

alpha-protein synthetic milk was deficient in vitamin B<sub>12</sub>, two animals were maintained without supplementation of this vitamin until death. These two animals received a single initial injection of 0.4γ per kg. of body weight of vitamin B<sub>12</sub> which allowed them to gain "normally" until the third week, at which time they began to lose weight and finally expired in the fourth and fifth weeks, respectively (Table II).

The performance of the animals in the second experiment is tabulated in Table II. Thirty-two animals were randomly assigned to the treatment groups originally, but two of them are excluded from the analysis: One animal did not learn to eat and was sacrificed during the first week just prior to starvation, while the second gained only 0.39 kg. during the 5-week experimental period.

Analysis of covariance showed that there was no significant difference in average final weight and feed consumption between treatments, each group being adjusted to equal initial weights. In this experiment the effect of variation

**Table I. Composition of Basal Ration**

| Constituents <sup>a</sup>                             | %              |
|---|----------------|
|   | Dry Basis      |
| Alpha-protein (isolated soybean protein) <sup>b</sup> | 29.4           |
| DL-Methionine <sup>c</sup>                            | 0.6            |
| Sucrose   | 30.9           |
| Lard  | 30.8           |
| Mineral salts <sup>d</sup>                            | 8.3            |
|   | Mg./Liter Milk |
| Water-soluble vitamins <sup>e</sup>                   |                |
| Thiamine  | 0.65           |
| Riboflavin  | 1.30           |
| Calcium pantothenate                                  | 7.80           |
| Nicotinic acid  | 2.60           |
| Pyridoxine hydrochloride                              | 1.30           |
| Choline chloride                                      | 130.00         |
| β-Amino benzoic acid                                  | 2.60           |
| Pteroylglutamic acid                                  | 0.052          |
| Ascorbic acid   | 16.00          |
| Biotin  | 0.01           |
| Inositol  | 26.00          |
| Fat-soluble vitamins                                  |                |
| Vitamin A   | 2000 I.U.      |
| Vitamin D   | 200 I.U.       |
| α-Tocopherol acetate                                  | 1              |
| 2-Methyl-1,4,-naphthoquinone                          | 26             |

<sup>a</sup> Homogenized into a synthetic milk containing 19.5% solids and 6% fat.

<sup>b</sup> Obtained from Glidden Co., Chicago, Ill.

<sup>c</sup> Supplied by Dow Chemical Co., Midland, Mich., through courtesy of Julius Johnson.

<sup>d</sup> For composition of mineral mixture, see (7).

<sup>e</sup> Thiamine hydrochloride, riboflavin, pyridoxine hydrochloride, calcium pantothenate, biotin, nicotinic acid, ascorbic acid, and α-tocopherol acetate were generously supplied by Merck and Co., Inc., Rahway, N. J., through the courtesy of H. H. Draper. Pteroylglutamic acid was supplied by the American Cyanamid Co., Pearl River, N. Y., through the courtesy of T. H. Jukes.

**Table II. Effect<sup>a</sup> of Sewage Sludge and Crystalline Vitamin B<sub>12</sub> on Vitamin B<sub>12</sub>-Deficient Baby Pig**

(5-Week experimental period)

| Treatments                             | 1            | 2                            | 3                            | 4   |
|--|--------------|------------------------------|------------------------------|---|
|  | Basal Ration | Basal Ration + Sewage Sludge | Basal + Vit. B <sub>12</sub> | Basal Ration + Sewage Sludge + Vit. B <sub>12</sub> |
| No. of pigs                            | 2            | 10                           | 10                           | 10  |
| Average initial wt., kg.               | 3.21         | 1.88                         | 1.88                         | 1.85  |
| Average final wt., kg.                 | 5.68         | 8.74                         | 8.99                         | 8.02  |
| Average total gain, kg.                | ...          | 6.86                         | 7.11                         | 6.16  |
| Average daily gain, kg.                | 0.079        | 0.196                        | 0.203                        | 0.176   |
| Average total dry matter consumed, kg. | 7.48         | 7.74                         | 7.59                         | 6.52  |

<sup>a</sup> None of the differences indicated between groups 2, 3, and 4 are statistically significant.

in original weight on final weight was less, when the conditions of temperature and humidity were kept constant, by an amount significant at the 5% level.

From these results it appears that dried activated sewage sludge fed at the 2% level is a satisfactory source of vitamin B<sub>12</sub> for the pig. When fed at this level, dried activated sewage sludge does not appear to have any harmful effect on the animals over a 5-week period and in a pilot experiment carried out under adverse conditions gave some evidence of growth stimulation above that due to vitamin B<sub>12</sub>.

Baby pigs, maintained on a synthetic milk shown to be deficient in vitamin B<sub>12</sub> and containing alpha-protein as the nitrogen source, made gains over a 5-week period which were not significantly different when supplemented either intramuscularly with crystalline vitamin B<sub>12</sub> at a level of 0.8γ per kg. of body weight per day or orally with 20 grams of dried activated sewage sludge shown to contain 3.11γ of vitamin B<sub>12</sub> activity per gram (dry weight). The effect of variations in initial weight on final weight was shown to be reduced when temperature and humidity were kept constant.

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#### Correction

In the article on "Malodorous Fermentation. Acidic Constituents of Zapterera of Olives" [Delmouzos, J. G., Stadtman, F. H., and Vaughn, R. D., *J. Agr. Food Chem.*, **1**, 333 (1953)] the fourth line should read: "fermentation also sometimes found in storage fruit."