ALTERATIONS IN THE MOUSE CECUM AND ITS FLORA PRODUCED BY ANTIBACTERIAL DRUGS*

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Antibacterial drugs added to the drinking water of mice rapidly bring about marked qualitative and quantitative changes in their gastrointestinal flora; these changes are characteristic for each type and concentration of drug (1). It is known, furthermore, that chicks treated orally with penicillin from the time of birth until 4–10 wk of age, have less lamina propria tissue in the mucosa of their small intestine than do untreated controls (2, 3). On the assumption that these facts are related, we have designed experiments to determine whether antibacterial drugs affect the lamina propria of the intestinal wall of adult mice.

In the course of these experiments, it was observed that a variety of changes occurred in the cecum within less than a day after oral administration of penicillin. The size of the cecum was markedly enlarged, and its contents became extremely fluid; its bacterial flora was practically eliminated. Other antibacterial drugs also exerted rapid effects on the cecum, but somewhat different from those of penicillin. The findings with three different drugs are described in the present paper, along with a discussion of the possible role played by microorganisms in gastrointestinal physiology.

Materials and Methods

Mice.—Animals used were 5–6 wk old male mice from the special NCS (4) and NCS-D (5) colonies maintained at The Rockefeller University. They were housed in groups of 6 to 10 in plastic boxes with wood shavings for bedding and given D & G diet and acid-water ad lib. (4).

Antibacterial Drugs.—Penicillin, Terramycin, or kanamycin were administered in the drinking water in the concentrations and for the time periods indicated in the text and tables. When a given drug was first administered, the drinking water containing it was always placed in the box in the early evening (5:00 to 6:00 p.m.). Unless otherwise stated, the dilute drug solution provided the only drinking water available to the animals during the course of an experiment.

Determination of the Wet and Dry Weight of Cecums.—Mice were sacrificed with chloroform, weighed, and autopsied. The cecums were carefully removed, weighed with the contents intact, and then used as described below for dry weight determinations, quantitative enumera-

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tion of bacterial flora, or histology. For dry weights, the cecums with their contents intact were placed in tared aluminum cups and lyophilized in a Virtis lyophilizer (Virtis, Gardiner, N. Y.). The dried cecums were stored in a desiccator over anhydrous calcium sulfate until they were weighed.

TABLE I

Wet and Dry Weights of the Cecums and Their Contents from Mice Given Various Concentrations of Penicillin in Drinking Water

Penicillin	Hours after drug started		
	12	24	48
	Wet weight of cecum and contents		
g/liter			
0.1	0.95* (0.83–1.06)	0.90 (0.48–1.00)	0.87 (0.52–1.11)
0.3	0.91 (0.76–1.01)	0.80 (0.47–0.87)	1.10 (1.03–1.35)
0.6	0.80 (0.67–0.98)	1.18 (1.03–1.43)	1.12 (0.94–1.46)
None	0.41 (0.34–0.46)	0.32 (0.27–0.44)	0.48 (0.35–0.58)
	Dry weight of cecum and contents		
0.1	0.13 (0.10–0.14)	0.13 (0.11–0.15)	0.15 (0.13–0.16)
0.3	0.11 (0.10–0.13)	0.10 (0.08–0.12)	0.16 (0.15–0.22)
0.6	0.10 (0.09–0.11)	0.14 (0.12–0.20)	0.15 (0.11–0.18)
None	0.11 (0.10–0.12)	0.09 (0.07–0.10)	0.12 (0.09–0.15)

* 6–10 mice per group; median and range in parentheses.

Quantitative Enumeration of Cecal Bacterial Flora.—Weighed cecums with their contents intact were homogenized in a glass tissue grinder in 5 ml of charcoal water (6). 10-fold dilutions of the homogenates prepared with charcoal water were spread on the surfaces of various selective agar media for different bacterial groups. The media and methods of incubation have been described elsewhere (6, 7).

