ORIGINAL RESEARCH

Use of betaine in gilts and sows during lactation: effects on milk quality, reproductive parameters, and piglet performance

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Summary

Objectives: To study the effects of betaine inclusion in the feed on gilts and multiparous sows and their litters.

Material and methods: Forty-eight sows and gilts were randomly assigned to Control and Betaine groups, with the Betaine group receiving betaine-supplemented feed from 5 days before their due dates until the end of the lactation period of approximately 18 days. Production parameters were assessed over two consecutive parities, and colostrum and milk were quantitatively and qualitatively analyzed.

Results: In the first studied parity, only average daily feed intake differed between Betaine and Control group females $(5.43\pm0.10 \text{ kg})$ and $5.91\pm0.10 \text{ kg}$, respectively; P<.01). Litter weight at weaning was greater for the Betaine group than the Control group (P<.05). Weaning-to-estrus interval was shorter for the Betaine group $(4.7\pm0.4 \text{ days})$ versus $5.8\pm0.4 \text{ days}$; P<.05). In the second studied parity, means for the Betaine group were significantly greater for piglets born alive (13.9 versus 13.2, P<.05) and pigs weaned per sow (10.9 versus 10.5, P<.01). The content of betaine in milk was significantly greater

in the Betaine group (0.219 mg per g versus 0.125 mg per g; P < .05).

Implications: Treatment with betaine from 5 days before farrowing to the end of the lactation period can reduce the weaning-to-estrus interval, improve reproductive performance of gilts and sows, and increase body weight gain of the piglets.

Keywords: swine, betaine, wean-to-estrus interval, litter size, milk quality

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Resumen - Uso de la betaina en primíparas y multíparas durante la lactancia: efectos en la calidad de la leche, parámetros reproductivos, y desempeño del lechón

Objetivos: Estudiar los efectos de la inclusión de betaina en el alimento de hembras primíparas y multíparas y sus camadas.

Materiales y métodos: Se asignaron al azar, cuarenta y ocho hembras y primíparas a los grupos Control y Betaina; recibiendo el grupo Betaina, alimento suplementado con betaina desde 5 días antes de la fecha de parto hasta el término del periodo de lactancia de aproximadamente 18 días. Se evaluaron los parámetros de producción durante dos partos

consecutivos, y el calostro y la leche se analizaron cuantitativa y cualitativamente.

Resultados: En el primer parto estudiado, sólo el consumo de alimento diario promedio difirió entre las hembras del grupo Betaina y el Control $(5.43\pm0.10~{\rm kg}~{\rm y}~5.91\pm0.10~{\rm kg}$, respectivamente; P<.01). El peso de la camada al destete fue mayor en el grupo Betaina que en el grupo Control (P<.05). El intervalo de destete al estro fue más corto en el grupo Betaina $(4.7\pm0.4~{\rm d}{\rm a}$ s contra $5.8\pm0.4~{\rm d}{\rm a}$ s; P<.05). En el segundo parto estudiado, los promedios del grupo Betaina fueron significativamente mayores en lo que se refiere a lechones nacidos vivos $(13.9~{\rm contra}~13.2, P<.01)$ y los cerdos destetados por hembra

(10.9 contra 10.5, P < .01). El contenido de Betaina en la leche fue significativamente mayor en el grupo Betaina (0.219 mg per g contra 0.125 mg per g; P < .05).

Implicaciones: El tratamiento con betaina desde 5 días antes del parto hasta el término del periodo de lactancia puede reducir el intervalo de destete a estro, mejorar el desempeño reproductivo de primíparas y multíparas, y aumentar la ganancia de peso corporal en los lechones.

Résumé - Utilisation de la bétaïne chez les cochettes et les truies durant la lactation: effets sur la qualité du lait, les paramètres reproducteurs, et la performance des porcelets

Objectifs: Étudier les effets de l'inclusion de la bétaïne dans l'alimentation des cochettes et des truies multipares et de leurs portées.

Matériels et méthodes: Quarante-huit truies et cochettes ont été réparties de manière aléatoire aux groupes Témoin et Bétaïne, ce dernier recevant un aliment supplémenté en bétaïne à partir du cinquième jour précédant la date prévue de mise-bas jusqu'à la fin de la période de lactation d'une durée approximative de 18 jours. Les paramètres de production ont été évalués durant deux parités consécutives, et le colostrum et le lait ont été analysés quantitativement et qualitativement.

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Résultats: Lors de l'examen des données de la première parité, seule la quantité moyenne d'aliment ingéré différait entre les femelles des groupes Bétaïne et Témoin (respectivement, 5.43 ± 0.10 kg et 5.91 ± 0.10 kg; P < .01). Le poids de la portée au sevrage était supérieur pour le groupe Bétaïne comparativement au groupe Témoin (P < .05). L'intervalle sevrage-œstrus était plus court pour le groupe Bétaïne (4.7 ± 0.4) jours vs 5.8 ± 0.4 jours; P < .05). Lors de l'examen des données de la seconde parité, des moyennes significativement supérieures pour le groupe Bétaïne étaient notées pour le nombre de porcelets nés vivants (13.9 vs 13.2, P < .01) et de porcs sevrés par truie (10.9 vs 10.5, P < .01). Le contenu de bétaïne dans le lait était significativement supérieur dans le groupe Bétaïne (0.219 mg/g vs 0.125 mg/g; P < .05).

