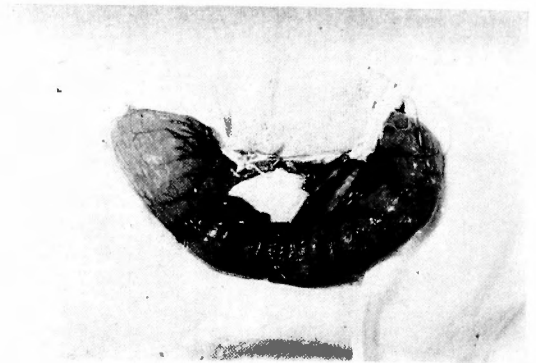
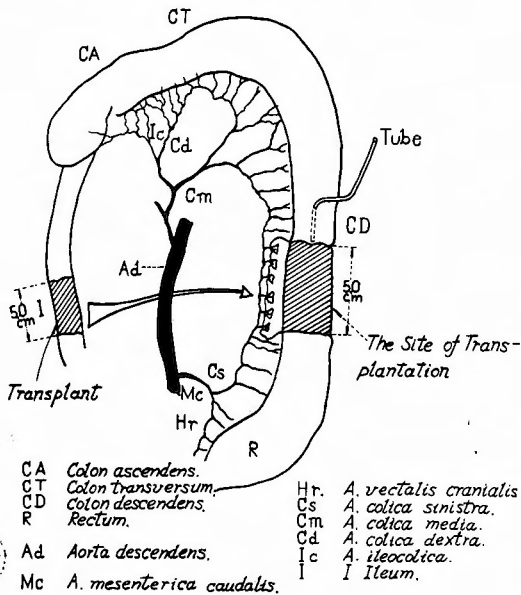


Fig. 16 #43. A segment of colon, after being divided and freed, was put back where it had been.

or by thrombosis and other factors. From these results, I concluded that it might be possible to transplant a completely devascularized free segment of a bowel. For study in this connection, experiments were made on 9 dogs.

i) Group i; cases in which a 5 cm long segment of colon was completely divided and freed, and then put back where it had been (#29, 43 & 45).

Figs. 15 and 16 show how the operation was carried on. A 5 cm long segment of the descending colon was divided and freed after completely devascularized by cutting its mesenteric vessels. The segment was then placed where it had been, and fixed there by means of end-to-end one-layer anastomosis. A thin plastic tube was introduced into the lumen from the healthy oral side of the anastomosis. The replaced portion was covered with the omentum, and the abdomen was closed in three layers. Then, antibiotics was given in quite the same way as mentioned before.

The 3 cases of this group survived the operation and were later sacrificed. The autopsy showed no purulent abdominal wound, but in the operated area a high degree of adhesion of the omentum and adjoining loops of small bowel to the site of anastomosis was observed.

The planted segment in all cases was found to be much shortened—1/3 to 1/10 times as long as it had been. Except #45, the other 2 cases had their planted section greatly strictured. (Fig. 18)

Histologically speaking, ulcers developed in the planted section, and connective tissues grew abundantly, in which were seen muscular fibers remaining. No new

epidermis, however, was found covering over the planted segment. (Fig. 19)

ii) Group ii; cases where a 5 cm completely devascularized and freed segment of ileum was transplanted.

A 5 cm segment was cut off from about the same portion of colon as in group i, into which was transplanted a 5 cm segment of ileum cut off from 20 cm oral to the ileocolic sphincter, by means of end-to-end one-layer anastomosis which was performed with a continuous suture using Albert's method. (Fig. 17) The transplanted section was covered with the omentum, and then the abdomen was closed in three layers.

