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Evaluation of Different Calcium Sources on the Performance of Highly Prolific Lactating Sows

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Abstract

Improvements in sow productivity have raised questions regarding dietary mineral recommendations. Current calcium (Ca) levels and/or Ca sources might not support milk requirements of the larger litter in the modern sow. Therefore, four hundred and eighty mixed parity sows of a high prolificacy genetic line were used to evaluate the impact of the calcium source on the performance of highly prolific lactating sows. Sows were distributed in a completely randomized experimental design among six treatments containing different levels of inorganic Ca (INO) and organic calcium carbon-amino-phospho-chelate (CQT) inclusion. The sows were allocated to one of the six treatments represented by increased replacement ratios between sources of calcium: 100% INO; 100% CQT; 80% INO and 20% CQT; 60% INO and 40% CQT; 40% INO and 60% CQT; 20% INO and 80% CQT. Farrowing duration was not influenced (P>0.10) by the treatments, and averaged 185 minutes. Average daily voluntary feed intake did not differ (*P*>0.10) between treatments (5.54 kg d^{-1} on average). The lactation BW, backfat losses and chemical composition of body weight loss was not influenced (*P*>0.10) by the treatments. Litter size and average piglet weight at birth were not influenced (*P*>0.10) by the treatments (13.7 and 1.26 kg, on average). Litter weight gain, litter size and average piglet weight at weaning were also not influenced (*P*>0.10) by the treatments. The Treatments did not influence (*P*>0.10) estimated daily milk yield; which averaged 11.41 kg d^{-1}. The treatments tended to influence (P<0.10) urinary pH levels at day 7 of lactation, were 100% CQT sows had a lower pH value than other treatments (6.72 *vs.* 7.27). 100% CQT sows also showed a significantly lower pH level at d 14 and 21 when compared to the other treatments (6.44 *vs.* 7.09; *P*<0.05; and 6.48 *vs.* 7.14; *P*<0.01; respectively for d 14 and 21 of lactation). Free Ca and parathyroid hormone (PTH) serum levels were not affected (*P*>0.10) by treatments during lactation and averaged 1.38 mmol/L and 14.62 pg/ml, respectively. In conclusion, this experiment demonstrated that in diets for lactating sows an inorganic Ca source can be fully replaced by a more available Ca source (i.e., calcium carbon-amino-phospho-chelate), without negatively impacting the productive and reproductive performance of these animals or the performance of their litters.

Keywords: Sows; Calcium; Lactation; Milk production; Parathyroid hormone

Introduction

The productivity of sows has risen substantially during the past 10 years, mainly due to management and genetic advances and selection based on parameters such as litter size, weaning-estrus interval, and lactation efficiency. Although genetic advances have made sows more productive, they are more nutritionally demanding and less resistant to nutritional challenges. Mineral requirements have changed, as have other nutrient requirements. Average levels of digestible phosphorus (Pd) and calcium (Ca) recommended by the NRC in 2012 are higher than the levels practiced by the NRC in 1998.

Ca is enrolled with important metabolic functions and is closely related to Phosphorus (P) metabolism, regulation of Vitamin D absorption, parathyroid hormone (PTH) and Calcitonin [1]. Ca is also involved in extracellular functions such as blood clotting, maintenance and stability of cell membranes, structural integrity of bones and teeth, renal and respiratory function [2]. In addition to ATP metabolism, Ca affects muscle contraction, particularly during the parturition process, which is directly related to how long delivery takes and how many piglets are born alive, given that hypocalcemia reduces myometrium contractions. Furthermore, this mineral is also present in the sow´s milk, as one of the most important minerals secreted in milk [3].

The primary sources of Ca supplements for swine diets come from inorganic sources (e.g., rocks). Inorganic minerals have a structure that limits satisfactory absorption by the organism, therefore resulting in significant losses through feces [4]. Several studies have focused on finding new, more efficient mineral sources that are highly soluble, have a stable chemical structure, and are electrically neutral in the gastrointestinal tract, factors that would prevent reactions that could impair the absorption of other minerals [5]. For minerals to be absorbed, they must overcome an intestinal barrier, entailing factors such as physical and chemical conditions, pH and intestinal viscosity. In this context, researchers have focused on developing studies aiming towards the use of organic mineral sources. Organic minerals can be found in various forms, including compounds with amino acids, chelated amino acids, protein minerals and complex polysaccharide minerals [6]. Although some variance has been seen, it has been suggested that organic mineral forms offer higher bioavailability in animals [7].

