PHENYLALANINE REQUIREMENT OF THE WEANLING PIG AND ITS RELATIONSHIP TO TYROSINE

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SUMMARY

The weanling pig's requirement for total dietary aromatic amino acids (TAAA) and the portion of this requirement that can be efficiently provided by tyrosine were studied. An amino acid-fortified corn diet that was first-limiting in TAAA was used in all feeding trials. Chick and microbiological growth assays established that the corn contained .31% bioavailable phenylalanine and .48% bioavailable tyrosine. The optimum crude protein level for the basal corn diet (adjusted by varying the level of L-glutamic acid) was found to be 12% (i.e., 6.72% supplemental glutamic acid), since pigs gained faster and more efficiently at this protein level than at 8, 16 or 20% crude protein.

The TAAA study consisted of adding varied levels of L-phenylalanine and L-tyrosine to the diet containing 12% crude protein, .24% L-tyrosine and .16% L-phenylalanine. Gain and gain/feed were assessed in young pigs (10 to 20 kg) during a 16-day feeding trial. Optimal gain and gain/feed occurred at .56% TAAA. Furthermore, there was no depression in gain or gain/feed when tyrosine was increased from 43% to 49% of the TAAA in the diet containing .56% TAAA. This indicates that the weanling pig's requirement for TAAA is .56% of the diet at 12% protein and that 49% of this requirement can be supplied as tyrosine.

(Key Words: Swine, Amino Acids, Phenylalanine, Tyrosine, Glutamic Acid, Availability.)

INTRODUCTION

Both the phenylalanine requirement and the portion of the TAAA requirement that can be furnished by tyrosine have been carefully worked out for both the young chick and rat. Tyrosine can effectively provide 42.5% (Sasse and Baker, 1972) to 47% (Sasse and Baker, 1973) of the TAAA requirement for chicks and from 45% (Rama Rao et al., 1961; Stockland et al., 1971) to 55% (Womack and Rose, 1946) for rats.

A single study with swine by Mertz and coworkers (1954) suggested a dietary phenylalanine requirement for weanling pigs of .46%. From this same study, though never intended, inferences were drawn that tyrosine could furnish no more than 30% of the TAAA requirement. In spite of the seemingly marked disparity between this estimate and that for chicks and rats, together with the lack of a good basis for even making such an estimate from the Mertz et al. (1954) data, the 30% value for swine (N.R.C., 1973; Henry et al., 1976) has remained unchallenged. This has probably resulted in large part from two primary factors: a) feeding TAAA-deficient diets to swine necessitates large quantities of synthetic amino acids and is, therefore, very costly and b) deficiencies of TAAA seldom, if ever, occur in practice. Nonetheless, because of the need to know more about the phenylalanine-tyrosine interrelationship in swine for purposes of formulating research diets, and because of its implications in intermediary metabolism of not only swine but also man, a study was undertaken to ascertain the weanling pig's requirement for TAAA and, moreover, to determine the maximal percentage that can be safely furnished by tyrosine. In the process of developing a suitable assay system to answer these questions, data emerged that also contributed information concerning the proper ratio of nitrogen from indispensable and dispensable amino acids and also the availability of TAAA in corn.

EXPERIMENTAL PROCEDURE

The objective of the first phase of this study was to accurately determine the concentration...
of utilizable phenylalanine and tyrosine in ground corn since this was to constitute 50% of all subsequent experimental diets. A microbiological method (Henderson and Snell, 1948) was used to first determine the total phenylalanine and tyrosine content of the ground corn. Concentrations of .40% phenylalanine and .61% tyrosine were found. Weight gain of the young chick was then used as a measure of the biologically available total aromatic amino acids (table 1). This was accomplished by first deriving a regression equation with TAAA intake as the independent variable and weight gain as the dependent variable. The gain due to inclusion of corn in a TAAA-deficient diet was then used to calculate available TAAA from the regression equation. Both the diet composition and conduct of the availability assay were as previously described (Sasse and Baker, 1973; Yen et al., 1976). Results indicated an available TAAA concentration in corn of .79%. If 1.01% TAAA determined by microbiological assay is taken as the total quantity present, then availability of TAAA in corn ~ 78%. Applying this percent to the microbiologically determined phenylalanine and tyrosine concentrations gives values of .31% for bioavailable phenylalanine and .48% for bioavailable tyrosine. These values were therefore used in subsequent assays in which TAAA were being evaluated.

