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# JOURNAL OF ANIMAL SCIENCE

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# Effects of ambient temperature on energy and nitrogen utilization in lipopolysaccharide-challenged growing pigs<sup>1</sup>

P. H. R. F. Campos,\*† E. Labussière,\*†<sup>2</sup> J. Hernández-García,\* S. Dubois,\*† D. Renaudeau,\*† and J. Noblet\*†

\*INRA, UMR 1348 PEGASE, F-35590 Saint-Gilles, France;  
and †Agrocampus Ouest, UMR1348 PEGASE, F-35000 Rennes, France

**ABSTRACT:** High ambient temperature impacts feed intake, growth, and nutrient utilization in pigs. However, little is known on its effects on immune function and, therefore, on how or if it could modulate the utilization of nutrients in pigs exposed to an inflammatory challenge. The aim of this study was to evaluate the effects of high ambient temperature on energy and nitrogen utilization in pigs submitted to repeated injections of *Escherichia coli* lipopolysaccharide (LPS). Twenty-eight catheterized and pair-housed female pigs (55 kg BW) were assigned to 1 of the 2 thermal conditions: thermoneutrality (TN, 24°C) or high ambient temperature (HT, 30°C). Within each condition, pigs had a 2-wk adaptation period in climatic-controlled rooms and then were transferred to open-circuit respiration chambers. Pigs remained in respiration chambers for a period of 18 d, which was divided into a 7-d period without LPS (baseline) and a subsequent 11-d period with LPS administration (LPS<sub>period</sub>). The interaction between ambient temperature and period was not significant for most of the traits studied. At baseline, pigs kept at HT had lower ADFI (1,500 vs. 2,003 g/d;  $P < 0.01$ ) and ADG (449 vs. 684 g/d;  $P = 0.01$ ) and similar nutrient digestibility compared with those kept at

TN. Pigs kept at HT also consumed less ME (1,651 vs. 2,170 kJ·kg BW<sup>-0.60</sup>·d<sup>-1</sup>;  $P = 0.01$ ) and produced less heat (1,146 vs. 1,365 kJ·kg BW<sup>-0.60</sup>·d<sup>-1</sup>;  $P < 0.01$ ) than those kept at TN. Furthermore, HT pigs retained less protein and fat than TN pigs (−61 and −57 g/d, respectively;  $P < 0.01$  and  $P = 0.01$ ). The LPS challenge reduced ( $P < 0.01$ ) nitrogen (−13.7 and −7.4 g/d) and ME intake (−594 and −335 kJ·kg BW<sup>-0.60</sup>·d<sup>-1</sup>) in TN and HT conditions, respectively; fecal digestibility of nutrients was not affected by LPS. During the LPS<sub>period</sub>, total heat production (HP) was decreased ( $P < 0.01$ ) in both TN and HT groups (−190 and −104 kJ·kg BW<sup>-0.60</sup>·d<sup>-1</sup>, respectively), in connection with the lower short-term thermic effect of feeding ( $P = 0.01$ ) and resting HP ( $P < 0.01$ ). In addition, the LPS induced a reduction in protein ( $P < 0.01$ ) and fat deposition ( $P = 0.01$ ) in pigs kept at TN (−79 and −73 g/d, respectively) and at HT (−41 and −44 g/d, respectively). In conclusion, our study confirms that high temperature reduces feed intake, growth performance, and HP. Moreover, our results evidence that irrespective of thermal condition, an inflammatory LPS challenge affects energy utilization through changes in ME intake and maintenance requirements.

**Key words:** energy expenditure, growing pig, heat stress, immunological stress, lipopolysaccharide

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## INTRODUCTION

Demand for animal products in tropical and subtropical areas is continuously increasing (Thornton, 2010). However, climatic factors limit the development of livestock production in these regions. Among them, high ambient temperature is a well-known factor affecting feed intake and growth in pigs (Renaudeau et al., 2011). Moreover, the high ambient temperature and high relative humidity occurring in tropical areas benefit pathogen proliferation and dissemination, resulting in greater environmental pathogenic pressure (Patz et

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<sup>2</sup>Corresponding author: etienne.labussiere@rennes.inra.fr

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al., 2000). Activation and maintenance of immune function have a direct negative effect on feed intake and growth (Johnson, 1997; Pastorelli et al., 2012) in addition to inducing an increase in energy expenditure as a result of increased immune compounds synthesis and metabolic cost of fever (Rauw, 2012).

