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THE EFFECT OF RIBOFLAVIN AND THE FILTRATE FACTOR ON EGG PRODUCTION AND HATCHABILITY

S. LEPKOFSKY, L. W. TAYLOR, T. H. JUKES, AND H. J. ALMQVIST

THE VITAMIN-B COMPLEX AS RELATED TO GROWTH AND METABOLISM IN THE PIG

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AN EARLY REPORT of a nutritional disturbance in pigs is that of Rommel and Vedder (1915), who produced a condition in them resembling human beriberi with a ration of steam-polished rice and tankage; at that time, however, these workers thought it was analogous to cottonseed-meal poisoning. Blissett and Golding (1931) reported that pigs fed wheat flour and white fish meal grew irregularly and were unthrifty and rough-coated. The English workers, Birch, Chick, and Martin (1937) noted scouring in pigs fed a modified Goldberger pellagra-producing diet, 5 to 6 weeks after their experiments began, which was corrected by the addition of yeast. Very recently, after the work of Elvehjem and his associates (1937), and while this work was being prepared for publication, Chick, Macrae, Martin, and Martin (1938) showed that the active factor in yeast was nicotinic acid. Since nicotinic acid has been demonstrated as a factor in the cure of human pellagra (Fouts, et al., 1937) and since the basal pig diet used was a pellagra-producing one, this nutritional disorder in the pig would seem to be analogous and might be designated as “pig-pellagra.”

At the California Agricultural Experiment Station, pigs on deficiency diets frequently showed dry curly hair, faulty elimination, a lack of normal muscular control, and slow growth. Casein in small quantities when added to a barley diet, markedly increased the rate of gain (Hughes, 1937); moreover, young fattening pigs grew particularly well when certain dairy by-products were a part of the diet; and pigs given access to pasture gained more rapidly than similar pigs in dry lot (Hughes and Roadhouse, 1937).

Because milk products and pasture have an acknowledged importance in pork production, and because they are rich in the vitamin-B complex, it seemed particularly appropriate that the nutritional factors compris-

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2 Aided by a grant from American Dry Milk Institute, Inc.
3 Professor of Animal Husbandry and Animal Husbandman in the Experiment Station.
4 See “Literature Cited” at the end of this paper for complete data on citations, which are referred to in the text by author and date of publication.
ing the vitamin-B complex should be studied critically with the pig as the experimental animal. Much is known concerning these factors and their importance for growth and well being in the rat and the chick, but very little concerning their effects on the pig.

An attempt was made in these studies to employ a purified diet to approach as nearly as possible with the pig the technique so successfully in use with small laboratory animals. A basal diet as low as possible in the vitamin-B complex was used, and supplemented with pure vitamins where obtainable; where pure vitamins were not obtainable, the best possible extracts or adsorbates were employed. In addition, natural feedstuffs—whey powder, skim-milk powder, and yeast—were used as supplements.

Three experiments were conducted: the first began in November, 1936, and the third was finished in March, 1938.

EXPERIMENTAL PROCEDURE

Animals and Their Care.—A total of 83 pigs have been included in these studies—purebred Duroc Jerseys, Poland Chinas, and crossbred Duroc Jerseys and Poland Chinas. The breeds were distributed uniformly in all the lots. The number in each lot is given in table 1. Their average initial weight was between 40 and 50 pounds; their average age about 60 days. They were placed on experimental diets at weaning time or within 10 days thereafter. They were fed on concrete floors and had free access to pens inside and outside of the barn, but did not have access to dirt.

Pigs on the basal diet seemed cold even though the outside temperature was mild, for they huddled together in the corner of the pens; therefore, rectal temperatures and respirations per minute were taken and recorded every 14 days (tables 2 and 3) for most of the pigs.

