

# Assessment of dietary phosphorus adequacy for sows using urinary diagnostics

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## Take-Home Message

Precise information on phosphorus (P) requirements is fundamental for formulation of nutritionally adequate diets and to minimize environmental pollution associated with P oversupplementation. Urinary P excretion represents a sensitive response variable to evaluate dietary P intake and post-absorptive P utilization. In sows, the analysis of urinary P provides a non-terminal, minimally invasive method to determine P intake adequacy without interfering with maternal care of the offspring.

Mathematical modeling of P intake and urinary P excretion response curves provided estimates of P requirements during different stages of gestation and lactation. Additionally, urinary Ca to P ratio  $> 1.5$  was associated with P deficient diets, whereas a urine Ca to P ratio  $< 0.5$  reflected excessive P intake. Measurements of Ca to P ratio in random spot urine samples may be considered as a practical method to monitor P intake adequacy in sows. This approach has practical applications for mass screening of P intake adequacy in specific herds with consequent dietary adjustment that would favor a more efficient use of P.

## Introduction

Adequate P nutrition of sows during gestation and lactation is critical for optimal growth and development of the offspring, and maternal health and productive performance. Conventional methods applied to assess P requirements in young swine are impractical, or even unfeasible to implement in sows. Therefore, we have proposed and evaluated the use of urinary P excretion as an alternative method to determine P requirements (Grez-Capdeville and Crenshaw, in press), and to monitor P intake adequacy in reproducing sows (Grez-Capdeville and Crenshaw, 2021).

Kidneys are the main organs involved in the maintenance of P homeostasis through adjustments in the amount of P excreted via urine. Thus, measurements of urinary P excretion provide useful information on dietary P intake and post-absorptive P utilization. Previous studies with growing pigs have shown that urinary P excretion remains low and constant until P requirements are met, then urinary P excretion increases in proportion to additional P intakes (Rodehutscord et al., 1998; Ekpe et al., 2002; Gutierrez et al., 2015). These results underline the plausibility of using urinary P measurements as a practical method to assess P status in sows.

The objective of our studies was to use 24-hour urine collections to determine P requirements at different stages of gestation and lactation. Additionally, we evaluated the use of Ca to P ratio measured in spot urine samples as an indicator of P intake adequacy in reproducing sows.

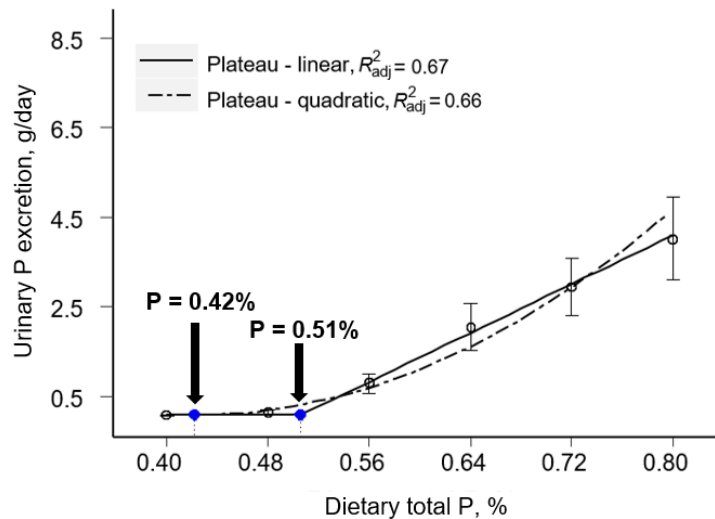
## Using 24-hour urinary P excretion to estimate P requirements

This experiment was conducted at the University of Wisconsin-Madison Swine Research and Teaching Center. Thirty-six sows (parity 3 to 7) were used to estimate total P requirements in mid (day  $77.1 \pm 2$ )

and late (day  $112.4 \pm 1$ ) gestation, and early (day  $4.5 \pm 1$ ) and late (day  $18.2 \pm 1$ ) lactation. At breeding, sows were assigned to one of six corn-soybean meal-based dietary treatments which provided incremental concentrations of total P (0.40, 0.48, 0.56, 0.64, 0.72, and 0.80%), and a constant Ca to total P ratio at 1.25:1 across diets. Sows were fed the experimental diets from day  $7.5 \pm 1$  after breeding until weaning (day  $26.6 \pm 1$ ).