Implications: Un traitement avec de la bétaïne à partir du 5e jour avant la parturition jusqu'à la fin de la période de lactation peut réduire l'intervalle sevrage-œstrus, améliorer les performances de reproduction des cochettes et truies, et augmenter le gain de poids corporel des porcelets.

Betaine, a natural extract from beet root, is used in animal nutrition to improve productive performance. 1,2 Much research in the literature regarding betaine applies to finishing pigs, 3,4 but there is little published information concerning the effects of betaine on reproductive traits in female breeders.

Several effects of betaine have been previously described. The extract acts as an osmotically active substance, retaining water in the cells, aiding the function of ionic pumps, and saving energy used in electrolyte balance.^{5,6} It also improves liver function, contributing to homeostasis, and acts as a methyl-donating compound.² When feed intake is reduced or limited (eg, during the summer season in hot climates), pregnant and lactating sows are unable to obtain enough energy through the feed, resulting in a severe body-weight loss in 3 to 4 weeks due to mobilization of fat stores or loss of muscle mass. Even when the current litter is not affected, the size of the subsequent litter may be smaller.⁷ The effects of betaine are more pronounced when feed intake is restricted, and this suggests that an advantage may be derived from its use to improve reproductive traits.8 Follicular development may be poor after weaning when the lactating sow has been in a catabolic state,⁹ and hormonal balance may be altered by nutrient consumption during lactation, resulting in a non-adequate luteinization of the corpus luteum.¹⁰ Thus, an increase in feed energy during lactation might raise the luteinizing hormone (LH) peak, favoring an increase in litter size in the following parity.¹¹

Betaine's activity as an osmolyte is particularly important in situations of cellular dehydration, as osmolytes help to minimize water loss against a prevailing osmotic gradient. 12 There is also some evidence that betaine reduces energy expenditure for ion pumping, and the spared energy may promote cell proliferation and consequently stimulate growth in young animals.^{2,6} In the gut, betaine's osmolytic activity might increase the thickness of the intestinal mucosa, providing a greater surface area for nutrient absorption. Betaine also regulates the transmethylation cycle, with a positive effect on protein metabolism in muscle, liver, and kidney.²

The aims of this study were to assess the effect of betaine on reproductive parameters in betaine-treated and control gilts and multiparous sows and to evaluate body-weight gain of the piglets during lactation.

Materials and methods

The University of Murcia Animal Care Committee reviewed and approved the experimental protocols. All facilities were committed to the Spanish and European Union Laws regarding welfare and protection of pigs.

Animals and treatment groups

The 24 Landrace × Large White gilts and 24 multiparous Landrace × Large White sows used in the study were housed in the Veterinary Teaching Farm of the University of Murcia (Spain), in a farrow-to-finish unit accommodating 250 breeders. The animals were specific pathogen free for porcine reproductive and respiratory syndrome virus, pseudorabies virus, *Mycoplasma hyopneumoniae*, and *Actinobacillus pleuropneumoniae*, as assessed by serological ELISAs and through a sentinel program.

Twelve gilts and 12 multiparous sows were randomly assigned to the treated group (Betaine), and the remaining 12 gilts and 12 sows were assigned to the control group (Control). Lactation length did not differ by group (18.6 ± 0.9 days and 18.0 ± 0.9 days for the Betaine and Control groups, respectively; P = .59).

Facilities and animal management

The farm had a farrowing barn divided into eight farrowing rooms with six farrowing crates $(2.10 \times 0.90 \times 0.65 \text{ m})$ per room. Each crate was provided with a feeder and a nipple drinker. The floor was partially solid, with a slatted section behind the sow over a manure pit.

The barn had an environmental control system with heaters, coolers, and exhaust fans. In each crate, a heating zone was provided for the piglets with either infrared lamps or a heating mat. The ambient temperature in the farrowing room was recorded on a daily basis using an environmental infrared thermometer. Temperature did not differ among rooms, ranging from 21.7°C to 26.8°C and averaging 22.3°C minimum and 25.1°C maximum.

Female breeders were housed in the farrowing rooms, with equal numbers of gilts and sows in each room. The animals remained in gestation crates until they were moved to a farrowing room 5 days before their predicted due dates. Farrowing rooms were managed all-in, all-out and were cleaned and disinfected between groups of females. Animals were assisted at parturition and care was provided to all piglets after birth, including an injection of 200 mg of iron dextran (24 hours post farrowing) and identification by tattooing. Litters were standardized by cross-fostering within study group within 24 hours after farrowing.