For the growth trials, 5- to 6-week-old crossbred pigs were used. They averaged approximately 10 days postweaning and 12 kg body weight when trials were initiated. A randomized complete block design involving four and six treatments in Trials 1 and 2, respectively, was employed. Blocks contained littermate pigs of similar weight and condition; assignment to treatment and pen was done randomly. The pigs were housed individually in raised cages with expanded metal floors in an environmentally controlled room. Feed was offered to appetite twice daily at 8 am and 5 pm. An equal weight of water was added to the feed at this time as well. Prior to initiating each trial, all pigs were fed a semi-purified pretest diet for 3 days so that any poorly performing pigs could be identified and replaced with littermates of comparable weight. Experimental diets were then offered on the fourth day following a 24-hr fast. The data from each trial were subjected to analysis of variance, and appropriate single degree-of-freedom comparison were made to test for treatment differences.

**Trial 1.** This trial was conducted to determine the crude protein level that would promote optimal performance when pigs were fed

| Added L-phenylalanine (%) | .25 | .30 | .35 | .25 |
| Added L-tyrosine (%) | .25 | .30 | .35 | .25 |
| TAAA (%) | .50 | .60 | .70 | .50 |
| Added ground corn (%) | .25 | .25 | .50 | 24.0 |
| TAAA intake (g) | .268 | .394 | .653 | .384 |
| Gain (g) | 16.0 | 29.9 | 49.7 | 39.7 |
| Gain/feed | .295 | .412 | .534 | .516 |

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*All additions to the basal diet were made at the expense of cornstarch. The basal crystalline amino acid diet (Sasse and Baker, 1973) was adequate in all known nutrients except TAAA. The TAAA requirement of the chick is .95% (Sasse and Baker, 1972).*

*Total aromatic amino acids. Refers to L-phenylalanine and L-tyrosine provided as crystalline amino acids.*

*Microbiological assay (Henderson and Snell, 1948) of ground corn revealed .40% L-phenylalanine and .61% L-tyrosine.*

*Data represent the mean of six replicate groups of seven chicks fed the experimental diets from day 8 to day 16 posthatching; average initial weight was 73 grams.*

*Regression of gain on TAAA intake for those treatments without added corn provided an equation: Y = -5.87 + 86.07X (r = .99) where Y = gain (g) and X = TAAA intake (g). Substituting into the regression equation 39.7 g gain for chicks fed the diet with 24% added corn gives an estimated TAAA consumption of .530 g of which .384 g was of crystalline origin and .146 g (by difference) was from corn. TAAA from corn as a percent of corn intake (i.e., .146 / (146 + 18.48 x 100) gives a value of .79% biologically available TAAA in corn.*
<table>
<thead>
<tr>
<th>Ingredient</th>
<th>% of diet</th>
<th>Amino acid mix</th>
<th>% of diet</th>
<th>% of diet</th>
</tr>
</thead>
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<tr>
<td></td>
<td></td>
<td></td>
<td>Trial 1</td>
<td>Trial 2</td>
</tr>
<tr>
<td>Cornstarch</td>
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<td>L-arginine•HCl</td>
<td>.08</td>
<td>.08</td>
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<tr>
<td>Dextrose</td>
<td>5.00</td>
<td>L-histidine•HCl•H₂O</td>
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<td>.17</td>
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<td>Wood cellulose</td>
<td>4.00</td>
<td>L-isoleucine</td>
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<td>.51</td>
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<tr>
<td>Ground yellow cornᵃ</td>
<td>50.00</td>
<td>L-leucine</td>
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<td>.24</td>
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<td>L-lysine•HCl</td>
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<td>Ca₃HPO₄</td>
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<td>DL-methionine</td>
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<td>.44</td>
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<td>L-threonine</td>
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<td>.45</td>
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<tr>
<td>Trace-mineralized saltᶜ</td>
<td>.50</td>
<td>L-tryptophan</td>
<td>.12</td>
<td>.12</td>
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<tr>
<td>KHCO₃</td>
<td>1.00</td>
<td>L-valine</td>
<td>.45</td>
<td>.45</td>
</tr>
<tr>
<td>Antibiotic premixᵈ</td>
<td>.50</td>
<td>L-phenylalanine</td>
<td>.23</td>
<td>var.</td>
</tr>
<tr>
<td>Vitamin mixᵉ</td>
<td>.20</td>
<td>Glycine</td>
<td>.61</td>
<td>.61</td>
</tr>
<tr>
<td>Ethoxyquinn</td>
<td>(50 mg/kg)</td>
<td>L-glutamic acid</td>
<td>var.</td>
<td>6.72</td>
</tr>
</tbody>
</table>

