Digestible Phosphorus in Poultry

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Animal Sciences
AGENDA

► Background Information on P

► Expression of P content of feedstuffs and the case for Digestible P in Poultry

► Towards a Standard Method for P Digestibility in Poultry???

► Other Digestible P Studies
PHOSPHORUS

- Essential mineral element

- Most vertebrate animals have a dietary requirement second only to calcium
PHOSPHORUS

▶ Skeletal Phosphorus

- 75% of the body P is stored in the skeleton
- Hydroxyapatite
- Framework as well as Reservoir of P
BODY PHOSPHORUS

Important roles in **BODY METABOLISM**

- **Component of Nucleic Acids** (DNA, RNA, ATP)
- **High-E compounds** (PEP, 1,3BPG, creatine phosphate)
- **Serves in Enzyme Regulation** (Phosphorylated proteins)
- **Phospholipids** (phosphoglycerides and sphingomyelin)
FEED PHOSPHORUS

► Inorganic Sources

► Organic Sources
FEED PHOSPHORUS

- **Inorganic**
  - Monosodium phosphate
  - Potassium phosphate
  - Mono-, di-, tri-calcium phosphates
  - Varying proportions of digestible phosphorus

\[
\begin{align*}
\text{Monosodium phosphate:} & \quad \text{HO-PO(OH)}^- \\
\text{Potassium phosphate:} & \quad \text{HO-PO(OH)}^- \\
\text{Mono-, di-, tri-calcium phosphates:} & \quad \text{Ca}^{2+}
\end{align*}
\]
Organic Phytate

- Organic form of P in plant
- Myo-inositol 1,2,3,4,5,6-hexakisphosphate complexed with mixed cations such as Ca, Zn, Mg, and Cu
FEED PHOSPHORUS

Phytate (Phytin)

- Constitutes up to 3% of many of the oilseed meals and cereals used in animal feeds

- Phytate P constitutes 50-80% of the total P in most Swine and Poultry feedstuffs of plant origin
DIETARY PHOSPHORUS

Dietary P supply **DOES NOT** guarantee total utilization

Dietary supply P ≠ Utilized P by animal

→ Excess P to Environment
ENVIRONMENTAL CONCERNS

► Immobile P in soil leads to build-up

► Erosion or run-off wash P into surface water
  ■ Algae & aquatic plants
  ■ Reduced dissolved O₂
  ■ Death & Decomposition of aquatic species
CURRENCY OF P UTILIZATION

► Feed Evaluation & Animals’ Requirement for Nutrients are Interdependent

► Underscores the importance of the development of appropriate systems for both
Poultry: Some Points for Awareness
EXPRESSION OF PHOSPHORUS (P) CONTENT OF FEEDSTUFFS

- Total P
- Non-Phytate P (nPP)
- Bioavailable P
  - Relative Available P
  - Digestible P
    - Total tract digestible P
    - Ileal digestible P

Proposed as a better approach to use
Non-Phytate P (nPP)
Corn and Canola Meal

<table>
<thead>
<tr>
<th></th>
<th>Total P, g/kg</th>
</tr>
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<tbody>
<tr>
<td>Corn</td>
<td>2.5</td>
</tr>
<tr>
<td>Canola meal</td>
<td>9.7</td>
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</table>

- A P evaluation system based on nonphytate P is NOT reflective of P digestibility in feed ingredients
- A portion of phytate-bound P is used by chickens
- In the study ~32% of phytate-bound P was utilized

Mutucumaran et al., 2014
Non-Phytate P (nPP)

Monosodium phosphate and Dicalcium phosphate

<table>
<thead>
<tr>
<th></th>
<th>Total P, g/kg</th>
</tr>
</thead>
<tbody>
<tr>
<td>MSP</td>
<td>250</td>
</tr>
<tr>
<td>DCP</td>
<td>190</td>
</tr>
</tbody>
</table>