Understanding how ambient temperature and inflammation interact with nutrient metabolism is of great interest, especially in the context of climate change (e.g., global warming, changes in seasonal and annual precipitation patterns, and occurrence of extreme weather events) and food animal production increase in hot-climate countries. Recently, a study performed in our laboratory has shown that the systemic effects of repeated administration of *Escherichia coli* lipopolysaccharide (LPS), at least in terms of hormonal and immune responses (e.g., cortisol and proinflammatory cytokines release), were attenuated in heat-acclimated pigs (Campos et al., 2014a). However, little is known on the associated effects of a high ambient temperature and an inflammatory challenge on nutrient metabolism. The objective of this study was to evaluate if ambient temperature affects energy and nitrogen utilization in growing pigs subjected to an inflammatory challenge through indirect calorimetry measurements in respiration chambers.

## MATERIALS AND METHODS

The experiment was conducted in accordance with the French legislation on animal experimentation and the experimental protocol was approved by the Regional Ethics Committee on Animal Experimentation of Rennes, France (authorization: R-2012-EL-03).

### Experimental Design and Animal Management

The study aimed to investigate the effects of ambient temperature on energy and nitrogen utilization in pigs subjected to an inflammatory challenge. It included 28 Piétrain × (Landrace × Large White) female growing pigs and was performed in 7 successive replicates. Each replicate was composed of 4 pigs from 2 different litters (2 per litter). Within each litter, 1 pig was assigned to 1 of the 2 thermal treatments: thermoneutrality (TN; 24°C) or high ambient temperature (HT; 30°C). Herein, the term high ambient temperature refers to ambient temperature above the upper limit of the thermoneutral zone, i.e., approximately 25°C for growing pigs (Renaudeau et al., 2008). At about 55 kg BW, pigs were surgically fitted with a jugular catheter, according to the procedure previously described by Campos et al. (2014b), and were transferred to temperature-controlled rooms, according to their allocation (TN or HT). Pigs remained in these rooms for a recovery and adaptation period of 14

**Table 1.** Composition of diet (as-fed basis)

Item	Amount
Ingredients, g/kg	
Corn	160.0
Wheat	262.0
Barley	255.5
Soybean meal	190.0
Palm oil	20.0
Molasses	30.0
Wheat bran	50.0
Bicalcium phosphate	5.0
Calcium carbonate	12.86
Salt	4.50
L-lysine HCl	3.33
DL-methionine	0.42
L-threonine	0.30
Vitamins, oligoelements, and phytase	6.10
Analyzed chemical composition, <sup>2</sup> %	
CP	18.0
Ash	5.6
Ether extract	3.5
Crude fiber	3.3
NDF	13.9
ADF	4.2
ADL	0.4
Starch	39.5
GE, MJ/kg	15.98
Nutritional energy values, <sup>2,3</sup> MJ/kg	
ME	12.81
NE	9.58

<sup>1</sup>Supplied per kilogram (as-fed basis) of diet: vitamin A, 5,000 IU; vitamin D<sub>3</sub>, 1,000 IU; vitamin E, 20 IU; menadione, 2 mg; thiamine, 2 mg; riboflavin, 4 mg; niacin, 15 mg; pantothenic acid, 10 mg; pyridoxine, 1 mg; biotin, 0.2 mg; folic acid, 1 mg; cyanocobalamin, 0.02 mg; choline chloride, 500 mg; Fe, 80 mg as ferrous carbonate; Cu, 10 mg as copper sulfate; Zn, 100 mg as zinc oxide; Mn, 37 mg as manganous oxide; I, 0.2 mg as calcium iodate; Se, 0.2 mg as sodium selenite; and Co, 0.1 mg as cobalt sulfate.

