et al.[14] Venning, *et al.*[3] have commented on this toxic effect found in extracts of human urine. Fractionation evidently removes some of the toxic activity since glycogen responses are higher in the ketonic fraction than in the total extracts. As was to be expected no evidence of cortin-like activity was found in the non-ketonic fractions.

As a standard preparation for comparison, Wilson's aqueous adrenal cortical extract has been used. Comparing liver glycogen responses to Wilson's extract with the average response of the ketonic fraction of the horse urine extracts it was calculated that 1 liter of normal horse urine contains an equivalent of 0.10-0.15 cc of Wilson's extract and 1 liter of pregnant mare urine contains an equivalent of 0.30-0.40 cc of this extract. These figures compare favorably with results of Dorfman *et al.*[15] and Venning *et al.*[3] on human urine extracts but appear to be definitely lower than the results reported for horse urine by Selye and Dosne.[8] The quantitative assay of the concentration of cortin-like material in urine is complicated by so many factors that the results presented here should be considered only as estimations.

Summary. The presence of cortin-like activity in extracts of horse urine has been confirmed. It is estimated that the concentration of this active material is approximately the same in urine of normal horses and normal humans. There is some indication that urinary cortin-like activity is increased in pregnancy.

14 Dorfman, R. I., Shipley, R. A., Schiller, S., and Horwitt, B. N., *Endocrinology*, 1946, **38**, 165.

15 Dorfman, R. I., Horwitt, B. N., and Fish, W. R., *Science*, 1942, **96**, 496.

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Nutrition of the Mouse. IV. Comparison of Bacterial Population of Two Highly Inbred Strains.*

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It was shown by Rogers, McElroy and Cowgill[1] that the nutritional requirements for reproduction and lactation differed for 2 strains of mice. Fenton and Cowgill[2] showed the existence of a strain difference with respect to the riboflavin requirement for growth. Unpublished work in this laboratory has shown that this is also true of the pantothenic acid requirement. This dissimilarity of the nutritional requirements of 2 strains of mice may in part be due to different rates of intestinal synthesis of essential nutrients. The cecum has been thought to be an important site of the synthesis of B vitamins.[3,4,5,6] Man-

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2 Fenton, P. F., and Cowgill, G. R., *J. Nutrition*, 1947, **34**, 273.

3 Guerrant, N. B., Dutcher, R. A., and Tomey, L. F., *J. Biol. Chem.*, 1935, **110**, 233.

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5 Schweigert, B. S., McIntire, J. M., Henderson, L. M., and Elvehjem, C. A., *Arch. Biochem.*, 1945, **6**, 403.

6 Mannering, G. J., Orsini, D., and Elvehjem, C. A., *J. Nutrition*, 1944, **28**, 141.

TABLE I.
The Weight of the Cecal Contents and the Bacterial Count per Gram of Cecal Contents and per Cecum of the 2 Strains of Mice.

Strain	Diet	No. of animals	Wt of cecal contents, g	Bacterial count	
				per cecum, billions	per g of cecal contents, billions
A strain	101	12	.065 ± .021*	1.5 ± .84	22.2 ± 11.5
C_{57} strain	101	12	.176 ± .047	5.11 ± 2.62	28.7 ± 11.5
Young A strain	stock	3	.386 ± .038	13.12 ± 3.03	32.4 ± 4.34
Young C_{57} strain	"	3	.352 ± .070	8.39 ± 1.76	23.9 ± 2.1
Older A strain	"	6	.417 ± .045	19.86 ± 6.1	48.0 ± 16.8
Older C_{57} strain	"	3	.406 ± .125	15.1 ± 5.9	37.0 ± 13.4

* Standard deviation.

nering *et al.*[6] worked with rats fed synthetic diets containing various carbohydrates and state that with certain exceptions the riboflavin found in the feces was related to the dry weight of the cecal contents.

The purpose of this investigation was to study the intestinal flora of 2 strains of mice, one with characteristically high susceptibility to spontaneous mammary tumors (strain A) and the other with low susceptibility in this respect (strain C_{57}). These two strains were chosen because they had been used in previous studies in this laboratory.

Experimental. The 39 mice used in this study had been maintained on one of 2 diets: a synthetic ration containing dextrose as the carbohydrate without added biotin or folic acid (diet 101 as described previously[7]) or a stock ration.† Twelve mice of the C_{57} strain (tumor-resistant) and 12 of the A strain (tumor-susceptible) ranging in age from 2 to 4 months were used after having been maintained on the synthetic ration. The remainder of the animals had been fed the stock diet and fell into 2 age groups, namely, 2-2.5 months and about 12 months old. The mice were sacrificed by etherization and the cecal contents removed and weighed. Bacterial counts and cultural work were carried out as described in another publication.[8]

Results. As shown in Table I, the mice of the A strain on the synthetic diet had smaller cecal contents than the C_{57} mice on the same diet. In only one case did the highest value given by an A strain mouse equal the lowest weight obtained from a C_{57} animal. The differences in weights of cecal contents between these 2 groups was found to be statistically highly significant ($p < 0.01$). The total bacterial count per g of cecal contents was about the same in both strains. However, the effect of the larger cecal contents of the black mice was reflected in the greater total bacterial population of the cecum of this strain when compared to the white animals. This difference was also found to be statistically highly significant ($p < 0.01$).