Feeds and Materials Used and Their Preparation.—As may be noted below, all diets contained brewers' rice or rice screenings, casein, cod-liver oil, and a salt mixture. Used interchangeably in the basal diet, brewers' rice (small cracked kernels of polished rice run through a fanning mill to remove all bran) and rice screenings (medium-sized pieces of commercial polished rice thoroughly cleaned at the mill) were chosen because of their high starch value and relative freedom from vitamins of the B complex; they contained 5.02 per cent protein. The granulated casein used would pass through an 80-mesh screen and had been manufactured by the acid-precipitation method. Except in diets containing whey adsorbate, whey powder, or skim-milk powder, vitamins of the B complex were removed from the casein by washing according to the method of Evans, Lepkovsky, and Murphy (1934), modified by that of Supplee,
Flanigan, Hanford, and Ansbacher (1936). The cod-liver oil was Squibb’s tested oil. The salt mixture had the following composition, in parts by weight:

<table>
<thead>
<tr>
<th>Part</th>
<th>Parts</th>
<th>Part</th>
<th>Parts</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calcium Phosphate (Dicalpho)</td>
<td>20.00</td>
<td>Magnesium oxide</td>
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</tr>
<tr>
<td>Sodium chloride</td>
<td>10.00</td>
<td>Copper sulfate</td>
<td>0.10</td>
</tr>
<tr>
<td>Calcium carbonate (oystershell flour)</td>
<td>5.00</td>
<td>Manganese sulfate</td>
<td>0.05</td>
</tr>
<tr>
<td>Iron (95 per cent citrate and 5 per cent oxide)</td>
<td>1.50</td>
<td>Zinc oxide</td>
<td>0.05</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Cobalt carbonate</td>
<td>0.05</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Potassium iodide</td>
<td>0.05</td>
</tr>
</tbody>
</table>

Fig. 1.—A, Riboflavin assay (using the chick as the experimental animal) of the yeast, autoclaved yeast, and whey adsorbate used; B, riboflavin assay of dried liver from pigs fed diets 1, 6, and 8 (basal, basal + autoclaved yeast, and basal + raw yeast). The arrows indicate when a change from 0.8 to 1.4 per cent of dried liver was made.
Most of the supplements used were commercial products. The whey adsorbate used as a concentrated source of riboflavin, however, was made at the California Agricultural Experiment Station by the process described by Jukes (1937b). It may have contained also a small amount of thiamin. The thiamin (synthetic crystalline vitamin B₁) was purchased from Merck and Company; the nicotinic acid from the Eastman Kodak Company; rice-bran filtrate (as a source of filtrate factor or factor 2), treated three times with fuller's earth (Lepkovsky, Jukes, and Krause, 1936) to remove most of factor 1, from the Vitab Products, Inc.; yeast foam tablet powder, pure dehydrated yeast, containing 44.0 per cent protein, from the Northwestern Yeast Company. The autoclaved yeast was prepared by autoclaving at 125°C and 25 pounds' pressure for 6 hours in shallow pans, with the yeast not over 1½ inches thick. The lactose powder U. S. P. was purchased from Eimer and Amend; the whey powder, containing 11.6 per cent protein, was furnished by the Western Condensing Company. The skim-milk powder was manufactured at the California Agricultural Experiment Station by the atmospheric-drum process; it analyzed 33.36 per cent protein.

The yeast, autoclaved yeast, and whey adsorbate fed were assayed for their riboflavin content by the chick-growth method (Jukes, 1937a), described in the accompanying paper (Lepkovsky, et al., 1938). The results are shown in figure 1, A. The autoclaved yeast contained less riboflavin than the raw yeast; part of the riboflavin in the yeast was apparently destroyed by autoclaving.

**Basal and Supplemented Diets.**—The diets fed to the various lots were as follows:

- Lot 1: Basal diet, brewers' rice or rice screenings, 88 pounds; casein, purified, 9 pounds; salt mixture, 3 pounds; cod-liver oil by mouth as described below.
- Lot 2: Basal, 99 pounds; whey adsorbate, 1 pound.
- Lot 3: Basal, 99 pounds; whey adsorbate, 1 pound; thiamin by mouth as described below.
- Lot 4: Basal, 99 pounds; whey adsorbate, 1 pound; thiamin and nicotinic acid by mouth as described below.
- Lot 5: Basal, 94 pounds; whey adsorbate, 1 pound; thiamin and nicotinic acid by mouth as described below; rice-bran filtrate, 5 pounds.
- Lot 6: Basal, 97 pounds; autoclaved yeast, 3 pounds.
- Lot 7: Basal, 97 pounds; autoclaved yeast, 3 pounds; thiamin by mouth as described below.
- Lot 8: Basal, 97 pounds; yeast, 3 pounds.
- Lot 9: Basal, 94 pounds; whey adsorbate, 1 pound; lactose, 5 pounds.