6 cases belonged to this group. One was given no antibiotics, but four received intraluminal administration (including one in which the transplanted portion was not covered with the omentum), and the other had intramuscular injection.

a) Cases A; given no antibiotics. (#36)

This case died two and a half days after the operation. The autopsy showed it had perforative peritonitis, with its transplanted segment nearly separated from its adjoining normal bowels at both suture lines, and the lumen was found dirty and dark brown. (Fig. 20)

Histologically, the tissues of the transplanted segment had disappeared, only to be replaced by the omentum and fibrinous masses. In other words, the segment can not possibly be considered as constituting a part of the bowel wall. (Fig. 21)

b) Group B; cases given antibiotics intraluminally. (#30, 44 & 46)

Of the 3 cases of this group, one died of perforative peritonitis 6 days and 7 hours after the operation, having two perforations about the size of the tip of the little finger in the transplanted portion. (Fig. 22) Histological examinations revealed that the transplanted section had disappeared, the omentum and fibrinous masses replacing it, just as was the case with case A. No new vessels were seen growing there, and only very few fibrocytes were found.

The other 2 cases survived and were sacrificed on the 30th and 39th day respectively. In both, the healthy colon on the oral side of the transplanted portion was much enlarged, while the transplanted portion was very much shortened $\dots 3/5$ to $2/5$ times as long as it had been when transplanted $\dots$ and fanshaped in the oral direction. (Fig. 24)

c) Case C; given antibiotics intramuscularly. (#47)

This case survived and was sacrificed 23 days after the operation. The autopsy showed that the transplanted segment had been greatly constricted, becoming $1/3$ as long as it had been when grafted, and resembling a fan in shape. Histological findings were much the same as those of group B; there were scarcely any muscular coat or newly growing epithelial cells.

d) Case D; given antibiotics intraluminally, but no covering with omentum (#48).

This case survived for 21 days and then was sacrificed. At autopsy, the loops of small bowel were observed strongly adherent to portions of the transplanted segment, which had been much strictured and fan-shaped, becoming $1/3$ as long as it had been when transplanted. Histologically, there were very few muscular fibers



Fig. 17 #45. Appearance of completely divided and freed segment of ileum transplanted into the loop of colon.



Fig. 18 Appearance of colon anastomosis of #43, in which completely divided and freed segment of colon was replaced where it had been.

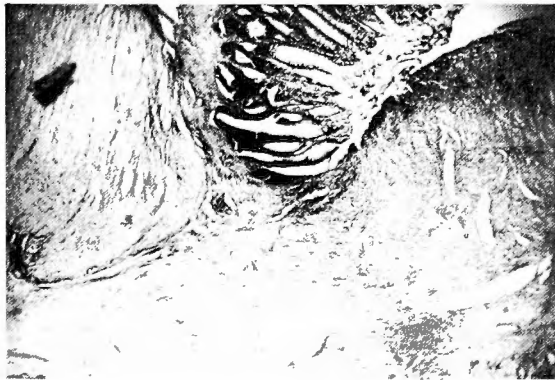


Fig. 19 Microscopic appearance of replaced segment of colon of #43. Note ulcers developed and muscular fibers remaining, but no new epithelial cells growing.

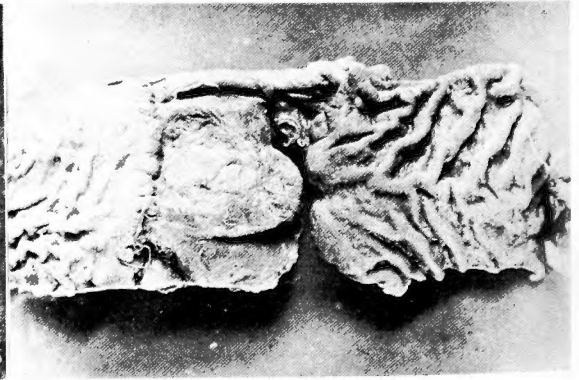


Fig. 20 Appearance of completely divided and freed segment of ileum transplanted into part of colon of #36, which received no antibiotics.