Histology.—As described in a previous publication (8), weighed cecums with their contents intact were frozen whole in methylcellulose (2% in 0.15 M saline) on the freezing shelf of a microtome-cryostat (International Model CTD, International Equipment Co., Needham

Heights, Mass.). Sections of the frozen tissues were stained with hematoxylin and eosin or with a Gram stain modified for histological studies.

RESULTS

Early Effects of Penicillin on the Wet and Dry Weights of the Cecum and on its Bacterial Flora.—Mice were given drinking water containing various concentrations of penicillin for 2 days. Some of the animals were sacrificed at sequential times during that period; determinations were made of the wet and dry weights

TABLE II

Effect of Penicillin in Various Concentrations in Drinking Water on the Bacterial Flora of the Cecums of Mice

Penicillin	Bacteria*	Hours after drug started		
		12	24	48
g/liter				
0.1	Lactobacilli	4‡	N	±
	Coliforms	±	N	7
	Enterococci	±	N	4
	Anaerobes	± (±)	N (−)	8 (−)
0.3	Lactobacilli	N	N	N
	Coliforms	±	±	5
	Enterococci	N	N	N
	Anaerobes	N (−)	N (−)	N (−)
0.6	Lactobacilli	N	N	N
	Coliforms	N	N	N
	Enterococci	N	N	N
	Anaerobes	N (−)	N (−)	N (−)
None	Lactobacilli	9	9	9
	Coliforms	4	4	4
	Enterococci	4	5	5
	Anaerobes	10 (+)	10 (+)	10 (+)

* The count of anaerobic streptococci (Gr. N) is included in the count of the lactobacilli.

The coliforms include *Klebsiella-Aerobacter* which often appear in mice given penicillin for 48 hr or more.

The anaerobes include bacteroides and clostridia which can be enumerated by viable counts and fusiform bacteria which have not yet been enumerated by viable counts. The bacteroides predominate on culture media before penicillin administration, the clostridia predominate after. The fusiform bacteria have not yet been cultured on agar media, but can be seen in enormous numbers in histological sections of the cecum (see reference 8).

‡ The data are recorded as the average of the $\log_{10}$ of the number of bacteria per gram of fresh tissue; N, no bacteria cultured; ±, bacteria infrequently cultured in very low numbers.

The parentheses indicate the results of determination of the amounts of fusiform bacteria from histological sections of the cecum; +, huge numbers; ±, infrequently observed in small numbers; −, not observed.

of the cecums and of the numbers of the various types of bacteria in their flora. The cecum weights are given in Table I and the numbers of bacteria in Table II.

Within 12 hr after administration of the drug, the wet weights of the cecums, including their contents, had increased 2- to 3-fold. This effect occurred with

TABLE III

Long-Term Effects of Penicillin in the Drinking Water on the Cecum and Body Weights of Mice

Drug	Days after drug started			
	1	2	30	90
	Wet weight of cecum and contents			
Penicillin*	0.80‡ (0.67–1.06)	0.79 (0.70–1.07)	0.91 (0.75–1.27)	1.04 (0.78–1.23)
None	0.48 (0.39–0.54)	0.41 (0.33–0.58)	0.41 (0.35–0.49)	0.51 (0.49–0.57)
	Dry weight of cecum and contents			
Penicillin	0.11 (0.09–0.15)	0.13 (0.12–0.16)	0.19 (0.16–0.23)	0.25 (0.12–0.30)
None	0.10 (0.09–0.12)	0.09 (0.08–0.10)	0.10 (0.09–0.11)	0.11 (0.09–0.19)
	Body weight§			
Penicillin	22.0 (17.5–22.7)	20.5 (16.0–23.8)	28.1 (23.5–30.1)	38.7 (32.8–48.5)
None	22.1 (17.5–23.2)	20.4 (18.5–25.3)	29.5 (26.4–36.7)	36.0 (32.5–42.6)

* Concentration of penicillin, 0.3 g/liter.

‡ 7–10 mice per group; median and range in parentheses.