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* Under the direction of G. A. Richardson.
* Under the direction of Thomas H. Jukes.
Lot 10: Rice screenings, 80 pounds; casein, 8 pounds; whey powder, 10 pounds; salt mixture, 2 pounds; cod-liver oil by mouth as described below.

Lot 11: Rice screenings, 81 pounds; casein, 5 pounds; skim-milk powder, 12 pounds; salt mixture, 2 pounds; cod-liver oil by mouth as described below.

The protein content of all these diets was kept at about 13.75 per cent. In those diets where yeast, autoclaved yeast, whey powder, and skim-milk powder were fed, the casein was reduced sufficiently to maintain the protein at this level.

Each individual was given 5 cc of cod-liver oil by mouth each week until about 70 pounds in weight, and 10 cc weekly thereafter. Thiamin and nicotinic acid, when included in the diet, were given to the pigs in water solution by mouth at the rates of 4.2 mg and 13.7 mg per head per 100 pounds daily, respectively.

The results obtained in this investigation are shown in table 1. It is a summary of three experiments: one major experiment—the third—which included all lots except 6, and two more-limited trials which included lots 1, 6, and 8 in one instance and lots 1, 2, 6, and 8 in the other. Because the results in all trials were similar, they are combined in this table. The experiments were in progress at least 98 days, with some curative measures continued thereafter.

RESULTS WITH PURE VITAMINS AND VITAMIN CONCENTRATES

As shown in table 1 and figure 2, the pigs did very poorly on the basal diet (lot 1). Their rate of gain was slow and their efficiency of food utilization poor.

The addition of whey adsorbate (lot 2) to supply riboflavin (and perhaps some thiamin) increased markedly the average daily gain and reduced the amount of feed consumed for a unit of increase in body weight.

Further addition of thiamin (lot 3) had an adverse effect as compared with the addition of whey adsorbate only. This experimental finding has at present no explanation, unless it is assumed that the balance of some of these water-soluble vitamins has a rôle in nutrition. All the pigs in lots 1, 2, and 3 gained slowly; anorexia was observed after the first two weeks; and most of the pigs were unthrifty, having dry curly hair.

The most striking improvement was observed when, in addition to riboflavin and thiamin, nicotinic acid was added. Pigs in lot 4 were much superior to those in lots 1, 2, and 3. Their rate of growth was much higher; their feed consumption for a pound of gain was much less; their appetites were better; and they surpassed the former groups in their thriftiness, in their sleek coats of hair, in the fine condition of their skins,
and in their general well-being (figs. 3, 4, and 5). Thus with a purified diet, using pure or purified vitamins, the importance of nicotinic acid to swine nutrition has been clearly demonstrated. This demonstration of the importance of nicotinic acid in swine nutrition is in agreement with those of Chick et al. (1938), who used a diet of natural foodstuffs. Since