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Some alternative organic ingredients have been explored to replace inorganic Ca sources (i.e., bone meal, meat and bone meal, ground shells and Ca from algae [5]. However, the main problem with using organic sources is the variation in composition, availability, and cost [8]. The use of chelated minerals comes as an alternative to inorganic sources of calcium. These are metallic ions chemically bound to an organic molecule, forming stable and highly bioavailable molecules and resulting in more efficient use by the animal [9].

Due to the evident importance of Ca for sows before farrowing and during lactation, the present study aimed to study the impact of a total and/or partial replacement of inorganic Ca with a more available source of Ca (i.e., calcium carbon-amino-phospho-chelate) on the productive performance of sows and their litters during the lactation period.

Materials and Methods

This study was conducted in compliance with and with approval from the Committee for Ethical Use of Animals at the Federal University of Paraná (UFPR) Agricultural Science Division, under the protocol number 016/2014.

Animals and experimental design

The study was performed in the facilities of a commercial sow unit, located in the South-eastern part of Brazil in the state of Minas Gerais, and was performed during summer, covering the period between January 2015 and March of 2015. According to the Köpen climate classification, the region is defined as Cwa (hot, temperate, rainy, and with a dry winter season and hot summer).

A total of 480 mixed parity sows of a high-prolificacy commercial genetic line (TOPIGS 20®) from four successive batches of 130 sows each were used in this study. Within each batch, sows were distributed in a completely randomized experimental design among six treatments containing different levels of inorganic Ca (INO) and organic calcium carbon-amino-phospho-chelate (CQT) inclusion according to parity order (1st, 2nd and 3rd-4th parity), body weight and backfat thickness at d 110 of gestation. The sows were allocated to one of the six treatments represented by increased replacement ratios between sources of calcium: 100% INO; 100% CQT; 80% INO and 20% CQT; 60% INO and 40% CQT; 40% INO and 60% CQT; 20% INO and 80% CQT. Each treatment consisted of 80 repetitions, and each sow was considered an experimental unit. Feed was formulated based on corn, soybean meal and soybean oil, and supplemented with industrial minerals, vitamins and amino acids to meet the requirements of this animal category according to recommendations in the feeding manual for the genetic breed (Table 1). For the ratios between essential amino acids and digestible lysine, the values indicated by Rostagno et al. were used as reference (Table 1) [10].

At 110 d of gestation, sows were weighed, backfat measured and transferred to the farrowing rooms and housed in farrowing crates (2.1 × 2.2 m) on fully slatted metal floor. During this period, the sows were fed the respective treatment diet as follows: day 110: 3.0 kg d^{-1}, 111: 2.80 kg d^{-1}, 112: 2.60 kg d^{-1}, 113: 2.40 kg d^{-1}, 114: 2.00 kg d^{-1}, and at d 115, if the sow had not yet farrowed, they were offered 1.50 kg d^{-1} until the farrowing day. The animals had *ad libitum* access to water throughout the entire experimental period.

Twenty-four hours after farrowing, all sows were again weighed, had their backfat measured (P2). Sows were then submitted to a step-up feeding regime to stimulate a gradual feed intake increase up to day 7 post-farrowing, starting with 2 kg on day 1 post-farrowing and reaching 8 kg d^{-1} on day 7. The allowance increased by 1 kg each day. This feeding management was applied to avoid over-consumption at the beginning of lactation and agalactia problems. After d 7 sows were fed 2 kg+0.5 kg $piglet^{-1}$ d^{-1}.

After birth, piglets were handled for tooth clipping, umbilical cord treatment and ear tagged for labelling. On d 3, they received an intramuscular injection of 200 mg of iron dextran. As necessary, cross-fostering was conducted within the first 48 h after birth to standardize litter size at 13 piglets. On d 10, male piglets were castrated. Creep housing equipped with infrared lights provided supplemental heat for the piglets during the lactation period. The day prior to weaning (i.e., d 24), sows were allowed 5 kg of feed (i.e., at least 1.5 kg lower than their usual feed intake) to standardize consumption for all sows for determination of sow weight at weaning. At weaning, sows were again weighed and backfat measured and moved to a breeding facility and were presented to a mature boar twice daily to detect onset of standing estrus. Sows were inseminated when positive to the back-pressure test. During the weaning-to-estrus interval, all sows were submitted to the same feeding management, receiving 3.0 kg d^{-1} of their respective lactation diet.