§ Body weight was adjusted by subtracting the wet weight of the cecum and its contents.

the three concentrations of drug tested, but the cecal wet weights continued to increase only in mice given the two higher concentrations.

Whereas the total cecal weights increased within a few hours after beginning the drug administration, the dry weights did not increase substantially and consistently until more than 24 hr had elapsed. The initial increase in weight was therefore primarily due to an increase in the amount of water in the cecum. Moreover, although the dry weight had increased after 24 hr, the major factor in the total change was still, at that time, an excess amount of water.

The enlarged cecums were swollen and black in color; their mucosa was under tension as if from pressure from within. When the organs were punctured, the black, fluid contents poured out. In contrast, the contents of normal cecums were greenish-brown in color and very mucoid in consistency. Despite the differences in gross appearance, however, the histology of the enlarged cecums did not differ from that of the normal state. Gross appearance (Fig. 1) and histology (Figs. 2 and 3) made clear that the excess of water in the enlarged cecums 24 hr after the beginning of drug treatment was located primarily in the lumen and not in the wall.

Although histological sections revealed no abnormalities in the mucosal tissue,

TABLE IV

Long-Term Effects of Penicillin in the Drinking Water on the Bacterial Flora of the Cecums of Mice

Drug	Bacteria*	Days after drug started			
		1	2	30	90
Penicillin‡	Lactobacilli	N*	N	N	N
	Coliforms	N	9	10	9
	Enterococci	N	±	10	9
	Anaerobes	N (−)	N (−)	10 (−)	10 (−)
None	Lactobacilli	9	9	9	9
	Coliforms	4	4	6	4
	Enterococci	4	5	5	4
	Anaerobes	10 (+)	10 (+)	10 (+)	10 (+)

* See the footnotes for Table II.

‡ Concentration of penicillin, 0.3 g/liter.

there were remarkable differences in the contents of the cecums. Figs. 2 and 4 show that normal cecums contained huge masses of bacteria and digesta in their lumens. In contrast, hardly any solid material remained on the microscope slides which had received sections of the enlarged cecums of mice given penicillin for 24 hr (Fig. 3).

The almost complete lack of solid material in the lumens agreed well with the results of bacteriological studies. As early as 12 hr after the penicillin was first administered, almost no bacteria could be cultured from the cecums of mice having received the two higher concentrations of the drug (Table II). After 24 hr, the cecums of mice in all treated groups were still essentially free of culturable bacteria. As noted also in Table II, no fusiform bacteria could be seen in histological sections of the cecums from mice receiving penicillin.

Long-Term Effects of Penicillin on Cecal Weights and on Bacterial Flora.—The results of experiments designed to test the effects of continuous long-term administration of penicillin on the cecal wet and dry weights and body weights are summarized in Table III. The findings can be summarized as follows:

(*a*) The continuous administration of penicillin did not markedly affect body weight during the experimental period.[1]

(*b*) The wet weights of the enlarged cecums did not diminish over the 3 month period of continuous administration of the drug.

(*c*) In contrast, the dry weights of the cecums and their contents progressively increased in proportion to the increase in wet weights; the ratio of dry

TABLE V

Effect of Normal Microflora on Wet Weights and Bacterial Populations of Cecums of Mice Given Penicillin for 24 hr

			Before treatment	Days after drug treatment started*				
				1	3	4	6	8
Wet weights of cecums and contents	C‡		0.4§ (0.3–0.5)	0.7 (0.6–1.0)	0.9 (0.7–1.0)	0.6 (0.4–0.9)	0.6 (0.5–0.7)	0.4 (0.3–0.5)
	NC					0.8 (0.6–1.1)	0.8 (0.5–1.0)	0.4 (0.3–0.5)
Bacterial flora	C	Lactobacilli	9‖	N	±	8	9	9
		Coliforms	4	N	10	10	6	4
		Enterococci	4	N	9	9	7	4
		Anaerobes	10(+)	N(−)	10(−)	10(±)	10(+)	10(+)
	NC	Lactobacilli				6	9	9
		Coliforms				5	10	5
		Enterococci				9	9	6
		Anaerobes				10(±)	10(+)	10(+)

* Penicillin solution (0.3 g/liter) was given in the drinking water for 1 day.