▶ A P evaluation system based on nonphytate P is **NOT** reflective of P digestibility in feed ingredients
▶ A portion of nPP is **NOT** used by chickens
▶ In the study ~48% of nPP was **NOT** utilized

Adapted from: Shastak et al., 2012
EXPRESSION OF PHOSPHORUS (P) CONTENT OF FEEDSTUFFS

- Total P
- Non-Phytate P (nPP)
- Bioavailable P
  - Relative Available P
  - Digestible P
    - Total tract digestible P
    - Ileal digestible P

Proposed as a better approach to use
RELATIVE AVAILABLE PHOSPHORUS

Slope-ratio bioassays

▸ Response criterion
  • Bone ash
  • Bone P concentration
  • Bone breaking strength
  • Weight gain
  • Feed efficiency
RELATIVE AVAILABLE  PHOSPHORUS

Slope-ratio bioassays

- The expression of Response criterion to increasing concentrations of P from a test feedstuff RELATIVE to that from a reference source

- Common-intercept multiple linear regression procedure

\[ Y = a + bX_1 + cX_2 \]
RELATIVE AVAILABLE PHOSPHORUS

BW gain, g

\[ y = 40.4 \text{MSP} + 200 \]
\[ y = 37.9 \text{DCP} + 213 \]
Biological availability = 93.8%

Tibia ash, g/bone

\[ y = 0.13 \text{MSP} - 0.18 \]
\[ y = 0.08 \text{DCP} + 0.01 \]
Biological availability = 61.5%

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RELATIVE AVAILABLE PHOSPHORUS

Factors affecting Slope-ratio Bioassays

- Response criteria selected
- Reference OR standard phosphate selected
- Diet composition – Purified or practical
- Ca:P ratio
- Species and stage of growth/development
- Bioassay Length
RELATIVE AVAILABLE PHOSPHORUS

► Not additive in mixed feed
► Labor-intensive and expensive
► Does not provide estimates of the quantity of P voided by animal

Expression of the utilization and requirement on a DIGESTIBLE BASIS
EXPRESSION OF PHOSPHORUS (P) CONTENT OF FEEDSTUFFS

- Total P
- Non-Phytate P (nPP)
- Bioavailable P
- Relative Available P
- Digestible P
  - Total tract digestible P
  - Ileal digestible P

Proposed as a better approach to use
DIGESTIBLE PHOSPHORUS

- Relationship between:
  - Input (Diet)
  - Output (Excreta)
  - Output (Ileal digesta)

\[
P \text{ Digestibility, } \% = \frac{P \text{ Intake} - P \text{ Output}}{P \text{ Intake}} \times 100
\]
Excreta or Ileal Digesta Collection in Poultry

Ileal digesta from distal two-thirds (or one-half) 5 cm anterior to the ileocelecal junction
Poultry

P Intake

P output in ileal digesta

P output in excreta
Poultry
Is ileal \( P \) digestibility different from total tract retention of \( P \)?
Poultry

Total tract / Ileal P Digestibility, %

Soybean Meal

Canola Meal

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## Poultry

### Total tract / Ileal P Digestibility, %

<table>
<thead>
<tr>
<th></th>
<th>Total tract</th>
<th>Ileal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peanut flour</td>
<td>75</td>
<td>67</td>
</tr>
<tr>
<td>Peanut flour + Phytase</td>
<td>84</td>
<td>75</td>
</tr>
<tr>
<td>Low-Phytate SBM</td>
<td>31</td>
<td>75</td>
</tr>
<tr>
<td>Monocalcium phosphate</td>
<td>85</td>
<td>80</td>
</tr>
<tr>
<td>Dicalcium phosphate</td>
<td>81</td>
<td>74</td>
</tr>
</tbody>
</table>
Poultry

Total tract / Ileal P Digestibility, %

Black-Eyed Pea

Black-Eyed Pea + Phytase

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Poultry Total tract / Ileal P Digestibility, %

Corn 1

Corn 2
Poultry
Total tract / Ileal P Digestibility, %

Wheat

Barley

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Poultry
Total tract / Ileal P Digestibility, %