Artificial insemination was used in all females, with each female receiving two doses of semen within a 24-hour interval. Semen was obtained on-farm using a commercial extender. Females were inseminated with fresh semen using a standard artificial-insemination catheter. Semen from two different boars was pooled in each dose, with 3×10^9 spermatozoa per dose. The same two boars were used for all matings. Fertility of the boars was assessed on a yearly basis using the homologous in vitro penetration test. 13

Feed

Pelleted feed was offered twice daily, and water was available ad libitum via nipple drinkers. Feed intake was assessed for each animal by weighing the feed remaining 30 minutes after it was delivered to the feeder.

Two different diets were provided: a basal diet for the Control group, and for the Betaine group, the same diet containing 2 kg per tonne of Betafin S1 (96% pure anhydrous betaine; Danisco Animal Nutrition, Naantali, Finland), with betaine replacing

an equal amount of wheat and providing 1.92 g of betaine per kg of feed. The study diets were offered to gilts and sows beginning when they were moved to the farrowing room, 5 days before their predicted farrowing date, and continued throughout lactation. Composition and nutritional values for the basal and betaine diets are shown in Table 1.

A pelleted prestarter feed, not supplemented with betaine, was offered to piglets in small removable feeders beginning when they were 9 days old, and water was available ad libitum from nipple drinkers.

Measurements

Two consecutive parities of each female were studied. All sows that began the trial farrowed a second time. Body weights of gilts and sows were recorded at the beginning and end of lactation. Parameters recorded or calculated included body weight loss during the first lactation, P2 backfat thickness at the beginning and end of lactation, backfat loss, average daily feed intake of gilts and sows, total born piglets, piglets born alive, stillborn piglets, litter weight at birth, litter weight at weaning, weaned piglets per

gilt or sow, weaning-to-estrus interval, and lactation duration. The following parameters were recorded for the second studied parity: total born piglets, piglets born alive, stillborn piglets, and weaned piglets per sow.

Gilts and sows were weighed on a digital mobile scale with a range of 0 to 300 kg, accurate to 0.1 kg, and litters were weighed on a digital mobile scale with a range of 0 to 30 kg, accurate to 0.1 kg. Backfat thickness at P2 for gilts and sows was measured with an A-mode ultrasonic instrument (Renco Lean-Meater series 12; Renco, Minneapolis, Minnesota).

Table 1: Composition of basal diet provided to sows and gilts during a study assessing the effects of betaine on reproductive and litter performance*

Component	Basal diet
Ingredients (g/kg)	
Wheat	242.8
Soy (44% crude protein)	160
Barley	160
Corn	150
Barley ecoprotein†	120
Beetroot	110
Calcium carbonate	11.8
Fat	11.6
Liquid lysine	6.6
Monocalcium phosphate	5.6
Calcium	4.0
Phosphorus	3.2
Sodium chloride	4.0
Vitamin-mineral premix‡	10.4
Calculated levels of nutrients (g/kg)	
Crude protein	170
Crude fat	35
Crude fibre	60
Crude ash	55
Lysine	10
Additive (g/kg)	
Phytase	0.01

- * For the Betaine diet the basal diet was supplemented with betaine anhydrous (1.92 g/kg) by adding 2 kg/tonne of Betafin S1 (96% pure anhydrous betaine; Danisco Animal Nutrition, Naantali, Finland) to replace an equal quantity of wheat. Treated sows and gilts were fed the basal and test diets beginning day 5 before the expected farrowing date and throughout lactation. Metabolizable energy = 13.6 MJ/kg. Twelve gilts and 12 multiparous sows were randomly assigned to the betaine group, with the remaining 12 gilts and 12 sows as controls.
- † Distillery dried grain with solubles.
- Provided (mg/kg): vitamin A, 3.9; vitamin D3, 0.05; vitamin E, 45; vitamin B12, 20; vitamin B6, 2; vitamin K3, 1; vitamin B2, 1; vitamin B1, 5; nicotinic acid, 20; CuSO₄, 10; ZnO, 100; FeCo₄, 80; Mn, 50; Se, 0.2; I, 1; Co, 0.5; calcium pantothenate, 10; folic acid, 3; biotin, 0.25; CoSO₄, 0.50; choline chloride, 250.

Colostrum and milk sample collection

The quality of colostrum and milk was assessed for each female in the study. Samples were collected from all sows and gilts on the day of farrowing (Day 0) and on Day 13. Sows and gilts were injected intramuscularly with 20 IU of oxytocin and were hand milked. A total of 50 mL of colostrum and 50 mL of milk were collected in sterile polypropylene tubes and immediately frozen and stored at -20°C until analysis.