Collected measurements and parameters

Variations in environmental conditions inside the farrowing barns were recorded daily using a thermo-hygrometer kept in an empty cage in the middle of the building, at the average midpoint of the animals' height. Temperature and humidity data were recorded daily at 0700; 1200; and 1700. Sows were weighed using a digital scale (Líder Balanças Ltda., Mod. LD 2000E, Araçatuba, SP, Brazil), and backfat thickness was measured at P2 (65 mm from the dorsal line) using ultrasound equipment (Renco Lean-Meater, Renco Corporation, Minneapolis, USA) at d 110, 24 h post-farrowing and at weaning in order to determine body weight and backfat thickness variation. The following litter parameters were collected at farrowing: total number of piglets born, born alive, stillborn, and mummies. Piglets were individually weighed using a digital scale (Líder Balanças Ltda., Mod. B150, Araçatuba, SP, Brazil) 24 h post-farrowing and at weaning to determine litter birth and weaning weights, and daily weight gain during lactation. All dead piglets during lactation were weighed in order to properly estimate growth rates and daily milk production.

Every morning, feed refusals were collected, and fresh feed was immediately distributed once per day between 0700 and 0800. Feed consumption was determined as the difference between feed allowance and the refusals collected on the next morning. Every day, one sample of feed and feed refusals were collected daily for DM content measurement, and successive samples were pooled and stored at 4°C for further analyses. The feed samples were analyzed for DM, ash, fat content (AOAC, 1990) and CP (N × 6.25 for feed) according to Dumas method (AOAC, 1990) and for crude fiber and for cell wall components (NDF, ADF, and ADL) according to van Soest and Wine (1967) at the Animal Nutrition Laboratory of Federal University of Paraná Animal Nutrition Laboratory (Curitiba, Brazil).

The impact of treatments on farrowing duration was evaluated by recording the time farrowing began, which was defined as the moment the first placenta break, and the time farrowing ended, which was defined as the moment the sow expelled the last placenta. To analyze the sows' urinary pH levels, urine samples were collected 24 hours after farrowing and at d 7,14 and 21 of lactation, in a predetermined subgroup of 20 sows per treatment. The subgroup consisted of sows chosen according to parity order to be the most representative samples of the treatments.

	Treatments[1]					
Ingredients	**100% INO**	**100% CQT**	**80% INO 20% CQT**	**60% INO 40% CQT**	**40% INO 60% CQT**	**20% INO 80% CQT**
Corn, kernel	52.38	52.27	52.36	52.34	52.32	52.30
Soybean meal 46%	25.00	25.00	25.00	25.00	25.00	25.00
Soybean hull	13.85	13.85	13.85	13.85	13.85	13.85
Soybean oil	4.94	4.94	4.94	4.94	4.94	4.94
Dicalcium phosphate	2.28	-	1.81	1.35	0.88	0.41
Calcitic limestone	0.30	-	0.25	0.19	0.14	0.08
Chelated Calcium[5]	-	2.70	0.54	1.08	1.62	2.16
Salt	0.48	0.48	0.48	0.48	0.48	0.48
Mineral mixture[2]	0.15	0.15	0.15	0.15	0.15	0.15
Vitamin mixture[3]	0.15	0.15	0.15	0.15	0.15	0.15
L-Lysine HCL 78%	0.24	0.24	0.24	0.24	0.24	0.24
L-Threonine	0.08	0.08	0.08	0.08	0.08	0.08
L-Tryptophan	0.02	0.02	0.02	0.02	0.02	0.02
L-Valine	0.06	0.05	0.06	0.06	0.06	0.06
DL-Methionine	0.02	0.02	0.02	0.02	0.02	0.02
TOTAL	100	100	100	100	100	100
Analyzed composition as fed[4]						
Metabolizable Energy, Mcal/kg	3.40	3.40	3.40	3.40	3.40	3.40
Crude protein %	17.1	17.1	17.1	17.1	17.1	17.1
Digestible Lysine %	0.95	0.95	0.95	0.95	0.95	0.95
Digestible Met+Cys %	0.51	0.51	0.51	0.51	0.51	0.51
Digestible Threonine %	0.61	0.61	0.61	0.61	0.61	0.61
Digestible Tryptophan %	0.18	0.18	0.18	0.18	0.18	0.18
Digestible Valine %	0.74	0.74	0.74	0.74	0.74	0.74
Digestible Arginine %	1.00	1.00	1.00	1.00	1.00	1.00
Total Calcium %	0.86	0.86	0.86	0.86	0.86	0.86
Total Phosphorous %	0.73	0.63	0.71	0.69	0.66	0.64
Digestible Phosphorus %	0.39	0.39	0.39	0.39	0.39	0.39
Digestible Ca:P ratio %	2.2	2.2	2.2	2.2	2.2	2.2
Sodium %	0.22	0.22	0.22	0.22	0.22	0.22