<sup>2</sup>For an average DM content of 87.0%.

<sup>3</sup>Values calculated according to Sauvant et al. (2004).

d. In each room, pigs from the 2 litters were pair housed in a metabolic crate (1.5 × 2.2 m), had free access to water, and received ad libitum a cereals and soybean meal-based diet (Table 1). The photoperiod was fixed to 12 h of artificial light (from 0730 to 1930 h). In the TN room, ambient temperature was held constant at 24°C. In the HT room, ambient temperature was held constant at 24°C during the first 7 d and thereafter held at 30°C. The temperature transition from 24°C to 30°C occurred gradually over 3 successive days at a constant rate of 2°C/d. The exposure of pigs to the HT temperature during 1 wk was sufficient to acclimate the pigs according to a previous study by Campos et al. (2014b) showing that the adaptation of pigs to an ambient temperature of 30°C (i.e., steady values of rectal and body temperature, respiratory rate, and thyroid hormones) required 3 to 4 d.

After the 2-wk adaptation period, the 2 pairs of pigs were transferred to 2 respiration chambers, 1 maintained at TN (24°C) and the other at HT (30°C) temperature, according to their allocation. Pigs remained in the respiration chambers during 18 consecutive days, which corresponded to the experimental period. The open-circuit respiration chambers (12 m<sup>3</sup> of physical volume each) were similar to those described by Vermorel et al. (1973). Irrespective of chamber and ambient temperature, relative humidity was maintained in the range of 65% to 70%, and animals were subjected to the same above-mentioned photoperiod program. During the experimental period, the actual air temperature of each chamber was continuously recorded, and it averaged 24.3°C ± 0.2°C and 30.3°C ± 0.2°C in the TN and HT respiration chambers, respectively. In the respiration chambers, pigs were also pair housed in metabolic crates similar to those used in the adaptation period that were mounted on force sensors that produced an electrical signal proportional to the physical activity of the pigs (Quiniou et al., 2001). The crates (1.5 × 2.2 m) had a fully metal slatted floor, back and side walls made of wooden planks, and a front wall made of a transparent material allowing visual observation of animals from outside through a window on the front door of each respiration chamber. Pigs had enough space to roam around freely and to have social interactions in the metabolic crate. Feces, urine, and water spillage were collected in a slurry pit located below the slatted floor. Urine and water spillage were drained, through a drainage channel along the slurry pit, into plastic tanks containing 250 mL of 10% sulfuric acid placed outside the chambers. The metabolic crates were equipped with a weight sensor placed under the trough that allowed continuous recording of daily feed intake of the animals. Pigs were fed ad libitum, and additional feed (Table 1) was offered twice a day at 0800 h (manually) and at 1550 h (automatically by a feed hopper mounted above the trough). The access to the trough was regulated by an electronic trap door equipped with a sensor recognizing each pig via an electronic transponder in an ear tag; from 0600 to 0900 h and from 1530 to 1600 h, the access to the trough was blocked to calibrate the gas analyzers (during the morning; see details below) and/or for feed distribution. Pigs had free access to water; the water tank for each chamber was placed outside the chamber, and its weight was recorded using a weight sensor.

Measurements in the respiration chambers were conducted in 2 successive periods: a 6-d period before (baseline; from d -6 to d -1) and a 10-d period during (LPS<sub>period</sub>; from d 1 to 10) the inflammatory challenge. The first day in the respiration chamber (d -7) was considered an adaptation day and was not included in the calculation of HP. At the end of the experiment (d 11), animals were maintained for an additional day (fasting day) in