Colostrum and milk analysis

Colostrum and milk samples were analyzed at the Laboratory of the Food Technology Department of the Veterinary School in the University of Murcia. Parameters analyzed included fat, protein, lactose, nonfat solids, total solids, and freezing point using a Milkoscan FT6000 (Foss, Hillerød, Denmark) and somatic cell count using a Fossomatic FC (Foss).

Methyl-donating compounds content was analyzed in the laboratory of Danisco Kantvik (Kantvik, Finland). The compounds analyzed were methionine, carnitine, betaine, and creatine plus creatinine using a YSI 2700 SELECT Biochemistry Analyzer (YSI Incorporated, Yellow Springs, Ohio).

Fatty acid composition of milk was analyzed in the Chemical Analysis Service of the University Autonoma of Barcelona (Spain) using a gas chromatography system (6890 Agilent with FID detector; Agilent Technologies, Santa Clara, California) and using as standard a Supelco 37 FAME mix (Sigma-Aldrich, Bellefonte, Pennsylvania). The fatty acids analyzed were butyric, caproic, caprylic, capric, undecanoic, lauric, tridecanoic, myristic, myristoleic, pentadecanoic, cis-10-pentadecenoic, palmitic, palmitoleic, heptadecanoic, cis-10-heptadecenoic, stearic, elaidic, oleic, cis-vaccenic, linole-

laidic, linoleic, arachidic, gamma-linolenic, cis-11-eicosenoic, linolenic, heneicosanoic, cis-11, 14-eicosadienoic, behenic, cis-8, 11, 14-eicosatrienoic, erucic, cis-11, 14, 17-eicosatrienoic, arachidonic, tricosanoic, cis-5, 8, 11, 14, 17-eicosapentaenoic, cis-13, 16-docosadienoic, lignoceric, nervonic, and cis-4, 7, 10, 13, 16, 19-docosahexaenoic. Results were expressed as percentages of the total fatty acid content.

Calculations and statistical analysis

Production data were subjected to a multi-factorial analysis of variance (MANOVA) in SPSS version 14 (SPSS Inc, Chicago, Illinois). A power study showed that a sample size of 18 females would have 90% power to detect a difference of 0.5 piglets born alive at $\alpha=.05$. For breeders, the following model was used:

$$\begin{split} Y_{ijklm} &= T_i + P_j + BWL_k + P2_l + (T_i \times P_j)_{ij} + \\ (T_i \times BWL_k)_{ik} + (T_i \times P2_l)_{il} + (P_j \times BWL_k)_{jk} \\ &+ (P_j \times P2_l)_{jl} + E_{ijklm} \end{split}$$

Where Y_{iiklm} = general mean

i = treatments (Betaine and Control)

j = parity (gilts and sows)

k = body-weight loss during suckling

1 = P2 backfat thickness (ultrasound)

 $ij = treatment \times parity interaction$

 $ik = treatment \times body$ -weight loss during lactation interaction

 $il = treatment \times P2 \ backfat \ thickness \\ interaction$

 $jk = parity \times body\text{-weight loss during lactation interaction}$

 $jl = parity \times P2$ backfat thickness interaction

 $E_{ijklm} = error$

For piglets, the following model was used:

$$Y_{ijkl} = T_i + P_j + LW_k + (T_i \times P_j)_{ij} + (T_i \times LW_k)_{ik} + (Op_j + LW_k)_{jk} + E_{ijkl}$$

where Y_{iik} = general mean

i = treatments (Betaine and Control)

j = parity (gilts and sows)

k = litter weight

 $ij = treatment \times parity interaction$

 $ik = treatment \times litter weight interaction$

 $jk = parity \times litter weight interaction$

 $E_{ijkl} = error$

Data for milk and colostrum quality were analyzed using an ANOVA in SPSS version 14. For all analyses, P < .05 was considered significant.

Results

Performance of females and litters

At the beginning of lactation in the first studied parity, weight averaged 219 \pm 9.1 kg and 223 ± 11.4 kg in gilts in the Control and Betaine groups, respectively, and P2 backfat thickness averaged 18 ± 1.6 mm and 21.1 ± 1.2 mm, respectively. In multiparous sows, weight averaged 246 ± 5.4 kg and 245 ± 5.2 kg for the Control and Betaine groups, respectively, and P2 backfat thickness averaged 20.5 \pm 0.9 mm and 19.4 \pm 0.4 mm, respectively. These parameters did not differ between treatment groups (P > .05). Body-weight loss and backfat loss during lactation did not differ (P > .05) either by treatment group or by parity group (gilt or sow) (Table 2). Average daily feed intake was lower in the Betaine group than in the Control group (Table 2), but there was no

difference between gilts and sows in either group (P > .05). The maximum difference in average daily intake was observed in gilts (5.20 ± 0.17 and 6.31 ± 0.77 for Betaine and Control groups, respectively; P < .001).