‡ After the 1 day of penicillin treatment, all mice were without food from the 2nd to the 3rd day; the C group was given food contaminated with homogenized cecums from normal mice on 3rd and 4th days; the NC group was not given the contaminated food.

§ 7–10 mice per group; median and range in parentheses.

‖ See footnotes to Table II.

weight to wet weight at the end of 30 days of continuous administration of the drug was almost the same as for untreated animals.

After 30 days of continuous administration of the drug, enormous numbers of bacteria could once more be cultured from the cecums, and the histological sections revealed that the lumens were again stuffed with bacteria and digesta (Table IV). However, the cecums remained enlarged as seen in Table III.

The microflora in the cecums of animals receiving penicillin was qualitatively different from that of the controls. No lactobacilli or Group N streptococci

[1] This statement appear in conflict with results reported earlier from this laboratory (9). The two sets of findings, however, are not comparable because the mice used came from colonies having different intestinal flora.

could be cultured from them, and no fusiform bacteria could be seen in histological sections. In fact, their flora consisted primarily of lactose-fermenting coliforms, *Klebsiella-Aerobacter* types, enterococci, and clostridia. These findings are in agreement with the ones earlier reported for the entire gastrointestinal microflora (1).

As evident from the figures presented in Table III, these bacteria present in mice treated with penicillin had not been able to induce a reduction of cecum weight to the normal level.

Administration of Normal Intestinal Flora to Mice Treated with Penicillin.—

TABLE VI

Effect of Normal Microflora on the Weights and Bacterial Populations of Cecums of Mice Given Penicillin for 14 Days

			Before treatment	Days after initiation of drug treatment*						
				15	16	17	19	22	25	30
Wet weights of cecums and contents	C‡		0.4§ (0.3–0.5)	1.4 (1.1–1.7)	0.9 (0.6–1.4)	1.1 (0.6–1.4)	0.6 (0.5–0.7)	0.5 (0.4–0.6)	0.45 (0.4–0.5)	
	NC					1.0 (0.8–1.2)		1.1 (1.0–1.2)	0.7 (0.6–0.8)	0.6 (0.5–0.7)
Bacterial flora	C	Lactobacilli	9‖	N	N	N	±	9	9	
		Coliforms	4	10	10	10	7	4	4	
		Enterococci	4	10	10	10	7	4	4	
		Anaerobes	10(+)	10(−)	10(−)	10(−)	10(±)	10(+)	10(+)	
	NC	Lactobacilli				N		N	N	±
		Coliforms				9		10	4	4
		Enterococci				10		9	4	4
		Anaerobes				10(−)		10(−)	10(±)	10(+)

* Penicillin solution (0.3 g/liter) was given in drinking water for 14 days.

‡ After the 14 days of penicillin treatment all mice were without food from the 15th to the 16th day; the C group was given food contaminated with homogenized cecums from normal mice on the 16th and 17th days; the NC group was not contaminated.

§ 7–10 mice per group; median and range in parentheses.

‖ See footnotes to Table II.

When mice were given penicillin in their drinking water for only 24 hr, the weights and bacterial flora of their cecums spontaneously returned to normal levels within 7 or 8 days after discontinuance of drug administration. This period was not significantly shortened by contaminating their food with material consisting of homogenized cecums from control, untreated mice (Table V).

When mice received the penicillin solution for 14 consecutive days, it took more than 2 wks before the weights and bacterial populations of their cecums returned to normal levels. This period, however, was shortened to 10 days when they were given food contaminated with cecal homogenates from untreated mice (Table VI).

These findings suggest that a minimum period of 8 days is required for correcting the cecal disturbance caused by penicillin, whether this normalizing effect is brought about by the bacteria that have survived in the intestine and start multiplying after very short drug treatment, or by the bacteria introduced in the form of cecal homogenate from untreated mice.