### TABLE 1

**Diets Fed, Initial Weights, and Feed Consumed for 100 Pounds of Gain for the Various Groups**

<table>
<thead>
<tr>
<th>Lot No.</th>
<th>Diet fed</th>
<th>Number of pigs</th>
<th>Average initial weight, in pounds</th>
<th>Average daily gain, in pounds</th>
<th>Feed consumed for 100 pounds gain, in pounds</th>
</tr>
</thead>
<tbody>
<tr>
<td>Purified diets</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1*</td>
<td>Basal†</td>
<td>17</td>
<td>43.4</td>
<td>0.17</td>
<td>705</td>
</tr>
<tr>
<td>2*</td>
<td>Basal + whey adsorbate</td>
<td>12</td>
<td>42.9</td>
<td>0.40</td>
<td>451</td>
</tr>
<tr>
<td>3</td>
<td>Basal + whey adsorbate + thiamin</td>
<td>5</td>
<td>46.4</td>
<td>0.23</td>
<td>657</td>
</tr>
<tr>
<td>4</td>
<td>Basal + whey adsorbate + thiamin + nicotinic acid</td>
<td>5</td>
<td>45.6</td>
<td>1.16</td>
<td>311</td>
</tr>
<tr>
<td>5</td>
<td>Basal + whey adsorbate + thiamin + nicotinic acid + factor 2 concentrate</td>
<td>5</td>
<td>44.5</td>
<td>1.59</td>
<td>303</td>
</tr>
<tr>
<td>Diets composed of natural feedstuffs</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>6*</td>
<td>Basal + autoclaved yeast</td>
<td>9</td>
<td>40.4</td>
<td>0.57</td>
<td>358</td>
</tr>
<tr>
<td>7</td>
<td>Basal + autoclaved yeast + thiamin</td>
<td>5</td>
<td>45.6</td>
<td>0.77</td>
<td>328</td>
</tr>
<tr>
<td>8*</td>
<td>Basal + yeast</td>
<td>10</td>
<td>41.2</td>
<td>1.14</td>
<td>284</td>
</tr>
<tr>
<td>9</td>
<td>Basal + whey adsorbate + lactose</td>
<td>5</td>
<td>45.2</td>
<td>0.31</td>
<td>620</td>
</tr>
<tr>
<td>10</td>
<td>Basal + whey powder</td>
<td>5</td>
<td>44.8</td>
<td>0.77</td>
<td>362</td>
</tr>
<tr>
<td>11</td>
<td>Basal + skim-milk powder</td>
<td>5</td>
<td>45.6</td>
<td>0.96</td>
<td>357</td>
</tr>
</tbody>
</table>

* Lots 1 and 8 average of three experiments; lots 2 and 6 average of two experiments; all other lots are one experiment only.
† Basal diet: rice screenings or brewers' rice, 88 pounds; casein, purified, 9 pounds; salt mixture, 3 pounds; cod-liver oil, 5-10 cc per head per week.

their diet was pellagra-producing, it should be pointed out that in this investigation a highly refined pellagra-producing diet has been developed.

While thiamin, riboflavin (in whey adsorbate), and nicotinic acid gave excellent growth, physical well-being, and efficient food utilization, improvement was obtained by the addition of rice-bran filtrate (lot 5), containing largely the filtrate factor (or factor 2) and some factor 1 (vitamin B	extsubscript{1}).

Dietary conditions for the pig have therefore been developed whereby all the factors of the vitamin-B complex, except factor 1, can be evaluated for their rôle in swine nutrition. While from these studies the rôle of factor 1 in the nutrition of the pig cannot be determined, the problem is under investigation.
Fig. 2.—Growth curves for pigs in lots 1 to 11 of table 1, with a "normal" growth curve for hogs of similar weight in the station herd. The upper curves are for lots fed purified diets, the lower for those fed natural feedstuffs.
RESULTS WITH YEAST, AUTOCLAVED YEAST, LACTOSE, WHEY POWDER, AND DRIED SKIM MILK

Using 3 per cent yeast as the source of the vitamin-B complex (lot 8), the results (table 1) indicated that some vitamin or vitamins were still partially inadequate, for less growth was obtained than that with the purified diet with all the vitamins added (lot 5).

Autoclaving yeast (lot 6) markedly reduced some of its vitamin content. Since thiamin is destroyed by autoclaving, it was added along with the autoclaved yeast in lot 7. Improved growth resulted, but the per-
formance of the pigs was still far short of those on the untreated yeast. Analysis of the yeast for riboflavin, using the chick as the experimental animal (fig. 1, A) indicated some destruction of the riboflavin during the autoclaving because those fed autoclaved yeast gained more slowly than those fed raw yeast. Analysis was also made in a similar manner of the

![Image of pigs in lots 4 and 5](image)

Fig. 4.—Thrifty, uniform, sleek-coated pigs in lots 4 and 5; note fine, smooth hair. Riboflavin content of the livers of pigs fed diets 1, 6, and 8. The livers were dried under vacuum at 194°F. This material was fed to chicks at 0.8 and 1.4 per cent, the change being indicated by the arrows (fig. 1, B). These results, like the former, indicate a loss of riboflavin in the autoclaved yeast. Whether this reduction of the riboflavin was responsible for its inferiority over the untreated yeast cannot be ascertained from the evidence available.
Pigs fed lactose (lot 9) did well for the first two weeks then developed anorexia, so that their final rate of growth and condition resembled those of lots 1, 2, and 3.