[1]100% Inorganic Calcium source (INO); 100% organic calcium source (calcium carbon-amino-phospho-chelate = CQT); 80% INO and 20% CQT; 60% INO and 40% CQT; 40% INO and 60% CQT; 20% INO and 80% CQT. [2]Contents in 1 kg of feed: Iron, 100 mg; Copper, 10 mg; Cobalt, 1 mg; Manganese, 40 mg; Zinc, 100 mg; Iodine, 1.5 mg; and excipient q.s. [3]Contents in 1 kg of feed: Vit A - 8000 IU; Vit D3 - 1200 IU; Vit E - 20 IU; Vit K3 - 2 mg; Vit B1 - 1 mg; Vit B2 - 4 mg; nicotinic acid - 22 mg; pantothenic acid - 16 mg; Vit B6 - 0.50 mg; Vit B12 - 0.020 mg; folic acid - 0.4 mg; biotin - 0.120 mg; choline - 400 mg; and antioxidant – 30 mg. [4]Composition calculated per TOPIGS (2012). [5]Contents: 25% Ca and 12% P (digestible).

Table 1: Composition of the lactation experimental diets.

Sampling was standardized within 2 hours after meals, and a pH meter was used to measure sample pH immediately after collecting. On the d 14 and 21, blood samples were collected from the same subgroup of sows from which urine was collected. The sows were immobilized in their farrowing crates, and then 10 ml of blood were drawn from the jugular artery using a 40 mm × 12 mm needle. Blood samples were then sent to a laboratory, where they were centrifuged for 10 minutes at 3500 rpm for subsequent analysis of free Ca using the Selective Ion method, with ISELAB equipment – SL 0014 – DRAKE, and parathyroid hormone (PTH) using the Automated Chemiluminescent method, with Siemens IMMULITE 2000 equipment. The following parameters were assessed: time of farrowing duration, total feed intake of sows considering pre-farrowing and lactation, number of total born, live born, stillborn and mummified fetuses, birth weight of piglets, variations in the sows' physical conditions, variations in body composition, estimated milk production, piglet and litter weight gain, sow urinary pH levels, and circulating levels of free Ca and PTH (parathyroid hormone).

Calculations and statistical design

Daily maximum, minimum, mean, and variance of daily ambient temperatures and relative humidities were averaged and analysed for the entire experimental period. Body protein, fat, and energy contents at farrowing and at weaning were estimated according to the equations of Dourmad et al. [11]. Protein, lipid, and energy losses during lactation were estimated as the difference between calculated values determined at weaning and farrowing. Average daily milk production estimation was based on litter growth rate and size during lactation, according to the equations of Noblet and Etienne [12]. Data were submitted to normality tests and analysed using the generalized linear model procedure (GLM) and the mixed linear model (PROC MIX procedure) of SAS statistical package (SAS Inst., Inc, Cary, NC; version 9.2) and differences were considered significant at the $P<0.05$ level. The effects of parity order (O), batch (G) and treatments during lactation (TL) and litter size and weight as a covariate effect were included in the statistical model. Analyses were conducted using the following statistical model:

$$Y_{ijk}=\mu+A_i+B_j+C_k+D_l+E(X_{ijkl}-X)+F(Z_{ijkl}-Z)+e_{ijkl};$$

where μ=general average, A_i=fount type of Ca, B_j=parturition order, D_l=back fat, C_k=group effect, X_{ijkl}=observed value of the covariate "sow weight", X=average of the covariate "sow weight", E=regression coefficient between the covariate (X) and the response variable (Y), Z_{ijkl}=observed value of the covariate "litter weight", X=average of the covariate "litter weight", F=regression coefficient between the covariate (Z) and the response variable (Y), and e_{ijkl}=incidental residual effect of observation.