ᵃN.R.C. (1968) Ref. No. 4-02-995.
ᵇThe amino acid mix was formulated by taking the difference between amino acid values for corn and N.R.C. (1973) requirements of the pig for all indispensable amino acids plus an additional 10%.
ᶜBaker et al. (1970).
ᵈContained 22 g tylosin per kilogram premix.
ᵉVitamins (mg/kg diet): Thiamin•HCl, 100; niacin, 100; riboflavin, 16; Ca pantothenate, 20; vitamin B₁₂, .02; pyridoxine•HCl, 6; biotin, .6; folic acid, 4; inositol, 100; para-aminobenzoic acid, 2; menadione, 5; ascorbic acid, 250; and vitamin A acetate (250,000 IU/g), 10,000 IU; vitamin D₃ (200,000 IU/g), 600 IU.
the semipurified diet described in table 2. The basal ration without additional L-glutamic acid contained 8% crude protein from the corn and added indispensable amino acids. The crude protein (N x 6.25) content was then increased to 12, 16 and 20% by the addition of 6.72, 13.44 or 20.16% L-glutamic acid, respectively. All diets were brought to 100% with cornstarch. Diets were fed over a 14-day period with weight gain and feed intake monitored at 7-day intervals.

**Trial 2.** Using information obtained in trial 1, a basal phenylalanine-tyrosine deficient diet containing 12% protein was formulated (table 2). Graded levels of L-phenylalanine and L-tyrosine were then added to determine (1) the growing pig's requirement for TAAA and (2) the fraction of this requirement that could be efficiently provided by L-tyrosine. Diets 1 to 4 involved graded levels of added phenylalanine to provide TAAA concentrations of .40, .48, .56 and .64% of which 60, 50, 43 and 37%, respectively, came from tyrosine. Diet 5 contained added phenylalanine and tyrosine to provide .64% TAAA, 43% of which was tyrosine. In diet 6, additions of both amino acids were made such that 49% of the .56% TAAA came from tyrosine. Duration of the trial was 16 days.

**RESULTS AND DISCUSSION**

**Experiment 1.** Results from this trial (table 3) clearly demonstrated that addition of 6.72% L-glutamic acid optimized performance of the pigs. Pigs fed this diet gained faster and more efficiently (P<.01) than those fed diets with higher or lower levels. That pigs fed the 12% crude protein diet gained faster than those fed either 16% or 20% largely reflects the greater feed intake of pigs fed diets with 12% crude protein. The addition of 13.44% or 20.16% L-glutamic acid to the basal ration may have decreased the cation:anion ratio of the diet to an extent that palatability was reduced. Upon calculating the ratio of nitrogen contributed by indispensable amino acids to nitrogen contributed by dispensable amino acids (I/D ratio), it was found that diets 1 through 4 had ratios of 2.5/1, .67/1, .38/1 and .28/1, respectively. Previous work with young pigs (Mitchell *et al.*, 1968) had suggested that optimal nitrogen retention occurs when I/D ratios of close to 1/1 are present in purified diets containing large quantities of glutamic acid. Our study suggests that pig performance is excellent at an I/D ratio of .67/1 at 12% crude protein equivalent. Whether performance could be further improved at this same protein level by increasing the I/D ratio to 1/1 is a moot question.