Skim-milk powder appears to be a fairly good source of the vitamin-B complex for the pig (lot 11). It is, however, moderately inadequate in one or more vitamins, and is under investigation.

Whey powder seems to be similar to dried skim milk in supplying these factors to the pig (lot 10). While in table 1 and figure 2 its performance appears poorer than skim-milk powder, the difference is probably more apparent than real since difficulty was encountered in getting the pigs to eat the whey powder at the beginning of the experiment. After the pigs developed an appetite for the whey powder and ate it more readily, their rate of increase was equal to those fed skim-milk powder.

Graphic presentations of the growth rates of all lots are given in figure 2.

OBSERVATIONS ON THE BODY TEMPERATURE AND RESPIRATORY RATE AS RELATED TO THE PROBLEM

Tables 2 and 3 show average temperatures and respiration rates taken throughout the experiment. In general, the temperatures of all the pigs were higher and the respiratory rate was more rapid at the beginning of the experiment than at the close. There is obviously a relation between the diets fed, the rate of growth, the body temperature, and the respiratory rate. The pigs on the basal diet (lot 1) gained slowly, their respiratory rate was slow, and their average temperatures were below normal. Those pigs that grow slowly—namely, lots 1, 2, 3, and 9—had lower tem-
peratures and slower respiratory rates at the close of the experiment than
the other groups. Furthermore, pigs in lot 5 grew faster than any other
group, and their temperature and respiratory rate were the highest of
any at the close of the experiment. The average temperature and respira-
tory rate were a very good index of the rate of gain and the physical
well-being of the pigs on these experiments.

TABLE 2
AVERAGE RESPIRATORY RATE OF THE PIGS IN THE THIRD EXPERIMENT*
(In respirations per minute)

<table>
<thead>
<tr>
<th>Lot No.</th>
<th>October 1, 1937</th>
<th>November 10, 1937</th>
<th>December 18, 1937</th>
<th>January 26, 1938</th>
<th>February 14, 1938</th>
<th>Average for all</th>
</tr>
</thead>
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<tr>
<td>1</td>
<td>25.6</td>
<td>17.1</td>
<td>10.6</td>
<td>11.9</td>
<td>16.3</td>
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<tr>
<td>2</td>
<td>27.3</td>
<td>23.0</td>
<td>15.7</td>
<td>19.3</td>
<td>22.1</td>
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</tr>
<tr>
<td>3</td>
<td>21.8</td>
<td>20.6</td>
<td>16.8</td>
<td>15.1</td>
<td>18.6</td>
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</tr>
<tr>
<td>4</td>
<td>28.5</td>
<td>23.0</td>
<td>21.1</td>
<td>19.3</td>
<td>24.0</td>
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<td>28.6</td>
<td>21.1</td>
<td>19.0</td>
<td>25.6</td>
<td></td>
</tr>
</tbody>
</table>

* Lot 6 not shown because this diet was not fed in the third experiment.

TABLE 3
AVERAGE TEMPERATURE OF THE PIGS IN THE THIRD EXPERIMENT*
(In degrees Fahrenheit)

<table>
<thead>
<tr>
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<td>102.6</td>
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</tr>
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</table>

* Lot 6 not shown because this diet was not fed in the third experiment.

RESULTS OF CURATIVE MEASURES

At the conclusion of the experiment, curative measures for a 10-day
period were employed in an effort to obtain additional information. Two
pigs from lot 1 were fed 2 mg of thiamin daily for each 50 pounds in
weight, while 2 other pigs from the same lot were fed a like amount of
thiamin and also 1 per cent whey adsorbate. In each case there was a slight increase in body weight and an increase of slightly more than 1° F in body temperature. There was not much change in the average respiratory rate.

Thiamin, at the same rate as previously mentioned, was added to the diet of 2 pigs in lot 2 but produced no noticeable change in a month's time. Some of the animals in lots 2 and 3 (those of about the same weight were selected) were given nicotinic acid at the rate of 17.8 mg per 100 pounds per head daily, which doubled their rate of gain in 10 days; they had an increase of 1.4° F in body temperature and an increase of 6 respirations per minute.

The pigs in lot 4 were given the rice-bran filtrate (5 per cent of the diet), which in the same length of time increased their average daily gain from 2.1 (rate of gain last 14 days of the experiment) to 3.4 pounds per head daily. There was a slight increase in body temperature and an increase of 5 respirations per minute.

