deficiency symptoms or to accelerate death. When fed to rats receiving 25 γ and 100 γ of calcium pantothenate respectively, it failed to depress the weight curve or to precipitate symptoms of deficiency.

Discussion. The data presented demonstrate that feeding of pantoyltaurine to rats maintained on a diet free from pantothenic acid does not affect the growth nor the survival time. Daily administration of 1 and 25 mg of pantoyltaurine respectively to rats receiving graded doses of pantothenic acid did not influence significantly the growth of these animals. Our data, therefore, do not adduce evidence of an interference with the utilization of pantothenic acid by pantoyltaurine in animals.

While these experiments were in progress, two reports appeared on the effect of pantoyltaurine in mice. Snell *et al.*[5] reported that mice maintained on Purina Fox Chow and fed daily 200 mg per kg of pantoyltaurine showed after 3 weeks rough fur and porphyrin deposits on the whiskers. However, no attempt was made to correct these manifestations which were interpreted as symptoms of pantothenic acid deficiency, by the addition of pantothenic acid or the withdrawal of pantoyltaurine. On the other hand, Woolley and White[6] were unable to produce symptoms of pantothenic acid deficiency in weanling mice maintained on purified diets to which 1% of pantoyltaurine was added; their animals received pantoyltaurine at a level 2000 times greater than that of pantothenic acid. Likewise, hamsters fed pantoyltaurine failed to show pantothenic acid deficiency symptoms.[6] The results of our experiments on rats are in agreement with the findings of Woolley and White.[6]

Summary. Feeding of pantoyltaurine failed to produce symptoms of pantothenic acid deficiency in rats. An antagonism between the two substances similar to that established in studies on bacterial growth could not be demonstrated.

[5] Snell, E. E., Chan, A., Spiridanoff, S., Way, E. L., and Leake, C. D., *Science*, 1943, **97**, 168.

[6] Woolley, D. W., and White, A. G. C., PROC. SOC. EXP. BIOL. AND MED., 1943, **52**, 106.

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The Necessity of Additional B-Complex Factors in the Diet of the Chick.

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Factor U, a chick growth factor, may be identical with a bacterial growth factor which occurs in an eluate fraction from norite adsorbate.[1-6] The name "folic acid" has been proposed for the bacterial factor.[6]

Identification of folic acid with a chick "eluate factor" appears to have been accomplished with the isolation of the former in crystalline state, and the demonstration of high growth-promoting activity for bacteria and for chicks, as well as an anemia-prevent-

[1] Snell, E. E., and Peterson, W. H., *J. Bact.*, 1940, **39**, 273.

[2] Hutchings, B. L., Bohonos, N., and Peterson, W. H., *J. Biol. Chem.*, 1941, **141**, 521.

[3] Hutchings, B. L., Bohonos, N., Hegsted, D. M., Elvehjem, C. A., and Peterson, W. H., *J. Biol. Chem.*, 1941, **140**, 681.

[4] Mills, R. C., Briggs, G. M., Jr., Elvehjem, C. A., and Hart, E. B., PROC. SOC. EXP. BIOL. AND MED., 1942, **49**, 186.

[5] Stokstad, E. L. R., *J. Biol. Chem.*, 1941, **139**, 475.

[6] Mitchell, H. K., Snell, E. E., and Williams, R. J., *J. Am. Chem. Soc.*, 1941, **63**, 2284.

TABLE I.
Effects of Concentrates of the *Lactobacillus casei* Factor on Growth and Survival of Chicks from Hens on Experimental Diets.

Exp.	Group	No. of chicks	Chick diet*	Hen diet*	Age of chicks: 2 wks Avg wt, g	2 wks Mortality, %	4 wks Avg wt, g	4 wks Mortality, %
I	1	10	Uns	Practical	59	30	76	70
	2	9	SupA	"	74	0	117	0
II	1	17	Uns	Uns	47	94	—	100
	2	11	SupA	"	69	10	117	10
	3	10	"	"	67	14	120	19
III	1	19	Uns	"	36	95	—	100
	2	12	"	SupC	55	8	58	75
	3	12	SupC	SupB	76	8	130	17
IV	1	15	Uns	Uns	47	93		
	2	15	"	SupB	62	7		
	3	15	"	SupC	65	7		

* Uns = unsupplemented diet, SupA = diet containing preparation A to provide 250 μg of the *L. casei* factor per 100 g, SupB = diet containing preparation B to provide 150 μg per 100 g, SupC = diet containing preparation C to provide 150 μg per 100 g.

ing (vitamin B_c) activity.[7] Recently, it has been postulated that 2 unidentified B-complex factors, different from folic acid, are also required by the chick.[8] It appears, however, that the original folic acid may be multiple in character, since preparations have been reported which are relatively more active for *Streptococcus lactis* than for *Lactobacillus casei*.[9]

It is the purpose of the present report to offer further evidence on the effects of the *Lactobacillus casei* factor on chick growth and survival, in reference to the diet of both the chick and the parent hen. S. C. White Leghorn chicks were obtained from hens fed an experimental diet to be described in another report. This hen diet is deficient in at least one factor contained in an eluate fraction and needed for normal hatchability.[10] The chicks were placed at once upon a synthetic type of diet plus supplements as indicated later.

