TABLE I.
Effect of Anoxia on Peristalsis of the Small Intestine in Rats.

PO_2 mm Hg	Altitude, feet	Control		Anoxia		"P"*
		No. of animals	% of gut traversed at end of 40 min.	No. of animals	% of gut traversed at end of 40 min.	
94	14,000	11	71	11	64	0.19
80	18,000	9	82	11	64	<0.01
63	24,000	10	76	10	50	<0.001
53	28,000	12	78	10	40	<0.001

* Significant when "P" (according to Fisher) is 0.05 or less.

mental animals.

Results and Discussion. The data, Table I, show that anoxic anoxia did not decrease the propulsive motility of the small intestine significantly at a simulated altitude of 14,000 feet. Above this level, however, the motility was significantly decreased by the anoxia.

The results indicated that the threshold for anoxia on the peristalsis of the small intestine lay between a simulated altitude of 14,000 feet and 18,000 feet. In previous work[1] it was shown that the propulsive motility of the intestine of the mouse was not significantly decreased until a simulated altitude of 18,000 feet was reached. The results obtained with rats, therefore, show fair agreement with those for mice. Since anoxic anoxia stimulates the sympathetic division of the autonomic nervous system it would be expected that intestinal movements would be decreased.

The authors still can offer no adequate explanation for the negative effect of anoxic anoxia on the propulsive motility of the small intestine in the dog. The current experiments suggest that as far as the propulsive motility of the small intestine is concerned the rat is somewhat less resistant to anoxia than the dog. However, it should be pointed out that both of these animals are relatively resistant to anoxic anoxia so that the difference observed by us between the two species probably cannot be attributed entirely to this factor.

Summary. The effect of anoxic anoxia on the propulsive motility of the small intestine of the albino rat was studied at the following partial pressures of oxygen: 94 mm Hg, 80 mm Hg, 63 mm Hg, and 53 mm Hg; corresponding to simulated altitudes of 14,000, 18,000, 24,000, and 28,000 feet. At a simulated altitude of 18,000 feet or higher there was a significant decrease in propulsive motility. The threshold lay between 14,000 and 18,000 feet. The results differ from those obtained in the dog but show fair agreement with those in the mouse.

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Protein Level and Pteroylglutamic Acid Intake on Growth Rate and Hemoglobin Level in the Rat.

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In attempts to produce a pteroylglutamic acid (PGA) deficiency in the pig, baby pigs were fed a PGA-low "synthetic" diet containing 30% casein and 2% sulfathalidine. However, no blood dyscrasias were observed.[1] Kornberg, Daft and Sebrell[2] and Daft[3] have shown that limitation of the casein in the

1 Johnson, B., Connor, James, Marian, F., and Krider, J. L., *J. Animal Science*, 1947, **6**, 486.

2 Kornberg, Arthur, Daft, Floyd S., and Sebrell, W. H., *Science*, 1946, **103**, 646.

3 Daft, Floyd S., *Fed. Proc.*, 1947, **6**, 405.

TABLE I.
Ration Fed to Each Group of Rats.*

Ingredient	Groups 1	2	3	4	5	6
Casein, Labco	5	5	10	15	20	30
PGA 0.4 mg/100 g	0	0	0	0	0	0
Glucose	83	83	78	73	68	58
Cod Liver Oil	2	2	2	2	2	2
Salt Mixture 446	4	4	4	4	4	4
Corn Oil	4	4	4	4	4	4
Sulfathalidine	2	2	2	2	2	2

* The following vitamins were added per 100 g basal ration:

	mg
Thiamine	.25
Riboflavin	.50
Pyridoxine	.25
Nicotinic Acid	1.0
Calcium-pantothenate	2.0
Biotin	.01
p-Aminobenzoic Acid	5.0
Inositol	10.0
Choline	100.0
2-methyl-1,4-naphthoquinone	0.1
Vit. A, Vit. D	Haliver oil, 2 drops/rat/week
α-Tocopherol	1 mg/rat/week

diet of the rat is a significant factor in the production of the blood dyscrasias typical of PGA deficiency. In order to determine what level of protein might be fed and still produce anemia on a PGA-deficient ration containing sulfathalidine, the influence of different casein levels and the effect of PGA intake on anemia production in growing rats were investigated.

