the tissues. Moreover, it is also possible that when the serine level is suddenly elevated, the formation of abnormal products through side reactions takes place to a large extent.‡

‡ Thus, ethanolamine, which in sufficient amounts is toxic for rats,[2] could possibly be formed by decarboxylation of serine. In this regard, the action of the intestinal flora should of course be considered. However, since serine by parenteral injection, as well as by stomach tube, is highly injurious, a role of the intestinal bacteria in the mechanism of serine injury appears unlikely.

It should be pointed out that these interpretations may apply to one or both of the components of the racemic *dl*-serine.

Summary. *dl*-Serine in 100 mg amounts daily is injurious to rats on an experimental diet when supplied by stomach tube and by parenteral injection, but is not harmful when administered mixed in the diet. Some hypotheses on the mechanism of the production of the toxic action of serine are discussed.

[2] Artom, C., and Fishman, W. H., *J. Biol. Chem.*, 1943, **148**, 423.

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Some Dietary Factors Which Reduce the Toxicity of *dl*-Serine in Rats.*

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Previous data[1] point to a relationship between the diet and the severity of the injury, caused by administration of *dl*-serine. Rats on a stock diet, receiving the amino acid by stomach tube, exhibited a transient cessation of growth, and only slight ill-effects; no fatalities were observed. This contrasted with the high mortality observed under the same conditions in animals maintained on both experimental diets 1 and 2. One of these diets (Diet 2, with 30% casein) provided a very satisfactory rate of growth, almost identical with that of rats on the stock diet. It seems, therefore, that the protective action of the stock diet against serine toxicity was not due merely to the maintenance of a state of good nutrition, but rather to some specific factor, not present in sufficient amount in the experimental diets.

In this connection, casein diets are known to be relatively poor in choline, cystine, and glycine. On the other hand, the role of a deficiency of some members of the vitamin B complex could also be suspected, as the amounts of these vitamins present in our experimental diets were probably inadequate.† Accordingly, in the present study, the effect of the administration of a choline, cystine, glycine mixture, pure B vitamins, or both simultaneously, has been investigated. Special attention has been directed to the action of pyridoxine.‡

Experimental. Two experimental diets were used, diet 4[3] and diet 4 supplemented with choline HCl, glycine, and *l*-cystine (50 mg of each, daily). The daily intake of vitamins in animals on the stock diet and in animals receiving pure B vitamins is indicated in Table I. *dl*-Serine was administered by stom-

* This work was aided by a grant from the John and Mary R. Markle Foundation. Preliminary report in *Federation Proceedings*, March, 1944, **3**, 10.

[1] Fishman, W. H., and Artom, C., *J. Biol. Chem.*, 1942, **145**, 345.

† 10 g of diets 1 and 2[1] contained 100 γ nicotinic acid, 10 γ riboflavin, 10 γ thiamin HCl, 1 γ pyridoxine, 1 γ Ca pantothenate.

‡ We are indebted to Dr. E. E. Snell for having pointed out to us the possibility of pyridoxine having an action antagonistic to serine, on the basis of his then unpublished results.[2]

[2] Snell, E. E., and Guirard, B. M., *Proc. Nat. Acad. Science*, 1943, **29**, 66.

[3] Artom, C., and Fishman, W. H., PROC. SOC. EXP. BIOL. AND MED., 1944, **57**, 239.

TABLE I.
Approximate Daily Intake of Individual B-vitamins.

B-vitamins	B-vitamin sol. (complete)* γ	Stock diet† γ
Pyridoxine	50	23-47
Thiamine HCl	50	54-65
Riboflavin	50	21-28
Nicotinic acid	1500	1440
Calcium Pantothenate	200	200-257
Inositol	200	Not determined
p-Aminobenzoic Acid	200	Not determined

* The pure vitamins were generously supplied by Merck and Co. The vitamin solutions were injected in a volume of 0.5 cc once daily for the first 3 weeks of experiment.

† An average daily intake of 10 g of diet was assumed in calculating these values. Data on the B-vitamin content of our stock diet (Rockland Rat diet, complete) were supplied by Arcady Farms Milling Company, Chicago, Ill.

ach tube according to the general plan of experimentation previously described.[3]

Results. Data on mortality are recorded in Table II, while composite curves of changes in body weight for most of the experimental groups are shown in Fig. 1 and 2. Separate curves are given for the animals dying (Curve VIa) and for those surviving (Curve VIb).

On the stock diet (Group I) no deaths occurred from serine administration and growth was affected only for a few days. In the animals on the experimental diets receiving serine but no B vitamins, mortality was high and the loss in weight was marked, even though supplements of choline, cystine, and glycine were given (Group II). When serine and the complete mixture of B vitamins were supplied (Group IV and V),

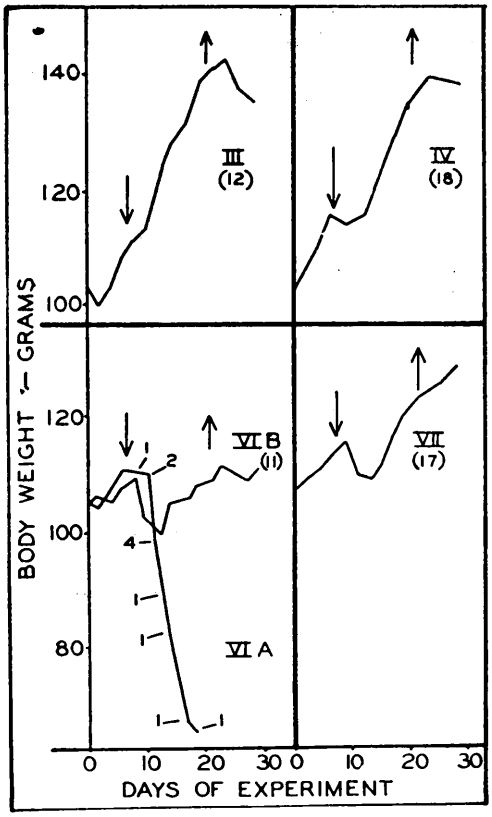

FIG. 1.