The results obtained by studying the Gram stains and the cultures isolated from the ceca of both strains were similar with respect to the occurrence of cocci and Gram-positive rods. However, there appeared in the highest dilutions of the C_{57} mice in 10 cases out of 12 a short Gram-negative, anaerobic rod. This type of organism occurred in only 2 white mice out of 12. Coliform organisms were encountered slightly more frequently in high dilutions of cecal contents of the A strain mice (5 out of 12 animals) than in the C_{57} strain (2 out of 12 mice).

These two strains of mice, when fed a stock ration, showed no differences in weights of cecal contents and total bacterial population per cecum. The lower values shown by C_{57} mice for bacterial counts per g of cecal contents and per cecum were not considered significant owing to the wide variation in counts and to the small number of animals in each series. On microscopic examination of the

[7] Fenton, P. F., and Cowgill, G. R., *J. Nutrition*, 1947, **33**, 703.

† Purina Laboratory Chow.

[8] Gall, L. S., Fenton, P. F., and Cowgill, G. R., *J. Nutrition*, in press.

Gram stain and from cultural results the bacterial flora of the cecum of the older animals of both strains seemed to be similar, but there was a tendency for the younger black animals (C_{57}) to have a more varied flora than was shown by the white mice of the same age. It is unsafe to draw a general conclusion from so small a series of mice, even though this difference was observed on all animals.

In Table I the values for the stock ration animals are broken down into groups of young and older mice, so that these stock animals may be compared with those on diet 101. As was pointed out in a previous paper by these authors,[8] the weight of the cecal contents and the bacterial count per g and per cecum were considerably higher in the A strain mice on the stock diet than in the A strain mice fed a synthetic ration. By inspection of the data in Table I, it is evident that even though the black animals fed a synthetic diet had greater cecal contents and higher bacterial counts per cecum than the A strain mice, they still fell short in these respects of the C_{57} animals of the same age on the stock diet.

Discussion. There has been no really satisfactory explanation offered for the strain differences in requirement for certain of the B vitamins, as manifested by mice of the C_{57} and A strains. (Grüneberg[9] discussed the various anatomical and physiological differences in the alimentary tracts of mice of several strains, but there is no mention made of any variations in the amount of cecal contents, or in the size of the cecum. It is therefore of interest to note that in our study a significant difference in the weight of the cecal contents of 2 strains of mice was found when these animals were fed a synthetic diet, especially since the A strain mice, which had the smaller cecal contents also had a higher dietary requirement for certain B vitamins. It has been shown[3,4,5] that the cecum is one of the sites of bacterial synthesis of B vitamins in the intestinal tracts of rats. For this reason it may well be that the larger bacterial population offers an explanation for the lower requirements of the C_{57} strain for certain B vitamins. Further support for this theory is found in the cultural work done on the cecal contents of animals fed this synthetic diet in which it was demonstrated that bacteria isolated from top dilutions of these contents in numbers of 100 million or more per gram were able to synthesize some of the B vitamins *in vitro*.[10]

Summary and Conclusions. 1. Studies were made to determine whether there is a difference in the weight and bacterial population of the cecal contents of mice of the C_{57} and A strains, which were the same age and received the same diet.

2. The C_{57} animals fed a synthetic diet (No. 101) had a significantly greater amount of cecal contents and bacterial count per cecum than the A strain mice of the same age and fed the same diet. Although most of the bacterial flora appeared to be the same in the 2 strains, a Gram-negative, anaerobic rod did occur more frequently in the C_{57} mice than in the A strain.

3. There was no significant difference between the black and white mice of the same age fed the stock ration with respect to weight of cecal contents or bacterial count per cecum. The young black mice may have had a more diverse flora than the white mice of the same age.

4. It was postulated that this greater weight of cecal contents with its higher bacterial population in the C_{57} mice might help to explain the lower B vitamin requirements of this strain when comparing it with animals of the A strain, both fed the same synthetic diet.

[9] Grüneberg, Hans, *Genetics of the Mouse*, 1943, Cambridge University Press, London, England.

[10] Gall, L. S., Illingworth, B. A., Cowgill, G. R., and Fenton, P. F., *J. Nutrition*, in press.