Experimental. Six groups of weanling rats, 5 in each group, were fed the rations given in Table I. The rats were fed *ad libitum* and kept on experiment for six weeks. Their growth curves are given in Fig. 1. Starting with the third week on the experiment, blood hemoglobin levels and red blood cell counts were determined weekly for the next 4 weeks. The hemoglobin values are given in Fig. 2. The red blood cell counts at 6 weeks are given in Table II.

Discussion. From this experiment it appears that the anemia produced by a protein deficiency on a sulfathalidine-containing PGA-low ration will respond to either PGA or to protein (Fig. 1 and Table IV). On the other hand, the lack of growth on a 5% casein diet was not affected by the addition of PGA to this diet (Fig. 2 and Table III).

The differences between the average gains of the rats for each group were analyzed statistically and no difference was found be-

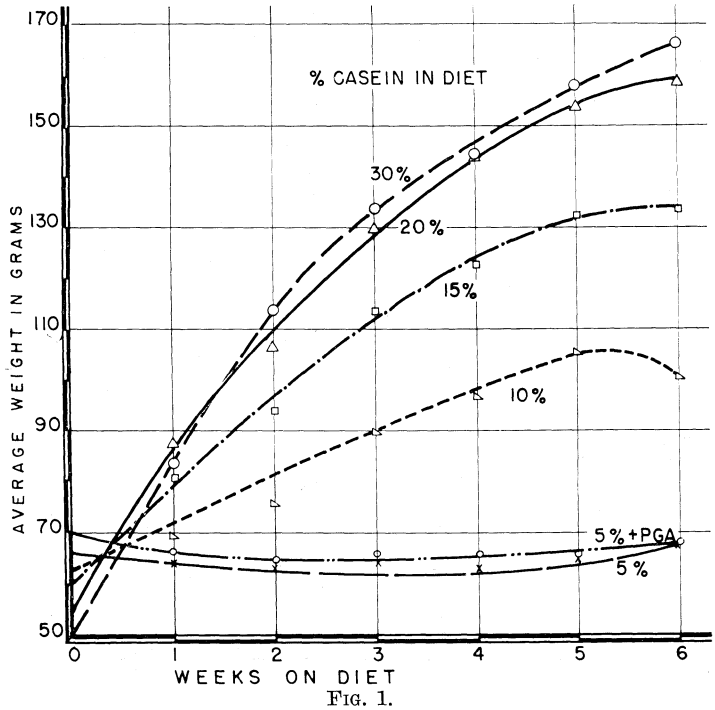

FIG. 1.
Growth curves of experimental rats.

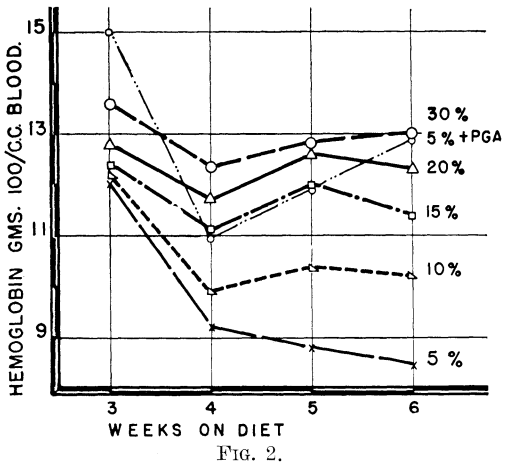

FIG. 2.

Hemoglobin concentration of blood from experimental rats.

TABLE II.

Average Red Blood Cell Count at 6 Weeks for Groups of Rats Fed Increasing Levels of Casein.

Group No.	%	Ration	Avg RBC millions per mm³
1	5	Casein + PGA	7.16
2	5	Casein	5.11
3	10	"	5.95
4	15	"	6.22
5	20	"	6.60
6	30	"	7.15

tween those receiving 20 and 30% protein ($P = 0.45$), but significantly lower rates of gain were found at lower casein levels (30% or 20% versus any other, $P < .01$). The statistical analysis of this data is given in Table III.