Litter weight at weaning in the first studied parity and liveborn piglets and number of weaned piglets per sow in the second studied parity were higher in the Betaine group (Table 2). Weaning-to-estrus interval was shorter in the Betaine group (Table 2). Average daily body-weight gain of piglets during lactation was 200.0 g per day and 183.5 g per day for Betaine and Control groups, respectively. There was a significant negative correlation between body-weight loss during the first studied lactation and total piglets born in the second studied parity ($R^2 = 0.534$; P < .01)

Colostrum and milk composition. Results of colostrum and milk analyses

Table 2: Results for production and reproduction parameters over two successive parities in gilts and sows fed either a basal diet (Control) or the same diet supplemented with betaine (Betaine)*

Parameter	Control (n = 24)	Betaine (n = 24)	SE	P †
Body weight loss, first studied lactation (kg)	30.21	28.83	1.91	> .05
Average daily feed intake (kg)	5.91	5.43	0.10	< .05
P2 backfat thickness, start of lactation (mm)	19.44	19.83	0.80	> .05
P2 backfat thickness, end of lactation (mm)	16.17	17.00	0.80	> .05
Backfat loss (mm)	3.25	2.83	0.60	> .05
Piglets born alive, first studied parity	12.1	11.5	0.63	> .05
Stillborn piglets, first studied parity	0.7	1.6	0.51	> .05
Total born piglets, first studied parity	12.8	13.1	0.72	> .05
Litter weight at birth (kg)	18.57	18.23	0.98	> .05
Litter weight at weaning (kg)	51.26	57.35	2.12	< .05
Weaned piglets per sow, first studied parity	10.4	10.4	0.22	> .05
Lactation duration (days)	18.0	18.6	0.9	> .05
Weaning-to-estrus interval (days)	5.7	4.7	0.4	< .05
Piglets born alive, second studied parity	13.2	14.0	0.45	< .05
Stillborn piglets, second studied parity	1.4	1.1	0.16	> .05
Total born piglets, second studied parity	14.6	15.1	0.25	> .05
Weaned piglets per sow, second studied parity	10.5	11.0	0.11	< .01

^{*} Diets described in Table 1. Study diets were fed beginning 5 days before the expected farrowing date and throughout the lactation of approximately 18 days.

[†] P for treatment effect. Production data were subjected to a multifactorial analysis of variance. Data were collected over the course of two successive parities. Average daily feed intake was calculated for the entire lactation period (approximately 18 days).

expressed as least squares means are shown in Table 3. Colostrum and milk composition did not differ either by parity or treatment group.

Methyl-donating compounds analysis

The results for methyl-donating compounds are shown in Table 4. The only parameter that differed significantly between the Betaine and Control groups was the content of betaine in milk, which was higher in the Betaine group. Although the content of betaine in colostrum was numerically higher in the Betaine group, there was no significant difference.

Fatty acid composition

The results for analysis of fatty acid composi-

tion are shown in Table 5. There were no significant differences between the Betaine and Control groups for any of the fatty acids. Fatty acids of the highest percentages were oleic, palmitic, linoleic, palmitoleic, stearic, myristic, and vaccenic (Table 5). Total fatty acids were 961.80 g per kg and 961.22 g per kg for the Betaine and Control groups, respectively.

Discussion

There were no significant differences in reproductive performance in the first studied parity, as was expected because the treatment with betaine started 5 days before farrowing. However, litter weight at 18 days of age was greater in the litters of treated sows and gilts. This might suggest that milk production

was higher in the Betaine group, as qualitative analysis of colostrum and milk did not differ between groups. However, it should also be taken into account that the content of betaine in the milk was higher in treated sows than in controls. Piglets in the treated group received 0.219 g of betaine per kg of milk, compared with control piglets that received 0.125 g betaine per kg milk. Betaine's activity as an organic osmolyte might lower the maintenance energy requirements of the piglets, resulting in energy savings. 4 Betaine supplementation in the diet enhances intestinal function and increases digestibility of most nutrients in piglets. 14,15 It also alters water retention in muscle cells, which might increase total body weight.¹⁶ In addition, betaine is a methyl donor that promotes betaine-homocysteine-methyltransferase

Table 3: Mean composition (least squares means) of colostrum and milk samples for females fed either a control diet (Control) or the same diet supplemented with betaine (Betaine)*

	Colostrum				Milk				
Parameter	Control n = 24	Betaine n = 24	SE	P †	Control n = 24	Betaine n = 24	SE	P †	
Fat (g/kg)	74.42	95.96	13.66	.30	79.65	77.75	8.43	.88	
Protein (g/kg)	103.24	80.31	13	.26	50.322	54.41	8.12	.75	
Lactose (g/kg)	34.87	37.21	4.90	.75	57.61	55.61	3.87	.75	
Non-fat solids (g/kg)	146.22	124.26	10.46	.18	107.02	110.03	5.98	.75	
Total solids (g/kg)	203.10	206.18	6.74	.76	191.48	191.57	8.66	.99	
Freezing point (m°C)	- 606	- 610	5.8	.65	- 597	- 593	4.7	.58	
Somatic cell count (cells/mL)	128,804	166,429	622	.70	231,345	239,367	1.09	.96	

^{*} For the treated group, the basal diet was supplemented with 1.92 g/kg of betaine anhydrous administered beginning 5 days before the due date and throughout the lactation period of approximately 18 days. Colostrum and milk samples were collected by hand milking after administration of oxytocin (20 IU). Colostrum was collected on the day of birth (Day 0) and milk was collected Day 13. Samples were stored frozen (-20°C) until analysis.