Effects of Broad Spectrum Antibacterial Drugs on Cecal Weight and Bacterial Flora.—As seen in Table VII, oral administration of the broad spectrum antibacterial drugs, Terramycin and kanamycin, brought about an increase in wet weights of the cecums. However, this increase was not as marked as that in-

TABLE VII

Comparative Effects of Penicillin, Terramycin, and Kanamycin Given in Drinking Water on the Wet and Dry Weights of Cecum and its Contents

Drug	Days after drug started		
	1	2	3
	Wet weights of cecum and contents		
Penicillin*	1.00‡ (0.82–1.09)	0.99 (0.93–1.08)	0.93 (0.80–1.21)
Terramycin	0.55 (0.48–0.69)	0.68 (0.59–0.74)	0.67 (0.60–0.75)
Kanamycin	0.63 (0.48–0.75)	0.64 (0.58–0.87)	0.60 (0.36–0.87)
None	0.30 (0.26–0.35)		
	Dry weights of cecum and contents		
Penicillin	0.13 (0.12–0.13)	0.15 (0.14–0.18)	0.15 (0.11–0.16)
Terramycin	0.09 (0.08–0.10)	0.13 (0.11–0.14)	0.17 (0.13–0.18)
Kanamycin	0.10 (0.08–0.11)	0.10 (0.09–0.12)	0.11 (0.07–0.12)
None	0.08 (0.06–0.10)		

* Concentrations of drug per liter of drinking water: penicillin, 0.3 g, Terramycin, 1.7 g, kanamycin, 1.0 g.

‡ 6–10 animals per group; median and range in parentheses.

duced by penicillin. Both Terramycin and kanamycin also induced an increase in cecal dry weight; after a three-day test period, however, only Terramycin proved as effective as penicillin in this respect.

As in the case of penicillin (Table III) no gross physiological disturbances could be detected in the animals receiving the two broad spectrum drugs. Neither the body weights (Table VIII) nor the histology of the cecums were significantly affected during the course of the experiments.

The results of the effects of the broad spectrum drugs on the bacterial flora of the cecums are presented in Table IX. Neither of the drugs was as effective as penicillin in clearing the cecums of their bacterial flora at least during the initial period of treatment. Enterococci and bacteroides could be cultivated from the cecums of animals given Terramycin, and some types of fusiform bacteria could be seen in histological sections (Fig. 5). The cecums of mice given kanamycin yielded cultures of lactobacilli and bacteroides but with this drug, histological sections usually revealed only very small numbers of fusiform bacteria (Fig. 6).

TABLE VIII

Comparative Effects of Penicillin, Terramycin, and Kanamycin Given in the Drinking Water on the Body Weights of Mice*

Drug	Days after drug started		
	1	2	3
Penicillin‡	24.4§ (22.5–26.7)	25.7 (24.7–25.9)	24.4 (21.9–27.7)
Terramycin	25.3 (23.2–26.8)	23.8 (22.6–27.0)	24.2 (21.4–26.3)
Kanamycin	24.5 (23.8–28.2)	24.9 (23.3–28.2)	27.2 (23.8–28.2)
None	24.4 (19.9–28.2)		

* Body weights are adjusted by subtracting the wet weight of the cecum and its contents (see Table III).

‡ See the footnote in Table VII for the concentrations of the drugs.

§ 6–10 animals per group; median and range in parentheses.

As was found in animals given penicillin (Table II and reference 1), both Terramycin and kanamycin induced, in the cecums, the proliferation of characteristic microfloras qualitatively different from those found in control animals.