Results

The average maximum and minimum environmental temperatures and average maximum and minimum relative humidity recorded during the experimental period were 32.9 and 19.3°C and 86.6 and 58.8%, respectively. A total of 29 sows were removed from the experiment due to low litter size at weaning (<9 piglets) and/or health problems. According to the experimental design, the average parity order was 3.17 and did not differ (P>0.10) between treatments. No differences in lactation length were observed between treatments (24.5 d on average). Farrowing duration was not influenced (P>0.10) by the treatments, and averaged of 185 minutes.

Average daily voluntary feed intake did not differ (*P>0.10*) between treatments (5.54 kg d^{-1} on average; Table 2). The lactation BW and backfat losses were not influenced (*P*>0.10) by treatments (9.51 kg and 2.83 mm, on average), as shown in Table 3. The chemical composition of body weight loss was not influenced (*P*>0.10) by the treatments (7.64 kg; 2.36 kg; and 280 MJ; respectively for body protein, lipids and energy losses; Table 2).

Litter size and average piglet weight at birth were not influenced (*P*>0.10) by the treatments (13.7 and 1.26 kg, on average; Table 3). Litter weight gain during lactation was not influenced (*P*>0.10; Table 3) by the treatments and averaged 2.39 kg d^{-1}. Litter size and average piglet weight at weaning were also not influenced (*P*>0.10) by the treatments (12.2 and 6.29 kg on average, respectively; Table 3). The Treatments did not influence (*P*>0.10; Table 3) estimated daily milk yield; which averaged 11.41 kg d^{-1}.

The treatments tended to influence (P<0.10) urinary pH levels at day 7 of lactation, where treatment 2 sows presented a lower pH value than other treatments (6.72 *vs.* 7.27; Table 4). Treatment 2 sows also showed a significantly lower pH level at d 14 and 21 when compared to the other treatments (6.44 *vs.* 7.09; *P*<0.05; and 6.48 *vs.* 7.14; *P*<0.01; respectively for d 14 and 21 of lactation; Table 4). Free Ca and PTH serum levels were not affected (*P*>0.10; Table 4) by treatments during lactation and averaged 1.38 mmol/L and 14.62 pg/ml, respectively.

Discussion

The effects of high temperatures on the performance of lactating sows is well established [13]. In tropical climate conditions, the temperature observed in different seasons is always higher than the upper limit of the sows' thermoneutral zone (i.e., 22°C; Quiniou and Noblet, 1999). While conducting this study, the average maximum temperatures (26.1°C) exceeded 22°C. Therefore, lactating sows suffered from heat stress most of the time during our experiment.

There is quite extensive research on the benefits of chelated mineral sources in animal nutrition, in particular for non-ruminants [14-17]. These associated mineral sources have been reported to improve mineral absorption, and when replacing traditional inorganic mineral sources are capable of maintaining or improving performance [14,15]. However, the vast majority of studies focus their work on "trace" minerals like Cu, Zn, Fe, Mn and Se. Research focusing on the study of Ca sources chelated to organic molecules in animal feed is rare [15] and in particular those focusing on their effect on reproductive and productive aspects of pigs are nonexistent.

During the farrowing period, intramuscular Ca reserves are extremely important for uterine contraction and expulsion of the fetus, but over the course of the parturition the uterine musculature response tends to decrease due to muscle fatigue and decreased intramuscular Ca reserves, sometimes requiring the application of hormones like oxytocin or medication like Ca gluconate to prevent increased total

Variables	Treatments							
	100% INO	100% CQT	80% INO 20% CQT	60% INO 40% CQT	40% INO 60% CQT	20% INO 80% CQT	RSD[2]	Statistics[3]
Number of sows	76	74	73	76	75	77		
Parity order	3.1	3.1	3.1	3.1	3.2	3.2	1.2	
Lactation duration, d	24.7	24.6	24.4	24.5	24.5	24.3	1.2	
Av. daily feed intake (d 1 to weaning), kg d^{-1}	5.51	5.54	5.54	5.61	5.38	5.55	0.73	
Body weight, kg								
At farrowing,	226.6	234.1	224.1	222.0	224.9	227.2	29.1	OP***, G***
At weaning	219.2	225.2	218.8	219.7	223.7	220.3	3.2	OP*, G†
Weight loss	-7.41	-8.91	-5.34	-5.14	-5.16	-6.94	3.2	OP***, G***
Backfat thickness, mm								
At farrowing,	15.4	16.0	15.8	15.7	15.7	15.5	3.2	OP***
At weaning	12.6	13.1	12.9	12.9	12.6	12.8	2.8	OP*
Backfat loss	-2.8	-2.9	-2.9	-2.8	-3.1	-2.6	1.9	OP**, G*
Chemical composition of body weight loss[4]								
Protein, kg	-6.09	-10.35	-4.46	-2.02	-1.21	-2.72	2.38	OP***, G***
Lipids, kg	-2.19	-3.26	-1.63	-0.90	-0.97	-0.95	1.16	OP***, G**
Energy, MJ	-250.6	-326.7	-224.2	-178.1	-176.1	-183.5	59.5	OP***, G***
Weaning to insemination, d	3.8	4.0	3.8	3.8	3.6	3.9	0.5	
Parturition duration, min.	178.9	166.9	205.4	176.7	195.4	184.5	71.6	