Rice Bran

Sunflower Meal
Poultry

Ileal \( P \) digestibility is different from total tract utilization of \( P \)
DIGESTIBLE P

TOTAL TRACT vs. ILEAL

Poultry

• Urine P contribution to excreta

• Total tract utilization
  ▪ Reflects Post-absorptive utilization
  ▪ Dietary supply of phosphorus and calcium relative to requirement and body status affect total tract utilization
DIGESTIBLE P

TOTAL TRACT vs. ILEAL

Poultry

• Utilization of P in feeds and Requirement for P should be expressed on an ILEAL DIGESTIBLE BASIS
DIGESTIBLE PHOSPHORUS

Digestible P in swine measured in feces as:

*Total tract digestibility*

Digestible P in poultry measured in ileal digesta as:

*Ileal digestibility*
Terminologies

Apparent digestibility...
AD

Standardized digestibility...
SD

True digestibility....
TD

Apparent digestibility does not account for confounding effect of endogenous losses
**ENDOGENOUS P LOSSES (EPL)**

Total EPL may be divided into 2 main components:

- **Basal losses**
  - Basal endogenous P losses

- **Specific losses**
  - Ileal endogenous P losses, g/kg of DMI
  - Specific endogenous P losses

- Dietary P content, g/kg

- P losses induced by specific feed ingredient characteristics
  - Dietary P dependent

- The minimum quantities of P inevitably lost by the animal
  - Dietary P independent
Definition

Apparent Digestibility

\[ = \frac{(\text{Intake} - \text{output})}{\text{Intake}} \]

Standardized Digestibility

\[ = \frac{(\text{Intake} - (\text{output} - \text{EPL}_{\text{basal}}))}{\text{Intake}} \]

True Digestibility

\[ = \frac{(\text{Intake} - (\text{output} - \text{EPL}_{\text{total}}))}{\text{Intake}} \]
SUMMARY

- Currency for P
  - Nutritionally adequate P
  - Additive in diet formulation
  - Minimize the excretion of phosphorus for environment
SUMMARY

- Relative available P and non-phytate P (nPP) served the industry well but may not be additive in diet formulation

- Digestible P should be considered a more appropriate currency for P
Poultry

Ileal $P$ digestibility is different from total tract retention of $P$

ILEAL DIGESTIBLE BASIS
Swine NRC (2012)

Recommend the use of Standardized Total Tract Digestible (STTD) P for Swine
Standard Method for P Digestibility in Poultry
Results of an international phosphorus digestibility ring test with broiler chickens

Rodehutscord et al., 2017
Poult. Sci. 96: 1679–168
Standard protocol (WPSA, 2013)

- Digestibility measured at the terminal ileum

- Defined details
  - Age of birds
  - Minimal number of replicates
  - Diet composition
  - P and Ca levels in diets
OBJECTIVE

Compare the Ileal Digestibility of P in soybean meal (SBM) among laboratories using the WPSA (2013) proposed method
MATERIALS AND METHODS

► 17 research stations from Europe and North America
► Ileal Digestibility of P in SBM following WPSA (2013) with regression analysis
MATERIALS AND METHODS

► The same diets mixed at one location in The Netherlands

► Diets shipped to all Participating Research Stations

► Chemical analysis of all Diets and Ileal Digesta at the same laboratory in Germany
## EXPERIMENTAL DIETS

### Table 1. Ingredient composition of diets

<table>
<thead>
<tr>
<th>Ingredient, g/kg</th>
<th>Diet</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Dehulled SBM</td>
<td>A</td>
<td>400.0</td>
<td>510.0</td>
<td>620.0</td>
</tr>
<tr>
<td>Cornstarch</td>
<td>B</td>
<td>448.6</td>
<td>336.6</td>
<td>224.6</td>
</tr>
<tr>
<td>Limestone</td>
<td>C</td>
<td>7.4</td>
<td>9.4</td>
<td>11.4</td>
</tr>
<tr>
<td>Titanium dioxide</td>
<td></td>
<td>5.0</td>
<td>5.0</td>
<td>5.0</td>
</tr>
<tr>
<td>Others</td>
<td></td>
<td>139</td>
<td>139</td>
<td>139</td>
</tr>
</tbody>
</table>
## EXPERIMENTAL DIETS