DISCUSSION

The most spectacular and completely unexpected of the findings reported in the present paper was the marked increase in the total weight and size of the cecum that occurred in mice within a few hours after the oral administration of certain antibacterial drugs. With penicillin, for example, the total weight

TABLE IX

Comparative Effects of Penicillin, Terramycin, and Kanamycin Given in Drinking Water on the Bacterial Flora of the Cecum of Mice

Drug	Bacteria*	Days after drug started		
		1	2	3
Penicillin‡	Lactobacilli	N*	N	N
	Coliforms	±	5	5
	Enterococci	N	±	N
	Anaerobes	N (−)	N (−)	N (−)
Terramycin	Lactobacilli	N	N	±
	Coliforms	N	±	N
	Enterococci	4	8	8
	Anaerobes	9 (+)	10 (+)	10 (+)
Kanamycin	Lactobacilli	6	7	9
	Coliforms	N	N	N
	Enterococci	N	N	N
	Anaerobes	10 (±)	10 (−)	10 (±)
None	Lactobacilli	9		
	Coliforms	4		
	Enterococci	4		
	Anaerobes	10 (+)		

* See the footnotes for Table II for a description of the bacteria involved, when penicillin is administered, and for method of recording the data.

When Terramycin and kanamycin are administered, the culturable anaerobes are predominantly bacteroides until the 3rd day, when clostridia appear as the dominant type. Thereafter, as long as the drug is continuously administered, the clostridia predominate.

‡ See footnote to Table VII for the concentrations of the antibiotics.

of the cecum, including its contents, had increased 2- or 3-fold within 12 hr after the oral administration of even small amounts of the drug. The increase in weight was due chiefly, if not exclusively, to the accumulation of water in the lumen.

There was no detectable evidence of histological abnormality in the epithelium during the early phase of cecal enlargement. In contrast, there were profound changes in the microflora of both the mucous layer and the lumen; bacteriological and histological techniques agreed in indicating that the bacteriological flora had been virtually eliminated within 12 hr after the beginning of penicillin treatment.

The results with Terramycin and kanamycin were similar to those obtained with penicillin but not as spectacular. One difference that may help in the interpretation of the findings was that certain types of bacteria survived longer in the cecum of mice treated with Terramycin and kanamycin than they did with penicillin; this was particularly true for the bacteroides and fusiform groups. The fusiform bacteria are of particular interest in this regard because recent studies have revealed that they constitute by very far the largest bacterial population in the normal cecum and are imbedded in the mucous layer of its epithelium (8).

When the animals were maintained on the drugs for prolonged periods of time, the cecum continued to enlarge and its epithelium eventually exhibited some histological abnormalities. The most striking changes, however, occurred in the cecal microflora. This flora did not return to its normal pretreatment state as long as drug administration was continued; instead, there was an enormous proliferation of enterococci, clostridia, and lactose-fermenting coliforms. As this new bacterial flora became established, the dry weight of the lumen content naturally increased, but there was no change back to normal in the size of the cecum, although the color of its contents usually became more green-brown.

The cecal enlargement produced by the drugs was long lasting. Even when penicillin was administered orally for only 24 hr, it took 7–8 days for the cecal size and wet weight to return to normal levels. The bacterial population returned to its normal pretreatment state, both qualitatively and quantitatively, after about the same length of time.

The alterations in size, weight, and contents of the cecum that have been observed indicate that administration of antibacterial drugs can produce profound disturbances of intestinal physiology. These disturbances should be studied, of course, by physiological techniques, but some bacteriological aspects of the problem are worth emphasizing here because they may be directly relevant to the functional disturbances in the cecum in drug-treated mice.

The physiological disturbances in the cecum occur long before histological changes in the epithelium can be recognized, but they are more or less concomitant with the elimination of certain types of bacteria, especially those of the fusiform group. Disturbances are more rapid and more pronounced with penicillin, a drug which can eliminate certain strictly anaerobic components of the intestinal flora within a few hours, than with Terramycin or kanamycin which are less effective in this regard. Restoration of the cecum to its normal anatomical and physiological state can be achieved by reassociating the drug-treated animal with the total intestinal flora of normal mice; however, not just any kind of bacteria can effect this restoration since anatomical and physiological abnormalities persisted after immense numbers of enterococci, clostridia, and lactose-fermenting coliforms had become established throughout the intestine during drug therapy.