[1]100% Inorganic Calcium source (INO); 100% organic calcium source (calcium carbon-amino-phospho-chelate=CQT); 80% INO and 20% CQT; 60% INO and 40% CQT; 40% INO and 60% CQT; 20% INO and 80% CQT. [2]RSD = Residual standard deviation. [3]Obtained from the analysis of variance (GLM including the effects of the parity order (OP), treatment during lactation (TL) and sow batch (G)). [4]Calculated based on the equations published by Dourmad et al. (1997). Protein (kg)=2.28 (2.22)+0.178 (0.017) × PV empty –0.333 (0.067) × P2 (*RSD=1.9*); lipids (kg) = −26.4 (4.5) + 0.221 (0.030) × PV empty + 1.331 (0.140) × P2 (*RSD = 6.1*); Energy (MJ) =−1.075 (159)+13.67 (1.12) × PV empty +45.98 (4.93) × P2 (*RSD=208*). PV empty (kg) = a × $PV^{1,013}$ (kg), where a = 0.912 at birth and a=0.905 at weaning. P2=P2 backfat thickness (mm) PV = live weight (kg). ***P<0.001; **P<0.01; *P<0.05; †P>0.05.

Table 2: Impact of Calcium source on the productive performance of sows during 24 days of lactation (least-square means)[1].

Variables	Treatments[1]							
	100% INO	100% CQT	80% INO 20% CQT	60% INO 40% CQT	40% INO 60% CQT	20% INO 80% CQT	RSD[2]	Statistics[3]
Number of litters	76	74	73	76	75	77		
Lactation duration, d	24.7	24.6	24.4	24.5	24.5	24.3	1.2	
Litter size								
At birth (liveborn)	13.69	13.71	13.47	13.93	13.81	13.54	1.35	
At weaning	12.04	12.41	12.06	12.18	12.00	12.05	1.22	OP***
Average piglet weight, kg								
At birth (liveborn)	1.27	1.33	1.27	1.21	1.24	1.24	0.20	OP**
At weaning	6.39	6.24	6.26	6.35	6.29	6.22	0.83	OP***
Litter weight, kg								
At birth	17.4	18.2	17.1	16.8	17.1	16.8	2.89	OP**
At weaning	76.6	77.1	75.1	77.1	75.1	74.5	10.8	
Litter weight gain, kg/d	2.40	2.38	2.38	2.46	2.36	2.38	0.43	
Milk production[4], kg/d	11.38	11.47	11.24	11.88	11.22	11.22	2.45	

[1]100% Inorganic Calcium source (INO); 100% organic calcium source (calcium carbon-amino-phospho-chelate = CQT); 80% INO and 20% CQT; 60% INO and 40% CQT; 40% INO and 60% CQT; 20% INO and 80% CQT. [2]RSD = Residual standard deviation [3]Obtained from the analysis of variance (GLM) including the effects of the parity order (OP), treatment during lactation (TL) and sow batch (G). [4]Daily milk production calculated based on the litter's weight gain, litter size, and milk MS (19%) using the Noblet and Etienne equation (1989). Milk yield (kg/d)=([0.718 × GPD − 4.9] × Number of piglets) / 0.19. ***P<0.001; **P<0.01.

Table 3: Impact of Calcium source for sows on the productive performance of their litters during 24 days of lactation (least-square means).