### Table 2. Nutrient composition of diets

<table>
<thead>
<tr>
<th>Nutrient, g/kg DM</th>
<th>Diet</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>A</td>
<td>B</td>
<td>C</td>
</tr>
<tr>
<td>Crude protein</td>
<td>231</td>
<td>288</td>
<td>339</td>
</tr>
<tr>
<td>Total P</td>
<td>3.02</td>
<td>3.80</td>
<td>4.59</td>
</tr>
<tr>
<td>Ca</td>
<td>4.57</td>
<td>5.65</td>
<td>6.78</td>
</tr>
<tr>
<td>$\text{InsP}_6-\text{P}^1$</td>
<td>1.63</td>
<td>1.97</td>
<td>2.28</td>
</tr>
</tbody>
</table>

$^1\text{InsP}_6-\text{P} = \text{phytic acid (myo-inositol hexakis-}$

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### SUMMARY OF TRIALS

<table>
<thead>
<tr>
<th>Station</th>
<th>Strain</th>
<th>Sex</th>
<th>No. of replicates</th>
<th>Birds per replicate</th>
<th>Age at start (d)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Ross 708</td>
<td>Male</td>
<td>8</td>
<td>8</td>
<td>20</td>
</tr>
<tr>
<td>2</td>
<td>Ross PM3</td>
<td>Unsexed</td>
<td>8</td>
<td>8</td>
<td>23</td>
</tr>
<tr>
<td>3</td>
<td>Ross 308</td>
<td>Unsexed</td>
<td>6</td>
<td>8</td>
<td>19</td>
</tr>
<tr>
<td>4</td>
<td>Ross 308</td>
<td>Unsexed</td>
<td>6</td>
<td>8</td>
<td>22</td>
</tr>
<tr>
<td>5</td>
<td>Ross 308</td>
<td>Male</td>
<td>6</td>
<td>10</td>
<td>22</td>
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<tr>
<td>6</td>
<td>Ross 308</td>
<td>Unsexed</td>
<td>6</td>
<td>8</td>
<td>23</td>
</tr>
<tr>
<td>7</td>
<td>Ross 308</td>
<td>Male</td>
<td>8</td>
<td>12</td>
<td>17</td>
</tr>
<tr>
<td>8</td>
<td>Ross 308</td>
<td>Male</td>
<td>6</td>
<td>12</td>
<td>20</td>
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<tr>
<td>9</td>
<td>Heritage 5632</td>
<td>Male</td>
<td>6</td>
<td>8</td>
<td>16</td>
</tr>
</tbody>
</table>
## SUMMARY OF TRIALS