The observations reported in this paper present analogies with those made in germfree rodents. In these animals, the cecum is extremely enlarged and contains large amounts of fluid. These abnormalities can be corrected by reassociating the germfree mice with the intestinal microflora of normal mice; in unpublished experiments from our laboratory, it has been found that, as in the case of drug-treated mice, not just any kind of bacteria are able to produce this effect in germfree mice.

Taken together, the observations reported in this paper and those made in germfree animals suggest that some particular types of bacteria play an essential role in maintaining the integrity of the water-transport mechanism in the intestinal epithelium. Fusiform bacteria are among the likely candidates for this function, since they are immensely numerous in the cecum and are imbedded in the mucus layer of its epithelium (8). Unfortunately, this hypothesis cannot yet be demonstrated convincingly for lack of bacteriological techniques to identify, enumerate, and cultivate in vitro some of the fusiform and other types of intestinal anaerobes.

SUMMARY

Addition of penicillin, Terramycin, or kanamycin to the drinking water of adult mice rapidly induced in them an enlargement of the cecum. In all animals, this occurred within 12 hr after the beginning of drug administration—the effect being most pronounced with penicillin. The cecums remained enlarged and generally continued to increase in size as long as the antibacterial drugs were administered.

The increase in wet weight of the cecums was due primarily to an accumula-

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FIG. 1. Left, cecum, approximate weight 0.5 g, of a normal NCS-D mouse; right, cecum, approximate weight 1.2 g, of an NCS-D mouse given drinking water containing 0.3 g of penicillin per liter continuously for 24 hr.

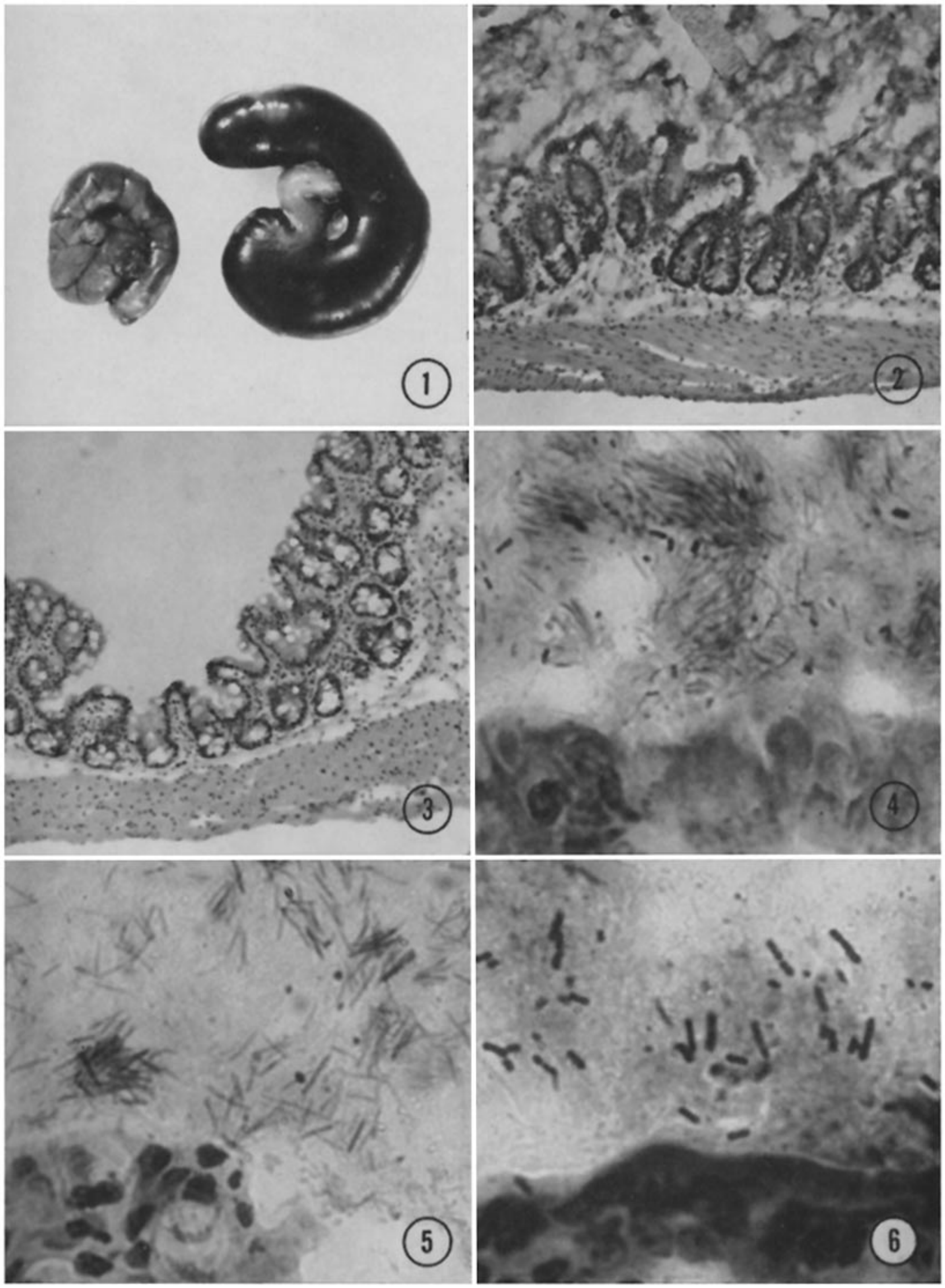
FIG. 2. Gram-stained histological section of the cecum of a normal NCS-D mouse; the lumen containing bacteria and digesta occupies the upper one-half of the photograph. × 100.