Variables	Treatments[1]							
	100% INO	100% CQT	80% INO 20% CQT	60% INO 40% CQT	40% INO 60% CQT	20% INO 80% CQT	RSD[2]	Statistics[3]
Number of sows	20	20	20	20	20	20		
pH								
On d 1	6.60	6.45	6.71	6.72	6.32	6.33	0.76	G*
On d 7	7.14	6.72	7.18	7.08	7.53	7.43	0.58	TL†
On d 14	7.25[a]	6.44[b]	7.26[a]	7.18[a]	6.91[a]	6.85[a]	0.71	TL*
On d 21	7.13[a]	6.48[b]	7.06[a]	6.98[a]	7.27[a]	7.28[a]	0.52	TL**
PTH[4], pg/ml								
On d 14	18.81	17.08	19.11	19.03	22.50	21.03	25.47	
On d 21	10.97	11.65	6.09	8.79	15.63	4.78	8.26	G*
Free Ca, mmol/L								
On d 14	1.40	1.36	1.38	1.35	1.36	1.36	0.07	G*
On d 21	1.41	1.39	1.36	1.42	1.38	1.37	0.09	

[1]100% Inorganic Calcium source (INO); 100% organic calcium source (calcium carbon-amino-phospho-chelate=CQT); 80% INO and 20% CQT; 60% INO and 40% CQT; 40% INO and 60% CQT; 20% INO and 80% CQT. [2]RSD=Residual standard deviation. [3]Obtained from the analysis of variance (GLM) including the effects of the parity order (OP), treatment during lactation (TL) and sow batch (G). [4]Parathyroid hormone. **P<0.01; *P<0.05; ∗P=0.0609.

Table 4: Impact of Calcium source for sows on the PTH and free Ca plasma levels and on pH of urine during 25 days of lactation (least-square means).

parturition time and consequently the incidence of higher stillborn rates. According to Frend et al. the increase in parturition duration from three to eight hours results in an increased stillborn piglet rate from 18.2 to 61.1%. On the other hand, Cunha et al. who evaluated total, ionic calcium and albumin serum levels in pregnant mixed-parity sows and their suckling piglets during lactation, found that the sows' average parturition duration was around 164 minutes, lower than the values found in our study (i.e., 185 min) [18]. This difference between studies can be attributed to the fact that the sows used by the previous authors farrowed on average 10.7 liveborn piglets while in our study the sows farrowed on average of 13.7 liveborn piglets, justifying the need for a longer delivery period. One benefit of using chelated Ca in the nutrition of sows is the role of Ca in activating oocytes during conception. Kishigami and Wakayama demonstrated that Ca chelation may induce meiosis activation after the second metaphase in the oocytes of mammals [19]. Studies on nutritional sources of chelated Ca are still rare and its responses in animal metabolism and performance are still unknown. More studies must therefore be conducted to better clarify its effects.

The lactation stage has often been investigated in swine nutrition, given that milk yield of sows is directly related to piglet weight gain. The lack of difference on average weight gain of litters and sow milk production between the different levels and sources of Ca is in agreement with the findings of Komegay and Kite and Kornegay et al. who evaluated the use of different Ca and P levels for sows and found no differences on litter size, birth weight nor weaning weight [20,21]. Similarly, Maxon and Mahan, studying first and second parity sows for two consecutive reproductive cycles using a Ca to P ratio of 1.3:1, also found no difference in litter parameters [22]. However, these same authors found that bone composition, litter size and weight are more closely related to sow body Ca than to dietary Ca.

In nature there are different forms of chelation between Ca and organic molecules [23] and their molecular nature and structure are still being studied, requiring new techniques in the field of biotechnology [24]. However, these chelation reactions do not always result in molecules with high nutritional bioavailability [25]. However, the association of Ca with organic molecules in the form of proteinate

has shown this high bioavailability [15]. One of the points that should be noted in the methodology when studying chelated minerals is the recommended nutritional level. Many studies indicate that, due to increased bioavailability, lower levels of chelated minerals (compared to saline sources) would be sufficient to meet the animals' requirements [5,14,16,17]. Perhaps this explains the absence of difference in the performance of sows fed with organic and inorganic Ca sources in the present study. Therefore, one may hypothesize that due to the fact that in our study we did not work with different dietary Ca levels, the sows had their requirements met independent of Ca source. Similarly to this work, Acda and Chae, that studied the performance of sows fed chelated trace minerals (Fe, Cu, Zn and Mn), observed no improvement in reproductive and productive responses in the animals fed with high levels of chelated minerals [14]. These researchers also observed similar results among animals fed with high levels and reduced levels of chelated minerals, confirming our hypothesis. Similar results were also observed by Martin et al. these authors tested saline and chelated sources of trace minerals in diets for piglets in the nursery phase, and did not find any performance improvement in the animals fed with organic when compared to inorganic sources [26].