The action of serine on the body weight of rats receiving B vitamins. For the periods between the arrows the rats received by stomach tube either water (III) or *dl*-serine (other groups). All rats were on diet 4. Group III and IV received the complete B vitamin mixture; group VI, the vitamin mixture minus pyridoxine; group VII, pyridoxine only. The number of rats surviving in each group is indicated in parenthesis. For group VI the number of deaths in each day of experiment is shown on curve VIA.

TABLE II.
Effect of Dietary Factors on Mortality of Rats Receiving *dl*-Serine (100 mg daily).

Group No.	Diet	Vitamins inj.	Administered by stomach tube	No. of rats	No. of deaths	Mortality %
I	Stock	None	*dl*-serine	19	0	0
*	4	"	H_2O	12	0	0
*	4	"	*dl*-serine	21	13	62
II	4 + choline HCl, *l*-cystine, glycine	"	"	19	10	53
III	4	B complete	H_2O	12	0	0
IV	4	"	*dl*-serine	19	1	5
V	4 + choline HCl, *l*-cystine, glycine	"	"	19	1	5
VI	4	B minus Pyridoxine	"	22	11	50
VII	4	Pyridoxine (100 γ per day)	"	19	2	11

* See Groups I and II of our previous series of experiments.[3]

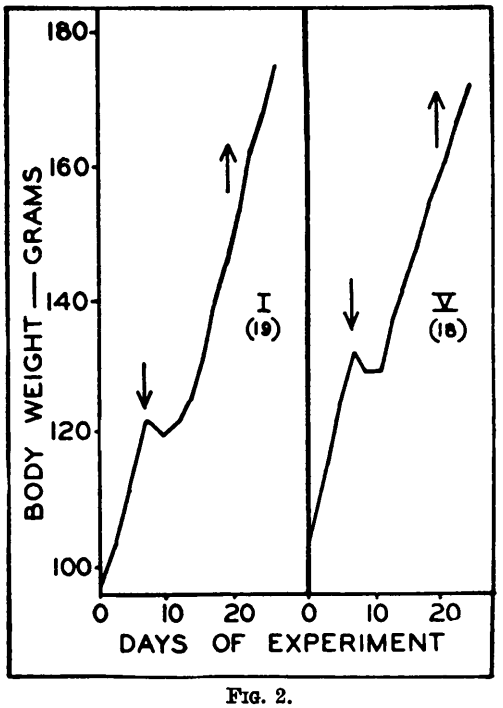

FIG. 2.

A comparison between the action of serine on the weight of rats on stock diet (I), or on diet 4, supplemented with choline, cystine, glycine and the complete B vitamin mixture (V). The number in parentheses indicates the number of rats surviving.

the mortality was very low. Growth was only slightly affected by serine administration and the animals appeared to be in good health as did those on the stock diet. Group V, on the experimental diet, supplemented with choline, cystine, and glycine and all the B vitamins, appeared to withstand the ill effects of serine best, the growth curves of these rats being almost identical to that of the animals on stock diet receiving serine (Fig. 2).

On the other hand, when pyridoxine was omitted from the B vitamin mixture (Group VI), the administration of serine produced severe clinical disturbances and many deaths resulted. The survivors did show recovery of growth in a manner very similar to that of corresponding animals receiving serine but no B vitamins.[3] Pyridoxine alone (Group VII) was able to reduce the mortality considerably; the drop in weight was less marked and the recovery more rapid than that of Group VI, receiving B vitamins without pyridoxine.

Discussion. It appears that a mixture of pure B vitamins is capable of alleviating the severity of clinical symptoms and of reducing considerably the mortality of rats receiving serine by stomach tube. As the stock diet contains adequate amounts of these vitamins, the protective action of this diet against serine injury may thus be explained. Of course, the possibility that other factors may also be involved cannot be excluded.

Of the B vitamins we have tested, pyridoxine appears to be most effective. It is difficult to state at present whether the action of this vitamin is a more general one on the nutrition of the tissues, or whether the vitamin is involved specifically in some phases of the metabolism of serine. However, pyridoxine has been shown to reverse the inhibition of growth in *Strep. lactis* caused by certain amino acids including serine.[2] In this respect, it may also be pointed out that a relationship between pyridoxine and the metabolism of proteins[4] and of tryptophane[5] has been shown previously.

Summary. The administration of ample amounts of pure B vitamins to rats on an experimental diet, receiving *dl*-serine by stomach tube, reduces considerably the mortality and the severity of the clinical symptoms. Of the B vitamins tested, pyridoxine was most effective.

[4] Lepkowsky, S., Roboz, E., and Haagen-Smit, A. J., *J. Biol. Chem.*, 1943, **149**, 195.

[5] McHenry, E. W., and Gavin, G., *J. Biol. Chem.*, 1941, **138**, 471; Foy, J. R., and Cerecedo, L. R., Abstracts, Division of Biological Chemistry, American Chemical Society, Atlantic City, Sept. 8, 1941; Voris, L., and Moore, H. P., *J. Nutrition*, 1943, **25**, 7.