FIG. 3. Gram-stained histological section of the cecum of an NCS-D mouse given drinking water containing 0.3 g of penicillin per liter continuously for 24 hr; the lumen is the empty space in the upper left hand corner. × 100.

FIG. 4. Same as Fig. 2; a higher magnification showing mixed types of bacteria but particularly Gram-variable fusiform rods in the mucus on the epithelium (shown at the bottom). × 1000.

FIG. 5. Gram-stained histological section of the cecum of an NCS-D mouse given drinking water containing 1.7 g of Terramycin per liter continuously for 24 hr; the lumen at the top of the photograph contains mixed types of Gram-variable fusiform bacteria. × 1000.

FIG. 6. Gram-stained histological sections of the cecum of an NCS-D mouse given drinking water containing 1.0 g of kanamycin per liter continuously for 24 hr; the lumen at the top of the photograph contains Gram-positive rods and cocci and a few small Gram-negative rods. × 1000.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tion of water in the lumens during the first 24–48 hr of drug administration. At that time, there were no detectable histological changes in any case, but the bacteriological picture differed from drug to drug. The cecums were free of bacteria in animals receiving penicillin, fusiform-shaped bacteria and bacteroides were present in those receiving Terramycin, and lactobacilli and bacteroides in those receiving kanamycin. After the initial 48 hr, an abundant and complex secondary microflora developed in all treated animals, its composition being characteristic for each type of antibacterial drug.

When penicillin was administered for 2 wk, the cecal weights and microbial populations did not return to normal levels for over 14 days after discontinuance of the drug. This recovery period could be shortened to 10 days by giving the mice food contaminated with cecal homogenates prepared from normal animals. A period of 7 or 8 days was required for the cecal weights and microflora to reach normal levels when the administration of penicillin lasted only 24 hr; this period could not be shortened by giving the animals contaminated food.

The effects of drugs on the size and bacterial contents of the cecum have been discussed in the light of earlier findings concerning the characteristics of the huge cecums uniformly found in germfree mice. Taken together, these observations support the hypothesis that certain elements of the intestinal microflora—not yet completely identified—play an essential role in maintaining the integrity of the water-transport mechanism in the intestinal epithelium.

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