<table>
<thead>
<tr>
<th>Station</th>
<th>Age at sampling (d)</th>
<th>BW before sampling (kg)</th>
<th>ADFI (g)</th>
<th>FCR (g/g)</th>
<th>Method of killing</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>25</td>
<td>1.12</td>
<td>123</td>
<td>1.63</td>
<td>CO₂ asphyxiation</td>
</tr>
<tr>
<td>2</td>
<td>28</td>
<td>1.86</td>
<td>174</td>
<td>1.22</td>
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<tr>
<td>3</td>
<td>27</td>
<td>1.46</td>
<td>138</td>
<td>1.32</td>
<td>Cervical dislocation</td>
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<tr>
<td>4</td>
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<td>1.84</td>
<td>163</td>
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<td>5</td>
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<td>9</td>
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<tr>
<td>Station</td>
<td>Strain</td>
<td>Sex</td>
<td>No. of replicates</td>
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<tr>
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<tr>
<td>10</td>
<td>Ross 708</td>
<td>Unsexed</td>
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<td>8</td>
<td>20</td>
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<tr>
<td>11</td>
<td>Ross 308</td>
<td>Unsexed</td>
<td>7</td>
<td>8</td>
<td>18</td>
</tr>
<tr>
<td>12</td>
<td>Ross 308</td>
<td>Male</td>
<td>8</td>
<td>8</td>
<td>16</td>
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<tr>
<td>13</td>
<td>Ross PM3</td>
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<td>14</td>
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<td>14</td>
<td>Ross PM3</td>
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<tr>
<td>15</td>
<td>Ross 308</td>
<td>Unsexed</td>
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<td>10</td>
<td>18</td>
</tr>
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<td>16</td>
<td>Ross PM3</td>
<td>Unsexed</td>
<td>10</td>
<td>4</td>
<td>12</td>
</tr>
<tr>
<td>17</td>
<td>Ross 308</td>
<td>Male</td>
<td>6</td>
<td>12</td>
<td>14</td>
</tr>
</tbody>
</table>
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<tbody>
<tr>
<td>10</td>
<td>25</td>
<td>1.11</td>
<td>109</td>
<td>1.44</td>
<td>CO₂ asphyxiation</td>
</tr>
<tr>
<td>11</td>
<td>23</td>
<td>1.03</td>
<td>93</td>
<td>1.42</td>
<td>CO₂ asphyxiation</td>
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<td>12</td>
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<td>1.32</td>
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<td>1.02</td>
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<tr>
<td>14</td>
<td>24</td>
<td>1.30</td>
<td>128</td>
<td>1.37</td>
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<td>15</td>
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<td>1.72</td>
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<tr>
<td>17</td>
<td>24</td>
<td>1.24</td>
<td>96</td>
<td>1.31</td>
<td>Sedanum and Ketamin</td>
</tr>
</tbody>
</table>
SUMMARY OF TRIALS

► Broiler strain (no. of stations)
  ▪ Ross 308 (10), Ross 708 (2), Ross PM3 (4), Heritage 5632 (1)
  ▪ Males (8 stations) Unsexed (9)
► Age
  ▪ Start: ranged from d 12 to 23
  ▪ Sampling: ranged from d 21 to 28
    • Minimum feeding period of 5 d
SAMPLING

1) The abdominal cavity was immediately opened
2) The digestive tract removed
3) The ileum dissected
   • Section between Meckel’s diverticulum and 2 cm anterior to the ileo-ceco-colonic junction
4) The digesta of the distal one-half of the ileum was obtained by
   • Flushing with water
   • Gentle squeezing

5) Digesta from all birds of one replicate cage were pooled into one sample
CHEMICAL ANALYSIS

► Ca, P, and Ti
  - By inductive coupled plasma optical emission spectrometer

► Conducted on Diets and Ileal Digesta Samples from all Stations at the same lab in Germany
Ileal P Digestibility of Diets, %

A: 67.3
B: 60.9
C: 55.3
Ileal Ca Digestibility of Diets, %

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Ileal Digestibility of P in SBM, %

\((P < 0.05)\)
Digestible Ca and P

\[ y = 0.97x + 0.66 \]

\[ r^2 = 0.69; \ Sy.x = 0.23 \]
REASONS FOR VARIATION

► Starter diets before experiment

- The concentration of P
  - Ranged from 0.5 to 0.7%
- The concentration of Ca
  - Ranged from 0.7 to 1.1%
- With or without phytase
- With or without coccidiostat
REASONS FOR VARIATION

► Experimental environment
  - Litter materials intake
    • Affect digestive tract development

► Epithelial phytase expression
  - Affected by genetic background
REASONS FOR VARIATION

► Sex of birds
  ▪ Male or unsexed

► Other differences among stations
  ▪ Growth
  ▪ ADFI
  ▪ Age at sampling
  ▪ Killing procedures
The protocol standardization must go beyond the standards already set