the respiration chamber, and they received no feed. The inflammatory challenge consisted of repeated injections of *Escherichia coli* O55:B LPS (Sigma-Aldrich, Saint Quentin Fallavier, France) on d 1 (LPS1), d 3 (LPS2), d 5 (LPS3), d 7 (LPS4), and d 9 (LPS5) following a procedure adapted from the one described by Rakhshandeh and de Lange (2012). The initial LPS dose of 30 µg/kg of BW on d 1 was increased by 12% at each subsequent injection. The first to fourth LPS injections were performed intravenously via the catheter, and the fifth injection was given intramuscularly in the neck muscle to avoid a potential transient malaise. This protocol was validated in our experimental conditions (Campos et al., 2014a). Because of complementary measurements and procedures performed on d 1 (e.g., BW measurement), LPS1 was administrated at 1000 h, whereas the subsequent LPS administrations were performed at 0830 h. Three hours after LPS1 administration, all pigs were administrated 250 mg of paracetamol (Paracetamol 10 mg/mL, solution for infusion; Panpharma, Fougères, France) through the catheter to avoid severe hyperthermia and possible death. At the end of the experiment, pigs were euthanized (T61, MSD Santé Animale, Beaucaouze, France).

### Measurements and Samplings

Pigs were individually weighed on d -7, d 1, and d 11. Every day, the respiration chamber measurements were stopped at 0800 h for about 1 h. From 0800 to 0830 h, an experimenter entered each chamber to collect feed refusals, refill the trough and feed hopper, collect feces accumulated on the walls and on the floor, provide care for the animals, and perform experimental procedures (e.g., LPS administration, catheters maintenance). Concomitantly, gas analyzers were calibrated with ingoing air as a baseline and air from a gas tube with known gas concentrations as a standard; water tanks were refilled with fresh water. At about 0900 h, measurements were restarted. Samples of offered feed were taken every day and pooled per period (baseline or LPS<sub>period</sub>) for immediate DM determination and further chemical analyses. Feed refusals were collected at the end of the baseline period and every 2 d during the LPS<sub>period</sub> and were similarly pooled per chamber and per period for immediate DM determination. Daily collected feces (i.e., from walls and floor) and feces that accumulated under the floor were cumulated per period and, at the end of the period, were weighed and homogenized, and 3 samples were taken: 2 were used for DM determination, and 1 was freeze-dried, ground, and stored at 4°C for further chemical analyses. Daily collected urine from each chamber was weighed and homogenized, and a sample (1% of daily weight) was taken, pooled per period, and frozen (-20°C) until further laboratory

analyses. Nitrogen losses due to  $\text{NH}_3$  evaporation in the respiration chamber originating mainly from urine were measured per chamber and per period by collecting the condensed water from the air-conditioning system and, through a representative aliquot of the outgoing air from the chambers, according to the methods previously described by Noblet et al. (1987). The difference between the inflow and outflow volumes of  $\text{CO}_2$ ,  $\text{O}_2$ , and  $\text{CH}_4$ ; ventilation rate; feed intake behavior; signals of force sensors; and the physical characteristics of the gas in the respiration chambers (temperature, relative humidity, and barometric pressure) were measured 60 times per second, averaged over 10-s intervals, and recorded for further calculations of the heat production (HP) components. The details of these measurements have been described previously by Labussière et al. (2013).

### Laboratory Analyses

Samples of offered feed, pooled per replicate, were analyzed for DM, ash, starch, crude fiber, fat, and nitrogen (Dumas method) content according to AOAC (1990) methods. Gross energy content was measured according to AOAC (1990) methods and using an adiabatic bomb calorimeter (IKA, Staufen, Germany). Cell wall components (NDF, ADF, and ADL) were determined according to Van Soest et al. (1991). For each replicate, samples of feces per chamber and per period were analyzed for DM, ash, nitrogen, and GE using the same methods as for feed. Urine samples were analyzed for nitrogen and GE; the latter was determined after freeze-drying approximately 30 mL of urine in polyethylene bags of known GE concentration. The ammonia content of condensed water and extracted air was determined using an enzymatic method (Enzytec™ fluid Ammonia, Thermo Fisher Scientific Oy, Vantaa, Finland).