Differently from our findings, Mello et al. evaluating the supplementation of piglet nursery diets using a chelated trace source, found an optimal level by replacing the saline source for the organic source at approximately 25% substitution [13]. Working with Ca proteinate supplementation for turkey hens, Grimes et al. reported greater hatchability values in the incubated eggs [15]. The authors attributed these findings to greater Ca bioavailability when provided in the form of proteinate in diets. This greater bioavailability resulted in improved deposition in egg shells, thus giving embryos a lower late mortality rate during incubation. However, still there are contradictory researches on the effect of mineral source types, for instance, Creech et al. studied the nutritional response of chelated trace minerals on the performance of nursing gilts and found similar performance between gilts fed with low levels of inorganic and chelated sources and those supplemented with appropriate levels of inorganic sources [5]. Trace mineral deficiency can be difficult to detect, due to the fact that these may be supplemented via the presence of raw materials used as ingredients to prepare feed.

Ca is important not only during parturition and lactation, but also plays an important role in other functions, given that Ca is soluble in acid and precipitates in alkaline pH, as is the case of urinary pH in certain situations. In sows with urinary tract infections, we expect to find alkaline urine, due to the microbiota located in the urinary tract which synthesize the urease enzyme and transform urea into ammonia, resulting in alkalization [27]. However, urinary pH at 14 days of lactation was more acidic when sows were fed with 100% organic Ca sources. This may indicate that Ca carbon-amino-phospho-chelate can help prevent the proliferation of pathogenic bacteria in the urinary tract or even acidify if an infected environment already exists, thus potentially preventing reproductive problems. However urinary pH levels from 5.5 to 7.5 are considered normal for sows, and levels from 6.5 to 8 represent urinary tract infections, according to Sobestiansky [3]. Thus, the levels found in our study would be within the normal urinary pH range for sows. Furthermore, the previous author stated that sows with and without urinary tract infections can show average pH levels of 6.4 and 6.2, respectively.

Ca plays a key role in many physiological processes, and its concentration in the extracellular fluid is regulated very precisely. In plasma, Ca is distributed into three major fractions: a biologically active ionized fraction, which corresponds to 50% of total Ca; the fraction bounded to proteins, especially albumin, which represents approximately 40% of total Ca; and the fraction bounded to anionic substances in plasma and interstitial fluids, such as citrate and phosphate, which account for the remaining 9%. Its extracellular concentration results from the balance between intestinal absorption, renal excretion and bone release or uptake. These processes are regulated by Vitamin D, by parathyroid hormone (PTH) and by calcitonin, hormones that are directly related to Ca metabolism [2]. The Ca ion analyses at 14 and 21 days of lactation were performed to observe Ca level fluctuation during this period, with no difference found in relation to treatments and sampling dates. Cunha et al. analyzed the ionic Ca of lactating sows and found ionic Ca values of 0.82 mmol/L, levels lower than those found in our study (i.e., 1.36 and 1.38 mmol/L at 14 and 21 days of lactation, respectively) [18]. Another issue is the concentration of Ca^{2+} inside the kidney cells, Andreoli and Mcateer reported an increase in Ca^{2+} concentrations within epithelial cells of pig kidneys (LLC-PK_1) related to injury due to hydrogen peroxide exposure [28]. However, Golconda et al. related a decrease in damage to LLC-PK_1 DNA due to the use of chelating agents (BAPTA) in intracellular Ca [29]. This helps us infer that nutritional intake of Calcium in chelated form could help preserve the integrity of the renal mucosa.

Secretion of the PTH hormone is related to Ca metabolism. It is responsible for stimulating bone demineralization to increase circulating Ca levels, but despite not having seen a difference between treatments with respect to circulating PTH levels, we observed in our data higher PTH levels at 14 days of lactation when compared to 21 days [2]. One may hypothesize that this numerical difference between 14 and 21 days of lactation could be related to the impact of the high ambient temperature, and consequently to the limitation on voluntary feed intake and milk production yield. Together these factors might have influenced PTH levels, since lactation demands a large amount of Ca from the body reserves, a higher milk production would necessarily have required more Ca and, as a result, a higher hormonal PTH response.

In conclusion, this study showed that in diets for lactating sows an inorganic Ca source can be fully replaced by a more available Ca source, without negatively impacting on the productive and reproductive performance of these sows or the performance of their litters. However, further studies are needed to assess the benefits of using more available Ca sources and its interactions with the bioavailability and longevity of sows.

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