Statistical analysis of the data on concentration of hemoglobin in the blood at the end of six weeks, given in Table IV indicated: (1) that the presence of either PGA or of 15, 20, or 30% casein in the diet produced a significantly greater concentration of hemoglobin in the blood than did a 5% casein diet; (2) that diets containing 5% casein plus PGA or 20 or 30% casein produced equal hemoglobin concentrations; and (3) that without PGA, 10% casein produced no significantly greater hemoglobin concentration than did 5% casein. This would indicate that for normal hemoglobin formation in the absence of PGA, over 15% casein is required in the diet, that is, between 15 and 20% casein provides adequate precursors for the synthesis of PGA.

It is also to be noted that even with 2% sulfathalidine in the diet, no significant anemia was observed in the absence of PGA on a 20% casein diet. Several workers[5–9] have produced typical symptoms of PGA deficiency using 18% casein diets with various sulfa drugs used as the intestinal bacteriostatic agents. Our inability to produce blood discrasias at 20% casein may be due to differences in the PGA content of Labco casein, to the inclusion

TABLE III.

Statistical Treatment of Data. Gains of Rats at 6 Weeks (g).

Group No.	1 5% casein + PGA	2 5% casein	3 10% casein	4 15% casein	5 20% casein	6 30% casein
Rat 1	1	8	32	74	126	125
2	—3	2	42	92	95	138
3	—3	1	34	74	108	111
4	—4	—3	52	55	105	104
5	dead	—1	31	82	95	108
Mean	—2.2	1.4	38.2	75.4	105.8	117.2
Rations compared		1 vs. 2	2 vs. 3	3 vs. 4	4 vs. 5	5 vs. 6
Probability of Difference*		.45	<.01	<.01	<.01	.45
		1 vs. 3	2 vs. 4	3 vs. 5	4 vs. 6	
		<.01	<.01	<.01	<.01	
		1 vs. 4	2 vs. 5	3 vs. 6		
		<.01	<.01	<.01		
		1 vs. 5	2 vs. 6			
		<.01	<.01			
		1 vs. 6				
		<.01				

* Student.[4]

TABLE IV.
Statistical Treatment of Data. Hemoglobin Levels in 100 g/100 cc of Rats at 6 Weeks.

Group No.	1 5% + PAGA casein	2 5% casein	3 10% casein	4 15% casein	5 20% casein	6 30% casein
Rat 2	13.1	8.0	8.3	12.0	12.0	12.4
3	12.4	8.3	11.0	10.7	11.2	14.2
4	13.8	8.9	11.2	10.6	12.4	13.1
5	12.7	8.9	11.0	12.4	13.8	12.4
Mean	13.0	8.5	10.4	11.4	12.4	13.0
Group compared			1 vs. 2	2 vs. 3	3 vs. 4	4 vs. 5	5 vs. 6
Probability of Difference*			<.01	.025	.15	.12	.2
			1 vs. 3	2 vs. 4	3 vs. 5	4 vs. 6	
			<.01	<.01	.025	.025	
			1 vs. 4	2 vs. 5	3 vs. 6		
			.01	<.01	.01		
			1 vs. 5	2 vs. 6			
			.15	<.01			
			1 vs. 6				
			.028				

* Student.[4]

of a more complete list of vitamins (including para-aminobenzoic acid, inositol, etc.) in our ration, to differences in carbohydrate, etc.; or it may be that sulfathalidine is not as useful for prevention of intestinal synthesis of PGA in nutritional experiments as are some of the other insoluble sulfa drugs. However, Teply, Krehl, and Elvehjem[10] have produced growth depression on a 15% casein "synthetic" diet containing 78% sucrose and 2% sulfathalidine, which was partially corrected by feeding a "folic acid" concentrate. Wright, Skeggs, and Sprague,[11] using diets containing 5% sulfasuxidine, have shown that on a high protein (30% casein) diet, "folic acid" stores in the liver and "folic acid" excretion in the feces are higher than on a normal (18% casein) diet. They also obtained better growth on the high protein diet, due to the lower PGA requirement.

Summary. Anemia was produced in growing rats by feeding a ration containing 5% casein and 2% sulfathalidine, to which no pteroylglutamic acid was added. This anemia was prevented either by the addition of pteroylglutamic acid to the diet or by increasing the casein to 20%.

There was no improvement in growth when PGA was added to the 5% casein diet, while 30% casein gave normal growth.

The sulfathalidine used in this experiment was very generously supplied by Sharp and Dohme, Philadelphia, Pa., through the courtesy of Dr. S. F. Scheidy.

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