### Calculations

All data were measured for each pair of pigs housed in the same respiration chamber and were subsequently expressed per pig or per kilogram of metabolic BW ( $\text{kg BW}^{0.60}$ ; Noblet et al., 1999). Apparent digestibility coefficients of nutrients and energy were calculated according to standard procedures. Nitrogen retention (NR) was calculated as the difference between nitrogen intake and nitrogen lost in feces and urine and evaporated as  $\text{NH}_3$ . Protein deposition (in grams) corresponded to  $\text{NR} \times 6.25$ . The DE and ME intakes were calculated according to standard methods, including energy lost as  $\text{CH}_4$  in the ME intake calculation.

Data from d -7 and 1, which were considered to be adaptation or transition days, were not included in period energy balance calculations. Total HP was calculated from

respiratory gas exchanges (i.e., indirect calorimetry), urinary nitrogen (including nitrogen evaporated), and  $\text{CH}_4$  production according to the formula of Brouwer (1965). Energy retention (ER) was calculated as the difference between ME intake and HP. Energy retained as protein ( $\text{ER}_p$ ) was calculated from nitrogen balance, assuming an energy value of protein gain ( $\text{NR} \times 6.25$ ) of 23.6 kJ/g (McDonald et al., 2011). Energy retained as fat ( $\text{ER}_f$ ) was calculated as the difference between ER and  $\text{ER}_p$ . Fat deposition was calculated from  $\text{ER}_f$  assuming an energy content of 39.7 kJ/g of deposited fat (Brouwer, 1965). From  $\text{ER}_p$ ,  $\text{ER}_f$ , and ME intake, maintenance ME requirements ( $\text{ME}_m$ ) were calculated as  $\text{ME intake} - \text{ER}_p/0.60 - \text{ER}_f/0.80$  (Noblet et al., 1999). The respiratory quotient (RQ) corresponded to the ratio between  $\text{CO}_2$  production and  $\text{O}_2$  consumption. Simultaneous measurements of  $\text{CO}_2$  and  $\text{O}_2$  concentrations, signals of the force sensors, meal information (i.e., time and ingested quantities), and the physical characteristics of the gas in the chamber were used as inputs to calculate components of HP according to the modeling approach of van Milgen et al. (1997) and using R software (R Development Core Team, 2010; Soetaert et al., 2010), as previously described by Labussière et al. (2013). The HP due to physical activity (AHP), the short-term thermic effect of feeding ( $\text{TEF}_{st}$ ), and resting energy metabolism (RHP) were then calculated on the basis of their respective estimated  $\text{O}_2$  consumption and  $\text{CO}_2$  production using the formula of Brouwer (1965), excluding urinary N losses and  $\text{CH}_4$  production. On d 11, fasting heat production (FHP) was calculated from the asymptotic  $\text{O}_2$  consumption and asymptotic  $\text{CO}_2$  production at the end of the fasting day and excluding HP related to physical activity. Daily feed intake, total HP, and RQ (including those on d 1 and 11) were graphically presented. Because measurements in respiration chambers on d 1 started later than usual (i.e., after 1300 h), volumes of  $\text{O}_2$  consumption and  $\text{CO}_2$  production on that day were extrapolated to the 24-h period assuming proportionality and were then used to calculate total HP and RQ on d 1.

### Statistical Analyses

Performance, fecal digestibility, nitrogen, and energy balance variables per period were analyzed using a linear mixed model (MIXED procedure, SAS Inst. Inc., Cary, NC) that included the fixed effects of ambient temperature (TN or HT), period (baseline or  $\text{LPS}_{\text{period}}$ ), the interaction between ambient temperature and period, and replicate. In a second approach, linear contrasts were generated using the contrast statement of the MIXED procedure to compare, within each thermal condition, mean values measured at the baseline and those measured during the  $\text{LPS}_{\text{period}}$ . Adjusted means were compared using the Tukey test. The MIXED models