Table 4: Results of analysis for composition for methyl-donor compounds in colostrum and milk in sows and gilts either fed a basal diet (Controls) or that diet supplemented with betaine (Betaine)*

	Colostrum			Milk				
Methyl-donor compound	Control n = 24	Betaine n = 24	SE	P †	Control n = 24	Betaine n = 24	SE	P †
Methionine (g/kg)‡	< 0.005	< 0.005	NA	NA	0.014	0.013	0.006	.77
Betaine (g/kg)	0.198	0.289	0.009	.45	0.125	0.219	0.028	.03
Carnitine (g/kg)	< 0.002	0.002	0.0002	.14	0.009	0.008	0.001	.61
Creatinine (g/kg) + creatine	0.196	0.183	0.021	.70	0.230	0.196	0.029	.42

^{*} Diets described in Table 1.

NA = not applicable

[†] P values for treatment effect in ANOVA.

[†] P values for treatment effect in ANOVA.

[‡] Free methionine.

Table 5: Result of analysis for composition of the most abundant fatty acids (percentage of total fatty acids in g/kg) in milk in sows and gilts either fed a basal diet (Control) or that diet supplemented with betaine (Betaine)*

	Treat	ment			
Fatty acids	Control n = 24	Betaine n = 24	SE	P †	
Oleic	31.5	30.6	1.68	> .05	
Palmitic	21.2	20.8	1.10	> .05	
Linoleic	9.3	9.6	0.56	> .05	
Palmitoleic	6.2	5.5	0.86	> .05	
Stearic	3.7	4.0	0.19	> .05	
Myristic	2.5	2.2	0.31	> .05	
Vaccenic	2.2	2.2	0.11	> .05	
Linolenic	0.7	0.7	0.03	> .05	
Arachidonic	0.4	0.4	0.04	> .05	
Cis-11-eicosenoic	0.4	0.4	0.03	> .05	
Cis-11-14-eicosadienoic	0.3	0.3	0.03	> .05	
Heptadecanoic	0.2	0.3	0.20	> .05	
Lauric	0.2	0.2	0.02	> .05	
Miristoleic	0.1	0.2	0.02	> .05	
y-linolenic	0.1	0.1	0.06	> .05	
Capric	0.1	0.1	0.01	> .05	
Cis-8-11-14-eicosatrienoic	0.1	0.1	0.02	> .05	
Arachidic	0.1	0.1	0.01	> .05	
Cis-11-14-17-eicosatrienoic	0.1	0.1	0.01	> .05	

^{*} Diets described in Table 1.

activity, enhancing the recycling rate of homocysteine and maintaining S-adenosyl methionine and homocysteine levels.² Other effects of betaine supplementation should be considered. For example, it cannot be ruled out that betaine might improve litter performance by increasing milk production, or that betaine or its metabolites in milk might be concentrated in the mammary gland when milk is collected by hand milking.

Interestingly, there was no difference in bodyweight loss during lactation, but it should be taken into account that average daily feed intake was lower in the Betaine group. This has previously been observed in finisher pigs, at least as a trend. ¹⁷⁻¹⁹ Greater weight loss during lactation has been associated with lower feed intake in other studies using various diets. ²⁰⁻²² The absence of a difference in body-weight loss could be the result of energy savings resulting in less catabolism of body reserves in the Betaine group, but further research is required in gilts and sows.

Backfat loss did not differ between treatment groups, and while Boyd et al²³ recommended that backfat loss be limited to approximately 2 mm, Willis et al²⁴ observed a backfat loss in early-weaned (14 days) and conventionally weaned (28 days) primiparous sows similar to that reported in the current study, and without any influence on the number of embryos surviving after mating. 23,24 In the current study, backfat loss during lactation apparently did not influence performance in the second parity studied. The lower average daily feed intake in the Betaine group's second parity suggests better use of the energy and resources mobilized during lactation, or possibly more efficient use of dietary nutritional resources.