When thiamin was removed from the pigs in lot 5, the rate of gain and the temperature did not change in 10 days, but the respiratory rate decreased slightly.

When thiamin was added to lots 10 and 11 (4.6 mg was given per 100 pounds daily), the average daily gain of the pigs was increased thereby from 1.4 pounds each daily during the last 14 days of the experiment to an average of 2.9 pounds during the 10-day curative period. There was an average increase of 0.5° F in temperature and of 5 respirations per minute.

POST-MORTEM FINDINGS

Several pigs died during these experiments, others were killed at the conclusion. Most, but not all, came to autopsy. Of 6 pigs in lot 1 that were autopsied, all were in poor condition, with hair dry and curly. The stomachs were full of feed, with some gastritis at the most dependent area of the stomach. All but 1 showed flabby hearts; and 4 had some fluid in the lungs, though there was no pneumonia in any of them. The small intestines of 3 contained some catarrhal exudate. The others appeared normal. In the large intestines of 1 there was a slight hemorrhagic area. In 2 of the pigs the gall bladders appeared small, and 1 was completely collapsed. The liver of 1 was small and pale. Two showed a few pin-point hemorrhages in their bladders.

When 2 pigs in lot 2 were autopsied, both their stomachs were full of feed, and 1 showed some gastritis. There was some exudate in the small intestines of both, with slight ulcerations below the illio-cecal valve in 1.
There was no comment about the heart of 1, the right heart of the other was slightly flabby.

When 3 pigs in lot 6 came to autopsy, all their stomachs were full of feed, and 2 showed some gastritis. There was a clot in the left heart of 1 at the semilunar valve which showed fibrin shreds. The hearts of the other 2 were apparently normal; both showed small gall bladders, and the liver of 1 was below normal in size; the spleens of both were small.

In general the pigs that died in lots 1, 2, and 6 were in poor condition; they were thin and had sometimes walked with difficulty. Their hair and skins were dry. Their stomachs were full of feed, with little or no feed in the small intestines. The small intestine often showed a catarrhal exudate, and the feces, if any, in the large intestine were hard and sticky. The heart was flabby, particularly the right heart; the spleen was often small; and the gall bladder was sometimes undersized. The musculature of the alimentary canal was not normal. While the stomach was full of feed, there was apparently not much mixing of the feed in it. The emptiness of the small intestine raises the question of why the feed did not pass out of the stomach. Possibly the pyloric valve failed in its function and there was a slowing down of the normal process along the tract.

DISCUSSION

That the basal diet used in these studies was sufficient except for the vitamins of the B complex there was little doubt. No vitamin-A deficit was indicated at any time. That the salt mixture was efficient seems certain because of the excellent growth obtained when the vitamins were supplied. The energy and protein content was such that where all factors were supplied, growth was rapid, and the physical well-being of the animals was excellent. The basal ration without (or with very small amounts of) the various factors of the vitamin-B complex caused a loss of appetite, exceedingly slow growth, lack of thrift, and sometimes death in the young weaned pig (lot 1, table 1).

While the addition of riboflavin in the form of fuller's-earth whey adsorbate (lot 2) resulted in additional growth, greater daily food consumption, and fewer deaths, the most striking result of the investigation was the demonstration of the value of nicotinic acid in the nutrition of the pig (lot 4). It produced striking results when added to the basal diet in the presence of riboflavin and thiamin, even in the absence of the filtrate factor (factor 2) and factor 1 (vitamin B₆). That these latter were deficient, however, was demonstrated by the further response secured when rice-bran filtrate (supplying largely the filtrate factor but some
factor 1) was given in addition to the supplements just listed (lot 5). A fuller understanding of the importance and function of the filtrate factor and nicotinic acid has, therefore, been obtained.

Unfortunately the value of the rôle of riboflavin and thiamin in the nutrition of the pig, was not so clearly demonstrated because both were studied in the purified diets, only in the absence of nicotinic acid and the filtrate factor. There was a response, however, when riboflavin was added to the basal diet even in the absence of the other factors of the vitamin-B complex. With respect to thiamin, however, some information was derived from the results of curative experiments (with lots 10 and 11). In these lots, thiamin, when added as a curative measure, produced striking improvements; this shows that thiamin was a limiting factor when skim-milk powder and whey powder were fed in these experiments.