Total 24-hour urine excretion was collected using bladder catheters in mid and late gestation, and early and late lactation. In parallel, spot urine samples were collected from the catheter tube on gestation day  $112.4 \pm 1$  and lactation day  $18.2 \pm 1$  at three different time points: early morning, late morning, and late afternoon. Total 24-hour and spot urine samples were analyzed to determine Ca and P concentrations, and Ca to P ratio was calculated.

Mathematical modeling of total P intake and urinary P excretion curves were used to predict P requirements at the different stages of gestation and lactation. Different regression models were tested, and the final model was selected based on statistical fit to the observed data and appropriate description of the biological response. Figure 1 shows the pattern of urinary P excretion in response to dietary total P intake in lactating sows, and results of estimates of P requirements using two different regression models.



**Figure 1.** Estimation of dietary total P requirements based on a single 24-hour urinary P excretion in early lactation. Minimum dietary total P requirements were predicted using the plateau-linear (0.51%) and plateau-quadratic (0.42%) models. Based on statistical fit and biological considerations, the selected model for estimating P requirements in lactation was the plateau-linear model.

Estimated total P requirements (%), feed intake, and the NRC (2012) nutrient composition values were used to express the requirements in daily amounts (g/day) of total P and standardized total tract digestible (STTD) P. Estimated daily dietary total P requirements in mid and late gestation were 10.3 g (6.0 g STTD P), and estimates for early and late lactation were 31.1 g (16.6 g STTD P) and 40.3 g (22.1 g STTD P), respectively.

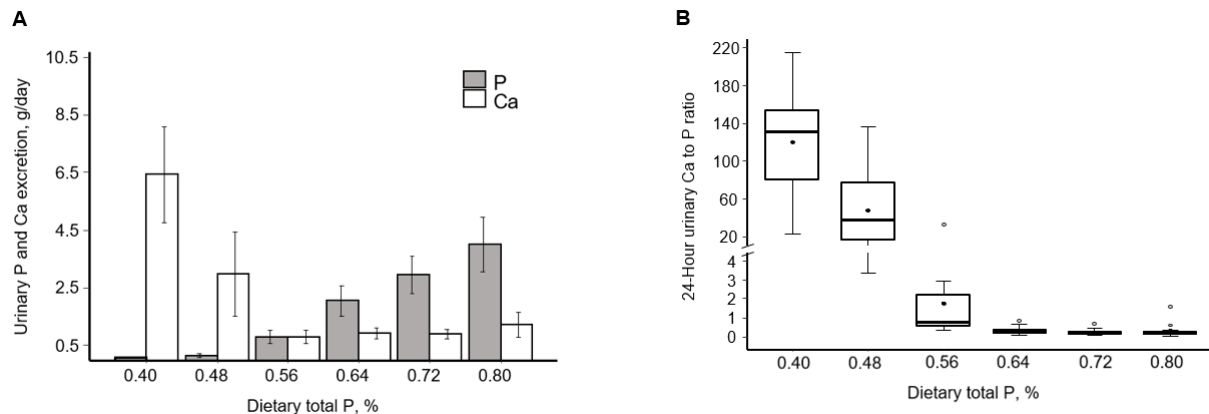
Outcomes from this experiment indicated that urinary P excretion is a sensitive response variable to dietary P intake. We conclude that the use of urinary P excretion is a reliable approach to estimate P requirements in reproducing sows. Estimated values for P requirements in gestation and lactation indicated that dietary P should not exceed current recommendations in dietary guidelines (NRC, 2012).

## Urinary Ca to P ratio to predict P intake adequacy

Additional findings from the described experiment showed an opposite response between 24-hour urinary P and Ca excretion to increments of P intake. As expected, urinary P excretion increased with dietary P supply. However, urinary Ca decreased as dietary P approached optimal levels, and remained constant at higher inclusions of P in the diet. Figure 2A shows results observed in early lactation. Similar patterns of urinary Ca and P excretion were observed in gestation and late lactation (data not shown).

Elevated urinary Ca excretions in pigs fed P-deficient diets have been reported in previous studies with (Vipperman et al., 1974; Fernández, 1995; Gutierrez et al., 2015). This response may be attributed to insufficient P to retain consumed Ca in the body, which results in excretion of circulating Ca via urine. The inverse response of urinary Ca and P with respect to dietary P intake presented a potential for use of urinary Ca to P ratio to determine P intake adequacy in sows.