Weaning-to-estrus interval was significantly shorter in the Betaine group: 4.7 days versus 5.7 days in Control animals, a difference of 19%. The greater availability of energy derived from betaine intake may improve hormonal balance as previously reported. 11,25 Circulating levels of LH influence measures

of reproductive performance such as weaning-to-estrus interval, and effects of energy intake on secretion of LH during mid- to late lactation have been previously reported in sows. ²² It has previously been demonstrated that improved crude fat digestibility follows dietary supplementation with analogues of betaine. ²⁶ Moreover, betaine is involved in the synthesis of chylo-microns, which are involved in the absorption of fat, enhancing energy intake. ²⁷

In the second studied parity, the numbers of piglets born alive and weaned per sow differed significantly by treatment. Several explanations may apply to the larger number of piglets born alive in the Betaine group. The metabolic status of the sow during lactation influences follicular maturation after weaning²⁸ and is a critical determinant of subsequent fertility, as follicles may become atretic or cystic due to a negative energy balance.²⁹ Greater availability of energy during lactation can improve follicular development, resulting in a larger number of

[†] P values for treatment effect in ANOVA.

oocytes present at mating. 9,30,31 The priming effect of ad libitum feeding during the entire lactation influences the quality of the growing follicles and oocytes and the responsiveness of the ovary to increased secretion of LH and follicle-stimulating hormone by the pituitary gland after weaning.³² As all stages of follicular development may be sensitive to such changes, the levels of hormones that influence development of follicles at any stage of lactation may have a lasting effect on the ultimate size of the recruited follicles and their quality at weaning.³³ In our study, although the sows and gilts in the Betaine group had a lower average daily feed intake than the Controls, their energy status might have been higher because of the addition of betaine to the diet.

Under the conditions of this study, supplementation of betaine beginning 5 days before the predicted farrowing date resulted in a greater litter weight at weaning, a shorter weaning-to-estrus interval, and better measures of reproductive performance during the subsequent parity, in terms of piglets born alive and weaned piglets per litter. No differences in colostrum or milk composition, methyl-donating compounds (except for betaine), or fatty acid profile were observed. When these effects are combined, it may be predicted that the dietary use of betaine would increase the economic profit gained from each sow per year (taking the cost of betaine into account). However, before a general recommendation can be made to use betaine as a feed ingredient in order to improve reproductive performance and production traits, further research is needed using a larger sample size.

Implication

Under the conditions of this study, feeding betaine beginning 5 days before parturition and continuing throughout lactation can reduce the weaning-to-estrus interval, increase the numbers of born-alive piglets and weaned piglets per litter in the subsequent parity, and increase the body-weight gain of the piglets at weaning.

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References

- 1. Virtanen E. Piecing together the betaine puzzle. *Feed Mix.* 1995;3:12–17.
- 2. Eklund M, Bauer EJ, Wamatu S, Mosenthin R. Potential nutritional and physiological functions of betaine in livestock. *Nut Res Rev.* 2005;18:31–48.

- 3. Lawrence BV, Schinkel AP, Aldeola O, Cera K. Impact of betaine on pig finishing performance and carcass composition. *J Anim Sci.* 2002;80:475–482.
- 4. Schrama JW, Heetkamp MJW, Simmins PH, Gerrits WJJ. Dietary betaine supplementation affects energy metabolism of pigs. *J Anim Sci.* 2003;81:1202–1209.
- Kidd MT, Ferket PR, Garlich JF. Nutritional and osmoregulation functions of betaine. Worlds Poult Sci J. 1997;53:125–139.
- 6. Moeckel GW, Shadman R, Fogel JM, Sadrzadeh SMH. Organic osmolytes betaine, sorbitol and inositol are potent inhibitors of erythrocyte membrane ATPases. *Life Sci.* 2002;71:2413–2424.
- 7. Evangelista JNB. Produccion de leche de cerdas cruzadas y crecimiento y mortalidad de lechones durante la lactacion: Analisis de algunos factores de variacion [Milk production of crossbred sows and growth and mortality of piglets during lactation: Analysis of some factors of variation]. PhD dissertation. 1995. Universidad Politécnica de Madrid, Madrid, Spain.
- *8. Casarin A, Forat M, Zabaras-Krick BJ. Interrelationships between betaine (Betafin-BCR) and level of feed intake on the performance and carcass characteristics of growing-finishing pigs [abstract]. *J Anim Sci.* 1997;75(suppl 1):75.
- 9. Quesnel H, Pasquier A, Mounier AM, Louveau I, Prunier A. Influence of feed restriction in primiparous lactating sows on body condition and metabolic parameters. *Reprod Nutr Dev.* 1998;38:261–274.
- 10. Einarsson S, Rojkittikhun T. Effects of nutrition on pregnant and lactating sows. *J Reprod Fertility Suppl.* 1993;48:229–239.
- 11. Tokach MD, Pettigrew JE, Dial GD, Wheaton JE, Crooker BA, Johnston LJ. Characterization of luteinizing hormone secretion in the primiparous, lactating sow: relationship to blood metabolites and return-to-estrus interval. *J Anim Sci.* 1992;70:2195–2201.
- 12. Klasing KC, Adler KL, Remus JC, Calvert CC. Dietary betaine increases intraepithelial lymphocytes in the duodenum of coccidia-infected chicks and increases functional properties of phagocytes. *J Nutr.* 2002;132:2274–2282.
- 13. Martínez E, Vazquez JM, Matás C, Roca J, Coy P, Gadea J. Evaluation of boar spermatozoa penetrating capacity using pig oocytes at the germinal vesicle stage. *Theriogenology*. 1993;40:547–557.
- 14. Siljander-Rasi H, Peuranen S, Tiihonen K, Virtanen E, Kettunen H, Alaviuhkola T, Simmins PH. Effect of equi-molar dietary betaine and choline addition on performance, carcass quality and physiological parameters of pigs. *Anim Sci.* 2003;76:55–62.
- 15. Eklund M, Mosenthin R, Tafaj M, Wamatu J. Effects of betaine and condensed molasses solubles on nitrogen balance and nutrient digestibility in piglets fed diets deficient in methionine and low in compatible osmolytes. *Arch Anim Nutr.* 2006;60:289–300.
- 16. Esteve-García E, Macks S. The effect of DL-methionine and betaine on growth performance and carcass characteristics in broilers. *Anim Feed Sci Technol.* 2000;87:85–93.
- 17. Matthews JO, Southern LL, Pontif JE, Higbie AD, Bidner TD. Interactive effects of betaine, crude protein, and net energy in finishing pigs. *J Anim Sci.* 1998;76:2444–2455.
- 18. Cera KR, Schinkel AP. Carcass and performance responses to feeding betaine in pigs. *J Anim Sci.* 1995;73:82.