**Experiment 2.** Pigs fed the diet with a 56% TAAA gained faster (P<.01) and more efficiently (P<.01) than those fed diets with lower TAAA levels (table 4). Also, no further response occurred when TAAA concentration was increased to .64%. Thus, it is clear that the growing pig's requirement for TAAA does not exceed .56% when a diet containing 12% crude protein is fed. The Mertz *et al.* (1954) requirement of .46% TAAA at 12.6% protein would therefore appear to be an underestimate, particularly since availability of TAAA in corn was not considered in making this estimate. The subcommittee on Swine Nutrition (N.R.C., 1973) set the TAAA requirement at .56% of the diet at 18% protein for pigs in the weight

### TABLE 3. PERFORMANCE OF PIGS FED GRADED LEVELS OF L-GLUTAMIC ACID (TRIAL 1)

<table>
<thead>
<tr>
<th>Diet no.</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
</tr>
</thead>
<tbody>
<tr>
<td>Added L-glutamic acid (%)</td>
<td>0</td>
<td>6.72</td>
<td>13.44</td>
<td>20.16</td>
</tr>
<tr>
<td>Crude protein (%)</td>
<td>8</td>
<td>12</td>
<td>16</td>
<td>20</td>
</tr>
<tr>
<td>Gainb,c (g/day)</td>
<td>275</td>
<td>387</td>
<td>294</td>
<td>226</td>
</tr>
<tr>
<td>Feedb,d (g/day)</td>
<td>622</td>
<td>728</td>
<td>575</td>
<td>525</td>
</tr>
<tr>
<td>Gain/feedb,c (g/kg)</td>
<td>441</td>
<td>533</td>
<td>507</td>
<td>425</td>
</tr>
</tbody>
</table>

*a* Crude protein calculated as total N x 6.25.

*b* Data based on the average of six individually-fed pigs; average initial weight was 11.9 kg; duration of trial was 14 days.

*c* Diet 2 > diets 1, 3, and 4 (P<.01).

*d* Diet 2 > diets 1, 3, and 4 (P<.05).
However, this value if applied to practical feed formulation, would also appear to be low, since availability of TAAA in feedstuffs that might be used was not considered.

The requirement estimated herein is for available TAAA at a protein level of 12%. Because an increase in protein level to 18% (N.R.C., 1973) would affect the TAAA requirement somewhat (cf., Baker et al., 1975), we would suggest an increase of .01% for each 1% increase in protein level, making the dietary requirement for available TAAA .62% at 18% protein. When an availability factor of 78% is applied (table 1) the TAAA requirement is calculated to be .79%. Hence, if a requirement for swine is to be extrapolated to practice from the work reported here,.79% would seem the most appropriate value. This value makes the TAAA requirement at 18% protein identical to the N.R.C. (1973) requirement for lysine at the same protein level, and this result is comparable to a similar relationship between TAAA and lysine requirements for chicks (Sasse and Baker, 1973).

Neither rate nor efficiency of gain differed significantly (P>.10) among pigs fed diets 3 through 6 (table 4). This result allows, therefore, a clear-cut estimate of the portion of the TAAA requirement that can be furnished by tyrosine. At .56% TAAA, a 49% tyrosine contribution (diet 6) was just as efficacious as a 43% contribution (diet 3). Our value of 49% is in line with estimates made on both rats and chicks (Womack and Rose, 1946; Rama Rao et al., 1961; Stockland et al., 1971; Sasse and Baker, 1972; 1973), but is markedly higher than the 30% estimate of N.R.C. (1973) based upon the pig data of Mertz et al. (1954).

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