The calculated urinary Ca to P ratio (Figure 2B) in lactating sows fed low concentrations (0.40 or 0.48% total P) was significantly higher than the ratio in sows fed above the estimated requirements ( $P < 0.01$ ;  $88 \pm 13$  vs  $1.4 \pm 0.8$ ).



**Figure 2.** (A) Urinary P and Ca excretion and (B) urinary Ca to P ratio in lactating sows fed different concentrations of dietary P

Statistical analysis based on receiver operating characteristic (ROC) curve was used to determine optimal cut-off values of urinary Ca to P ratio for predicting P intake adequacy. As a result, three different categories for P intake adequacy were defined (Table 1). These categories were then used to classify individual sows as being fed diets which were adequate, deficient, or excessive in P concentrations. Practical applications of this approach will allow appropriate dietary adjustments to balance between the P requirements and dietary supply. Assessments of P adequacy is essential to prevent negative effects of P deficiency on health and performance, but also to minimize P oversupplementation, which is particularly relevant when considering feed cost and environmental concerns associated with P overfeeding.

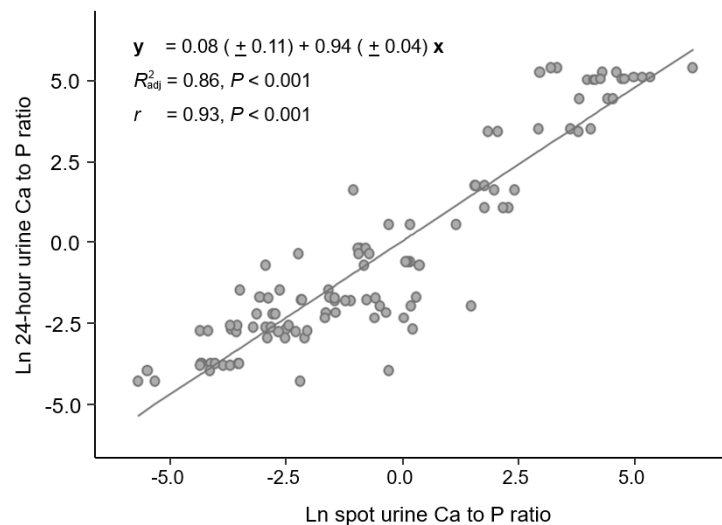
**Table 1.** Urinary Ca to P ratio cut-off values for predicting P intake adequacy in sows.

Urine Ca to P ratio	P intake category
> 1.5	Deficient
0.5 to 1.5	Adequate
< 0.5	Excessive

**Practical applications: use of spot urine samples to monitor P intake adequacy in sow herds**

Results in our study showed that measurements of 24-hour urinary Ca to P ratio is a useful method to predict P intake adequacy in sows. However, the collection of complete 24-hour urine samples may be inefficient when large number of animals are sampled. Single-void urine specimens, or spot urine samples, represent a more convenient method for urine collections in large-scale studies or for applications used to assess P adequacy of sow herds in commercial swine operations.

Statistical analyses were used to analyze the relationship and strength of association between urinary Ca to P ratio measured in 24-hour and spot urine samples (Figure 3). We found that regardless the physiological stage of sows (gestation and lactation), or time of spot sample collection (early morning, late morning, and late afternoon), urinary Ca to P ratio in spot urine samples explained most of the variability in 24-hour urinary Ca to P ratio ( $R^2_{Adj} = 0.86$ ). This indicates that urinary Ca to P ratio measured in random spot sample may be used as a predictor of P intake adequacy in reproducing sows.



**Figure 3.** Regression plot showing the relationship between natural log-transformed (Ln) 24-hour urine Ca to P ratio and Ln spot urine Ca to P ratio. The relationship was not affected by stage of gestation or lactation, or the time of day at which the spot urine sample was collected.  $R^2_{adj}$ , adjusted coefficient of determination;  $r$ , correlation coefficient.

Results from this study show that urinary Ca to P ratio predicts P intake adequacy in sows. However, further validation for the proposed cut-off values is required to verify relationships are consistent in herds with varied health status and genetic profiles, or in sows fed diets with different ingredient compositions. The current results provide a basis for future large scale screening of P adequacy and future approaches for development of precision feeding strategies applied to individual sows using electronic feeding systems.

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