The chick basal diet contained fibrin, 1; water and ethanol-washed casein, 20; gelatin, 4; cellulose, 5; calcium gluconate, 5; choline chloride, 0.2; inositol, 0.1; cholic acid, 0.1; salt mixture, 4.3;[11] crude soybean oil, 5; fish oil (100D-1000A), 1.0; vitamin supplements; and cerelose to complete 100 parts. Vitamin supplements as mg per kg of ration were: thiamin, 5; riboflavin, 10; pyridoxine, 5; calcium pantothenate (dl), 40; nicotinic acid, 50; 2-methyl-1, 4-naphthohydroquinone diphosphate (tetra sodium salt), 10; and a commercial biotin source, known by assay to be free of the *L. casei* factor, in an amount sufficient to prevent all indications of biotin deficiency. The preparations of the *L. casei* factor consisted of (A), a crude calcium salt; (B), a further refined preparation; (C), an eluate fraction prepared by Dr. S. Lepkovsky according to a published procedure;[1] and (D), a preparation approximately 9% pure.*

[7] Pfiffner, J. J., Binkley, S. B., Bloom, E. S., Brown, R. A., Bird, O. D., Emmett, A. D., Hogan, A. G., and O'Dell, B. L., *Science*, 1943, **97**, 404.

[8] Briggs, G. M., Jr., Luckey, T. D., Elvehjem, C. A., and Hart, E. B., *J. Biol. Chem.*, 1943, **148**, 163.

[9] Keresztesy, J. C., Rickes, E. L., and Stokes, J. L., *Science*, 1943, **97**, 465.

[10] Cravens, W. W., Sebesta, E. E., Halpin, J. G., and Hart, E. B., PROC. SOC. EXP. BIOL. AND MED., 1942, **51**, 106.

[11] Almquist, H. J., and Mecchi, E., PROC. SOC. EXP. BIOL. AND MED., 1941, **48**, 526.

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TABLE II.
Effects of Supplements of the *Lactobacillus casei* Factor and Additional Unidentified Factors on Chick Growth.

Supplement and amount of the *L. casei* factor per 100 g diet μg	Supplement of additional factors per 100 g diet	No. of chicks	Avg chick wt at 2 weeks g
0	none	29	55
C, 84	"	10	73
C, 150	"	25	73
C, 250	"	10	77
D, 150	"	16	60
C, 150	SL*, 2g	15	90
"	YE†, ≅ 6g	15	92
Practical diet only	none	38	89

* Solubilized liver.
† A 50% methanol extract of yeast, concentrated to remove methanol.

The level fed in chick diets was at least 50% greater than the requirement of the chick so far as reported.[3]

The results given in Table I (Exp. I) show a distinct difference in the growth and survival of chicks from hens on a normal breeding diet, depending upon the supplementation of the chick diet. In Exp. II it was found that, when fed the basal chick diet, chicks from hens fed an unsupplemented experimental diet showed particularly severe mortality, most of them dying within 2 weeks. These chicks also showed a very poor growth, as compared to chicks of the same origin receiving a supplement of the *L. casei* factor in their diet. In Experiments III and IV the same findings in respect to extremely heavy mortality and poor growth of chicks were obtained. Similarly fed chicks which originated with hens receiving a supplement of the *L. casei* factor in their diets showed comparatively good survival to at least 2 weeks, and an intermediate rate of growth.

Such results indicate an appreciable "carryover" of a factor (or factors) from the hen diet to the chick. This carryover was essentially the same whether supplement B or C was fed the hens. After 2 weeks, however, this reserve was depleted, the chicks ceased to grow and mortality became severe. Prior to death many chicks became extremely weak, resembling chicks previously described,[12] but it is doubtful whether such chicks should be referred to as "paralyzed."

The maximal weights attained under the most favorable conditions were considerably below the normal weights for the strain and species of birds used, being approximately 120-130 g at 4 weeks, while a normal weight for chicks of this age and breed is approximately 240 g.

In comparing the growth of chicks on diets supplemented with the *L. casei* factor with the growth of chicks receiving additional supplements (Table II) it became apparent that preparations A, C and D would permit no more than a distinctly subnormal rate of growth, although mortality was largely prevented. The purest of these preparations, D, permitted the least growth. Additional crude supplements, such as solubilized liver and yeast extract, caused an increase in rate of growth to normal (approximately 90 g weight at 2 weeks). This increase may be attributed to an additional unknown factor (or factors), as previously suggested.[4,12]

Summary. Concentrates of the *Lactobacillus casei* factor in the diet of the hen affect the growth and survival of chicks fed diets deficient in this factor. In addition, at least one unknown member of the B-complex is required by chicks.

12 Briggs, G. M., Jr., Luckey, T. D., Mills, R. C., Elvehjem, C. A., and Hart, E. B., PROC. SOC. EXP. BIOL. AND MED., 1943, **52**, 7.