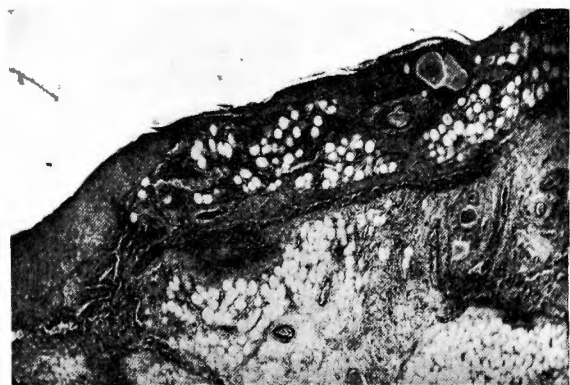


Fig. 21 Microscopic appearance of transplanted segment of #36. Note transplanted segment being replaced by omentum and fibrinous masses. (H-E stain $\times 40$)



Fig. 22 Appearance of completely divided and freed segment of ileum transplanted into part of colon of #46 which died under intraluminal administration of antibiotics.

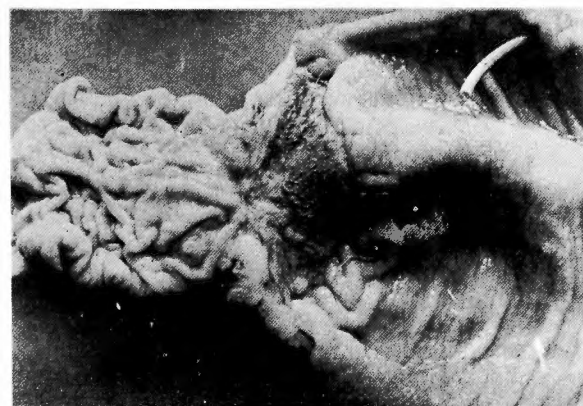


Fig. 23 Appearance of transplanted freed ileum of #30 which survived under intraluminal antibiotics.

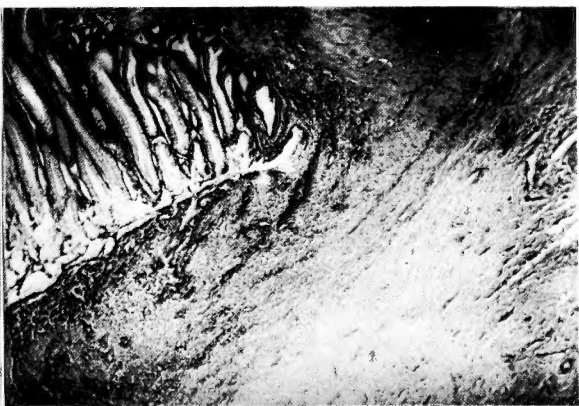


Fig. 24 Microscopic appearance of transplanted freed ileum segment of #44. Either granulation tissues or muscular fibers are scarcely seen.

remaining in the granulation tissues of the transplanted portion.

COMMENT

Summing up and commenting on the results of the above experiments concerning transplantation of a piece of the freed bowel performed on 9 dogs, one case that received no antibiotics died, but all 3 cases survived in which a segment of colon was used, and one of the 5 cases died where a piece of ileum was transplanted.

The transplanted section was observed to be greatly shortened, and constricted, having become, about 3 weeks after the operation, $1/6$ to $1/10$ of the former length in the case of colon, and $2/5$ to $3/5$ of it in the case of ileum. Indeed, this shows that antibiotics are effective in grafting but at the same time, that a completely devascularized segment of bowel cannot be transplanted as effectively as is intended, that is, to work properly as a substitute.

SUMMARY AND DISCUSSION

COHN, POTH, SUMIDA and others proved by experiments that antibiotics are effective in intestinal anastomosis. My experiments came to the same conclusion. If dogs had 'much' or 'very much' contents in the large bowel, intraluminal administration could not help them. On the other, some died even though they had only 'a little' contents in the bowel. This may sound contradictory, but I think it is because antibiotics introduced into the lumen of the colon do not always pervade over all parts of the devascularized area. My experiments are ample proof that by an ideal intraluminal administration, better histological findings of anastomosis could be gained than by intramuscular administration.

Clinically, therefore, at the time of bowel resection, if antibiotics are given by any other route, they should also be given systemically at the same time, which would bring about a constant effect.