- *19. Haydon KD, Campbell RG, Prince TJ. Effect of dietary betaine additions and amino:calorie ratio on performance and carcass traits of finishing pigs [abstract]. *J Anim Sci.* 1995;73:83.
- 20. Rojkittikhun T, Einarsson S, Uvnäs-Moberg K, Edqvist LE. Body weight loss during lactation in relation to energy and protein metabolism in standard-fed primiparous sows. *J Vet Med A*. 1993;40:249–257.
- 21. Weldon WC, Lewis AJ, Louis GF, Kovar JL, Miller PS. Postpartum hypophagia in primiparous sows: II. Effects of feeding level during gestation and exogenous insulin on lactation feed intake, glucose tolerance, and epinephrine-stimulated release of nonesterified fatty acids and glucose. *J Anim Sci.* 1994;72:395–403.
- 22. Koketsu Y, Dial GD, Pettigrew JE, Marsh WE, King VL. Influence of imposed feed intake patterns during lactation on reproductive performance and on circulating levels of glucose, insulin, and luteinizing hormone in primiparous sows. *J Anim Sci.* 1996;74:1036–1046.
- 23. Boyd RD, Touchette KJ, Castro GC, Johnston ME, Lee KU, Han IK. Recent advances in amino acid and energy nutrition of prolific sows. *Asian-Aus J Anim Sci.* 2000;13:1638–1652.
- 24. Willis HJ, Zak LJ, Foxcroft GR. Duration of lactation, endocrine and metabolic state, and fertility of primiparous sows. *J Anim Sci.* 2003;81:2088–2102.
- 25. King RH, Martin GB. Relationship between protein intake during lactation LH levels and oestrus activity in first-litter sows. *Anim Reprod Sci.* 1989;19:283–292.
- 26.Overland M, Rørvik KA, Skrede A. Effect of trimethylamine oxide and betaine in swine diets on growth performance, carcass characteristics, nutrient digestibility, and sensory quality of pork. *J Anim Sci.* 1999;77:2143–2153.
- 27. Yao Z, McLeod RS. Synthesis and secretion of hepatic apolipoprotein B-containing lipoproteins. *Biochim Biophys Acta*. 1994;1212:152–166.
- 28. Quesnel H, Etienne M, Père MC. Influence of litter size on metabolic status and reproductive axis in primiparous sows. *J Anim Sci.* 2007;85:118–128.
- 29. Hoffman CK, Bilkei G. Effect of body condition of postweaning "flushed" sows and weaning-to-mating interval on sow reproductive performance. *Vet Rec.* 2003;152:261–263.
- *30. Foxcroft GR, Cosgrove JR, Aherne FX. Relationship between metabolism and reproduction. *Proc IPVS*. Bologna, Italy. 1996;6–9.
- 31. Van Wettere W, Herde P. Supplementing gestation diets with betaine increases litter size of summer mated sows. In: Robert Van Barneveld, ed. *Manipulating Pig Production*. 12th ed. Bentley Delivery Centre Australia: Australian Pig Science Association; 2009:140–159.
- 32. Britt JH, Arsmtrong JD, Cox NM, Esbenshade KL. Control of follicular development during and after lactation in sows. *J Reprod Suppl.* 1985;33:37–54.
- 33. Foxcroft GR. Mechanisms mediating nutritional effects on embryonic survival in pigs. *J Reprod Fertil Suppl.* 1997;52:47–61.
- * Non-refereed references.

