

## Sparing Action of Protein on the Pantothenic Acid Requirement of the Rat.\*

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Studies on the requirement of the rat for pantothenic acid have, so far, been limited to diets containing a high proportion of carbohydrate with minimal levels of protein and fat. On these high-carbohydrate diets, it is well known that pantothenic acid deficient rats usually survive only 1 to 3 months.<sup>1,2,3</sup> (Table I.) The importance of the composition of the diet in relation to vitamin requirements is also well-known; *e.g.*, less thiamine is required for animals maintained on high fat<sup>4,5</sup> and high protein<sup>6,7,8</sup> diets than for diets high in carbohydrate whereas more riboflavin is required for rats receiving diets high in fat.<sup>9,10</sup>

In the course of experiments with diets vary-

ing in their proportions of carbohydrate, protein, and fat a marked sparing action of protein on the pantothenic acid requirement of the rat was discovered. When rats received diets deficient in pantothenic acid but containing 64% protein (casein) instead of 24% protein, growth was markedly accelerated and survival was greatly increased. This effect was observed both when the deficiency was instituted at birth and when it was instituted at weaning.

**Experimental and Results.** The basal (high carbohydrate) diet was composed of 64% sucrose, 24% alcohol-extracted casein, 8% hydrogenated cottonseed oil (Crisco), and 4% salts.<sup>11</sup> The high protein diet contained 64% casein and 24% sucrose with the remainder of the constituents identical. Both diets contained crystalline B vitamins per kg diet: 5 mg thiamine HCl, 5 mg pyridoxine HCl, 10 mg riboflavin, 10 mg *p*-aminobenzoic acid, 20 mg nicotinic acid, 400 mg inositol, and 1 g choline chloride. Each rat received weekly a fat-soluble vitamin mixture containing 325 mg corn oil (Mazola), 400 U.S.P. units vitamin A, 58 chick units vitamin D, and 3 mg alpha-tocopherol.

In the first series of animals, equivalent groups of male and female rats (Long-Evans strain) were placed on the deficient diets at weaning. Table I shows the marked effect of the high casein diet on growth and survival.

In the second series, litters of 6 young together with the mother were placed on the deficient diets the day of birth. The litters were weaned on the 21st day and continued on the same diet. This procedure results in a very acute deficiency<sup>12</sup> as can be seen in Table II. However, the sparing effect of the

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TABLE I.

Effect of Dietary Protein Level on Growth and Survival in Pantothenic Acid Deficiency Instituted at Weaning.

| Exp. group<br>% protein | No. of<br>rats | Avg wt<br>Day 21<br>g | Avg wt<br>Day 60<br>g | Survival<br>Day 60<br>% | Avg wt<br>Day 90<br>g | Survival<br>Day 90<br>% | Avg wt<br>Day 120<br>g | Survival<br>Day 120<br>% |
|-------------------------|----------------|-----------------------|-----------------------|-------------------------|-----------------------|-------------------------|------------------------|--------------------------|
| 24                      | 13 ♀           | 47                    | 84                    | 54                      | 93                    | 8                       | —                      | 0                        |
|                         | 10 ♂           | 48                    | 110                   | 80                      | 110                   | 50                      | 94                     | 10                       |
|                         | 12 ♂           | 47                    | 100                   | 58                      | 120                   | 42                      | 132                    | 25                       |
| 64                      | 12 ♀           | 48                    | 115                   | 83                      | 170                   | 83                      | 202                    | 83                       |
|                         | 10 ♂           | 47                    | 141                   | 90                      | 234                   | 90                      | 300                    | 90                       |
|                         | 12 ♂           | 47                    | 124                   | 92                      | 205                   | 83                      | 255                    | 83                       |

TABLE II.

Effect of Dietary Protein Level on Growth and Survival in Pantothenic Acid Deficiency Instituted at Birth.

| Exp. group<br>% protein | No. of<br>rats | Avg wt<br>Day 1<br>g | Avg wt<br>Day 21<br>g | Survival<br>Day 21<br>% | Avg wt<br>Day 30<br>g | Survival<br>Day 30<br>% | Avg wt<br>Day 45<br>g | Survival<br>Day 45<br>% | Avg wt<br>Day 60<br>g | Survival<br>Day 60<br>% |
|-------------------------|----------------|----------------------|-----------------------|-------------------------|-----------------------|-------------------------|-----------------------|-------------------------|-----------------------|-------------------------|
| 24                      | 40 ♂           | 6.3                  | 30                    | 81                      | 32                    | 35                      | 38                    | 7                       | 51                    | 1                       |
|                         | 38 ♀           | 6.1                  | 27                    |                         | 31                    |                         | 32                    |                         | —                     |                         |
| 64                      | 30 ♂           | 6.0                  | 31                    | 88                      | 31                    | 76                      | 53                    | 37                      | 83                    | 14                      |
|                         | 30 ♀           | 5.9                  | 30                    |                         | 30                    |                         | 44                    |                         | 73                    |                         |

64% casein diet on growth and survival is still clearly shown.

The superiority of the high protein diet may actually be due to the decreased level of dietary carbohydrate inasmuch as there is evidence that pantothenic acid is involved in carbohydrate metabolism.<sup>13</sup> On the other hand, it may also be due to some component(s) of the casein such as (a) traces of pantothenic acid; (b) the amino acid content; (c) the phosphorus content; (d) unknown factors present which may be related to the utilization of pantothenic acid or to biotin and folic acid which affect pantothenic acid utilization.<sup>14,15</sup> Extensive analyses of various purified and "vitamin-free" caseins have shown the presence of pantothenic acid in all samples, varying from 0.8 to 3.0  $\mu$ g per gram.

However, experiments now in progress indicate that the pantothenic acid content of the casein in use is responsible for only part of the sparing action.

*Summary.* Rats maintained either from birth or from weaning on a pantothenic acid-deficient diet containing 64% casein grow more and survive longer than littermates maintained on a 24% casein-deficient diet under the same conditions. Several possible explanations for this sparing action are given.

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