The impediments to the healing of gastric or intestinal anastomosis are due to three local factors:

1. The presence of solid or semi-fluid bowel contents and gas.
2. The circulation impediment at the zone of anastomosis.
3. The bacterial invasion (infection).

The method of operation used in the present experiments was intended to bring about quite a high degree of circulation impediment. It is highly, probable, however, that there was circulation left in the devascularized area immediately after making an incision in the mesentery of the colon, for hemorrhage was observed at the cut end of the devascularized colon. What degree of circulation was left, then? The answer depends upon, for example, the condition of vascular anastomoses which exist peripherally to the incision line of the mesentery, as HASEGAWAA asserts. This is true because, in the cases of group 1, which received no antibiotics, the extent of devascularized area that turned necrotic varied according to cases.

The changes subsequently brought about on the tissues of the devascularized segment were influenced by bacterial infection or by different quantities of bowel

contents.

In the cases of group 1 that received no antibiotics, necrotic change—a finding similar to what NIEDERSTEIN termed “hemorrhagische Gangrän”—was found less than 48 hours after the operation. On the contrary, in those cases which were given antibiotics, no necrotic change was observed in any layer of the bowel wall, though there was a high degree of hemorrhage, with the veins greatly enlarged. In the light of these findings, RAEINOVICI and COHN correctly said that antibiotics were effective in preventing both a circulation impediment caused by thrombosis due to bacterial infection or toxin, and a necrosis in the tissues.

It may be added here that even when complete devascularization has been brought about, administration of antibiotics, together with neighbouring viscera adherent to the segment, helps to some extent to protect against necrosis of the transplanted segment and ultimate death, but that the transplanted section cannot be a substitute for the original bowel.

CONCLUSION

The present experiments made on the colons of dogs comprised two parts. In one, end-to-end one-layer anastomoses were performed with a 5 cm incision made in the mesentery on one side of the anastomosis. In the other, transplantation of a 5 cm completely devascularized and freed segment of either large bowel or ileum was performed.

In both, antibiotics (SM and P) were administered in different ways and quantities, in order to evaluate their effects on the operation. The results were as follows:

(1) Among those cases where an end-to-end one-layer anastomosis was performed on the colon with an incision made in the mesentery on one side of the anastomosis, all the cases which did not receive antibiotics died, while all that treated with intramuscular administration of antibiotics survived, and two of the five cases that received intraluminal administration survived.

This is proof that antibiotics are effective in protecting bowel anastomosis.

(2) The cases given intramuscular antibiotics showed, with no exception, band-like formation of ulcer along the suture line, whereas those treated intraluminally indicated varying stages from an almost complete healing of the anastomosis to a development of ulcers and perforations caused by necrotic changes in the area of devascularization.

(3) One of the reasons why intraluminal administration showed different effects may be because the large bowel contents varied according to cases. Clinically, therefore, at the time of operation, antibiotics should be given not only locally but also systematically (intramuscularly), with the object to bring about comparatively constant effects.

(4) Among those cases on which transplantation was performed, all those which did not receive antibiotics died, while those which were given some did survive, the only exception being the one in which a piece of ileum was used, and which received intraluminal antibiotics.

The findings of those which survived showed that the transplanted portion had become ulcerous and highly constricted, with only little muscular fibers remaining deep in the tissues, and that there was no epithelial cells over the area of transplantation. In other words, the transplantation was of no use in the true sense of the word.

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和文抄録

抗生剤の腸吻合に対する保護作用

(実験的研究)

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血行障害に因する腸壊死に対する抗生剤の効果を検討する為に, Isidore Cohn の実験の追試を行い, 氏の重視する管内投与に対する全身投与の成績の比較, 更には抗生剤投与下に於ける遊離自家腸管移植の可能性に就ても検討を加へる為, 実験を行つて以下の如き成績を得た。