Yeast effectively supplemented the basal diet; and whey powder, skim-milk powder, and autoclaved yeast were partially effective. From the data available, it is evident that these materials were partially deficient in one or more factors. They (whey powder, skim-milk powder, and autoclaved yeast) responded favorably to thiamin, but it is impossible to determine in the cases of the first two whether this was their only deficiency. In the case of autoclaved yeast, other deficiencies were evident from the fact that when the basal diet was supplemented with autoclaved yeast and thiamin, the results were not equivalent to those obtained from the basal diet plus raw yeast. Apparently lactose does not supply in appreciable quantities any of the factors of the vitamin-B complex necessary for normal growth (other than riboflavin, which was supplied with it in lot 9). Of the natural feedstuffs employed, yeast was the most effective in supplementing the basal diet. As a matter of fact, the pigs fed yeast required less food for a unit of increase in weight than those fed the basal diet supplemented with riboflavin, thiamin, nicotinic acid, and the filtrate factor, but the latter were much larger at the end of the experiment. If the two lots are compared at the same weight, those fed the supplemented purified diet consumed 270 pounds of gain, as compared with 284 pounds for those fed the basal diet supplemented with yeast.

The respiratory rate and body temperature of the pigs can be roughly correlated with the dietary deficiencies. This is not surprising, since thiamin (Lohmann and Schuster, 1937), riboflavin (Theorell, 1935), and nicotinic acid (Warburg and Christian, 1936) have been shown to be important in the enzyme systems necessary to oxidize carbohydrates and make energy available for normal metabolism. Deighton (1935) has shown too that fasting hogs have a lower temperature than hogs on feed.

Post-mortem findings indicate heart and intestinal disturbances par-
particularly on those diets lacking in nicotinic acid and the filtrate factor (factor 2).

The investigation shows a definite need on the part of the pig for the factors of the vitamin-B complex studied. It emphasizes the pig's need for nicotinic acid. The supplementary value of milk products and green pasture (Hughes, 1937; Hughes and Roadhouse, 1937) for swine may be explained in part by the nicotinic acid, riboflavin, and filtrate factor that they contain. Thus, as a result of this investigation, the supplementary value of milk products and green pasture becomes more intelligible.

SUMMARY

With a basal diet of rice screenings, purified casein, cod-liver oil in sufficient quantities to protect the animals against deficiencies of vitamins A and D, and an adequate salt mixture, young pigs developed a condition similar to pellagra in humans as described in the literature. Nicotinic acid when added to this diet in the presence of riboflavin and thiamin fully protected similar pigs of like weights and ages.

That the pig cannot grow normally without some of the factors in the vitamin-B complex has been demonstrated.

Experimental diets used in these studies affected body temperature and respiratory rate.

The basal diet plus untreated yeast at the level fed did not produce growth so rapid as the diet composed of the basal plus riboflavin, thiamin, nicotinic acid, and the filtrate factor (rice-bran filtrate).

The addition of whey powder or skim-milk powder to the basal diet produced less rapid growth than the basal diet plus riboflavin, thiamin, nicotinic acid, and filtrate factor. When, however, thiamin was added as a curative measure, increased appetite and body weight resulted immediately.

The addition of riboflavin (as fuller's-earth whey adsorbate) to the basal diet resulted in better appetites, more rapid growth, and a more economical increase in body weight.

The addition of the filtrate factor (as rice-bran filtrate) to the basal diet plus riboflavin, thiamin, and nicotinic acid resulted in more rapid growth and body weight.

Anorexia, impaired locomotion, and lowering of the body temperature and the respiratory rate were caused by diets deficient in one or more of the factors of the vitamin-B complex.

At autopsy, pigs on deficient diets showed flabby hearts, stomachs full of feed at death with some gastritis, a catarrhal exudate in the small in-
testines, sometimes ulcers in the large intestines. A few showed small spleens, while some had collapsed gall bladders. There was no evidence of serious skeletal deformities; and though the lungs of 1 or 2 were small, there was little or no evidence of pneumonia.

A purified diet was successfully used in these studies.

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