- Starter diet composition
- Management condition
Standard Method for P Digestibility
Ileal Digestibility of P in Soybean Meal, %

Range: 19 to 51%; Mean: 32%

Ileal Digestibility of P in Soybean Meal, %

Range: 25 to 41%; Mean: 32%

Ileal P Digestibility, %

11% CP / 22% CP Diet

Corn/Soy Diet
Ileal P Digestibility, %
Adaptation Length: 24 h/48 h

Corn

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Ca to P ratio: 2.1:1; 15.8:1 to 3.5:1 at 0.95% Ca; 1:1 to 0.5:1 at 0.13% Ca

Ileal P Digestibility, %

- Ca:P 2.1:1: 41%
- 0.95% Ca: 25%
- 0.13% Ca: 57%
Poultry Ileal P Digestibility, %

<table>
<thead>
<tr>
<th>Ca/P 0.6/0.45</th>
<th>Ca/P 1.0/0.45</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.6% Ca, 0.45% P</td>
<td>1.0% Ca, 0.45% P</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Ca/P 0.6/0.75</th>
<th>Ca/P 1.0/0.75</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.6% Ca, 0.75% P</td>
<td>1.0% Ca, 0.75% P</td>
</tr>
</tbody>
</table>
Ileal P Digestibility, %

- Delfos: 95%
- Calfos: 87%
- Bone meal: 78%
- MCP: 89%
- DCP: 82%
Additivity in Feed Formulation
**Assumption**

\[
\text{Supply of nutrients in a mixed diet} = \text{Sum of the supply of nutrients from the individual ingredients}
\]

**Additivity of nutrients** is fundamental in the formulation of diets.
ADDITIVITY IN FEED FORMULATION

• Corn (0.26% P) & SBM (0.71% P)
• Mix together in ratio of 50 : 40
• Mixed diet P: $0.13 + 0.28 = 0.41\%$
  (Additivity)
## ADDITIVITY IN FEED FORMULATION

<table>
<thead>
<tr>
<th></th>
<th>Corn</th>
<th>SBM</th>
<th>50% Corn</th>
<th>SBM</th>
<th>40% Corn</th>
<th>SBM</th>
<th>Mixed Diet</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total P, %</td>
<td>0.26</td>
<td>0.71</td>
<td>0.13</td>
<td>0.28</td>
<td>0.41</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Digestibility, %</td>
<td>26</td>
<td>39</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>0.147</td>
</tr>
<tr>
<td>Digestible, %*</td>
<td>0.07</td>
<td>0.28</td>
<td>0.035</td>
<td>0.112</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Apparent Digestible OR Standardized Digestible OR True Digestible
HYPOTHESIS

Ileal digestibility of P in MCP and DCP for broiler chickens are additive in a mixed diet

Ho: TIDP_D = TIDP_P
9 experimental diets

- 3 Levels of MCP
- 3 Levels of DCP
- 3 Mixed diets with 3 levels of MCP:DCP in ratio 1:1
<table>
<thead>
<tr>
<th>P source</th>
<th>MCP</th>
<th>DCP</th>
<th>Mixture</th>
</tr>
</thead>
<tbody>
<tr>
<td>TID of P, %</td>
<td>80</td>
<td>74</td>
<td>77</td>
</tr>
</tbody>
</table>

Mixture: \[ TIDP_{\text{Predicted}} = 76\% \]
True ileal digestibility of P, %

Predicted: 76.3
Determined: 77.4

\[ P = 0.54 \]

Predicted is not different from determined TID of P in MCP and DCP for broiler chickens.
TAKE HOME MESSAGE

► Protocol requires standardization of

- Pre-experimental diets
- Management conditions
  - Cages vs. Floor Pens
  - Adaptation Length
  - Flushing vs. Squeezing for Ileal Digesta
TAKE HOME MESSAGE

► Poultry

■ Ileal digestible P

■ Ileal digestible Ca

■ Need more data on ileal digestibility of feed ingredients for poultry