A) 腸吻合に於ける抗生剤の効果に就て

健康成犬で, 下行結腸を一旦切断した上で単層連続端々吻合を施行し, 其の一侧の結腸間膜を腸壁に接して5.0cm切離するという, 極めて不安定な腸吻合を作製し, 次で合成樹脂細管を健康部より腸腔内に挿入し, 先端を吻合部に持ち来して留置した。

抗生剤は硫酸ジヒドロストレプトマイシン及結晶ペニシリンGナトリウムを使用し, 管内投与群では其の各々1.0g及20万単位を吻合施行時に管腔内に撒布し, 筋注群では同量を同様時期に筋注した。術直後より毎6時に夫々0.5g及10万単位を管内投与乃至筋注し7日間連続した。別に3例に於て懸独プロカインペニシリンG60万単位を管内投与に併用した。

1) 抗生剤非使用群は何れも血流遮断部の壊死穿孔による穿孔性腹膜炎によつて死亡した。抗生剤を術後に投与した16例中7例生存し9例穿孔性腹膜炎で死亡した。此の死亡例に於ても対照例に比して生存時間は延長していた。此の事実は抗生剤の救命的效果を示している。

2) 抗生剤投与例の内, 筋注群は全例生存したのに反し, 管内投与群は5例中2例生存, 3例は死亡している。一方, 其の吻合部の変化は筋注群では, 何れも血流遮断部に相当して帯状潰瘍を来しており, 其の部分に対する近傍臓器の癒着が高度であつたのに反し, 管内投与群では筋注群に見られないような良好な吻合部の状況を呈するものも認められるが, 又筋注群に於けるが如き帯状潰瘍の形成を示す例もあり, 更には血流遮断部の壊死穿孔を来した例もあつて多様な所見を示し, 近傍臓器の癒着は一般に軽度である。此の事実から管内投与の効果の不安定である事が明となり, 理想的に管内投与が効果を發揮する場合にのみ筋注群に於ける以上に良好な吻合部所見が得られる。

3) 管内投与群に於て生存例は大腸内容量の少量例に限られており, 他方極少量の大腸内容量でも死亡例を見ており, 管内投与の効果の一定せざる事実の一因として, 薬液の腸管内での分散の不規則なる事実が挙げられる。

4) 管内投与実施中合成樹脂細管の滑脱によつて管内投与不能となりその儘投与を中止し, 或いは筋注に切替た5例は全例死亡した。其の際, 管内投与継続の時間の短い程, 換言すれば筋注開始の早いもの程生存期間は長い傾向が認められる。一方, 1例ではあるが懸独プロカインペニシリンG60万単位を術直後たゞ一回併用した例は生存した。此の事は管内投与の不安定

性とSM及Pの腸管内腔よりの非吸収性とを併せ考えて、術後早期よりの全身投与の重要性を示唆するものと考えられる。

5) よつて臨床的に腸切除時に抗生剤を投与する場合、常に一定した効果ある全身投与は不可欠と考えられる。

B) 遊離腸管自家移植に於ける抗生剤の効果

健康成犬の下行結腸に於て其の5.0cmを切離し、腸間膜をも完全に切離し、完全に遊離した後再度単層端々連続吻合によつて連続せしめ、其の部を大網にて包み上述の管内投与を施行した。3例の全例は生存した。又廻腸片5.0cmを完全に切除し、其の遊離廻腸片を下行結腸部に移植した例に於ては、抗生剤非使用例は穿

孔性腹膜炎で死亡し、大網で被覆し管内投与を行つた3例の内1例は死亡した。又大網で被覆し筋注を行つた1例及び大網で被覆する事なく管内投与を行つた1例は共に生存した。生存例では何れも移植片の短縮を見、程度の差はあるが狭窄を認め、移植片は筋線維の遺残の僅少な肉芽組織と化し潰瘍を形成しており、上皮の新生は見られなかつた。以上により、完全に腸壁の血行停止を来した場合でも、周囲臓器の癒着と相俟つて、或る範囲の腸壊死に対して抗生剤は救命的に働く事が明らかとなつたが、移植片の本来の代用性と云う意味では、移植された腸管は殆どその役を果し得ないものと考えられる。