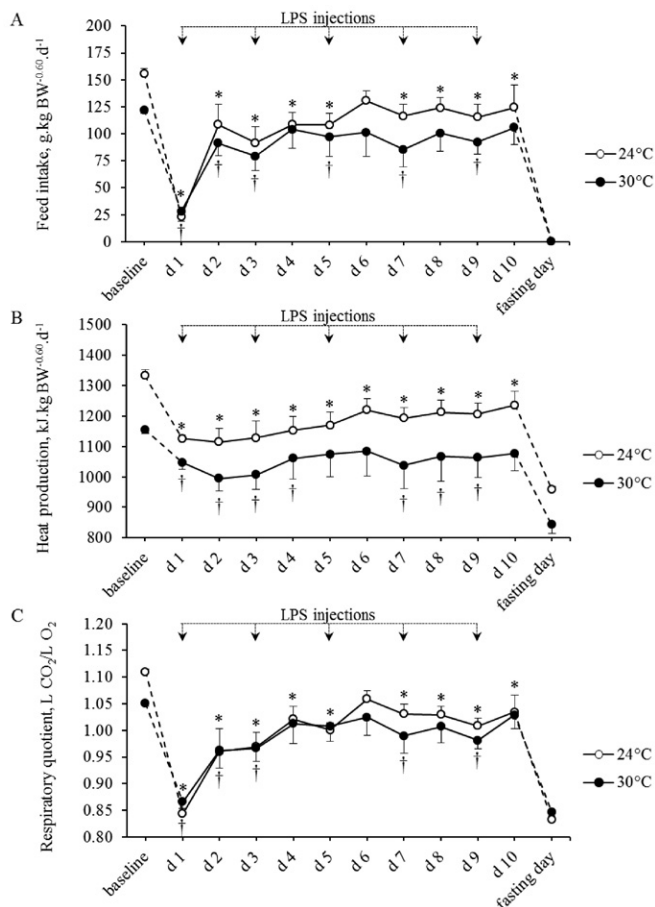
included the effect of period as a repeated effect, and a compound symmetry covariance structure was used to account for the experimental unit effect over time. In addition, within each thermal condition, daily mean values of feed intake, total HP, and RQ during the LPS<sub>period</sub> (i.e., from d 1 to 10) were compared to the mean value at baseline using the contrast statement of the MIXED procedure (SAS Inst. Inc.). For all analyses, the pair of pigs housed in 1 respiration chamber was the experimental unit; effects were considered to be significant if  $P < 0.05$ .

## RESULTS

Visually, most pigs had nausea and diarrhea and demonstrated signs of lethargy and hyperventilation after the first and, to a smaller extent, the second LPS administrations. On subsequent injections, pigs also demonstrated signs of lethargy and hyperventilation, but nausea and diarrhea were not observed. One pig from the HT group in the seventh replicate died after the first LPS administration; therefore, overall data from this replicate were excluded from the data set. Statistical analyses were then performed on data from 6 replicates comprising a total of 6 groups of 2 pigs per temperature. The interaction between ambient temperature and period was not significant for most of the traits studied, except for RHP (interaction  $P = 0.01$ ). Therefore, results will be presented as the average effects of ambient temperature irrespective of period, the average effects of the LPS challenge irrespective of thermal condition, and, when pertinent, the effects of the LPS challenge within each thermal condition. In addition, daily ADFI, HP, and RQ kinetics during the LPS challenge and within each thermal condition are presented in Fig. 1.

### Growth Performance and Nutrient Digestibility

Mean BW of pigs did not differ between thermal conditions and increased between the baseline and the LPS<sub>period</sub> (from 64.0 to 67.0 kg on average;  $P < 0.01$ ; Table 2). The ADFI and ADG were lower in pigs kept at HT than in those kept at TN, and both traits were also lower during than before the LPS challenge ( $P < 0.05$ ). Although not significant, the magnitude of the LPS-induced decrease in ADFI was numerically greater in pigs kept at TN than at HT (−504 vs. −279 g/d; interaction  $P = 0.30$ ). The LPS also induced a numerically greater decrease in ADG in pigs kept at TN than in those at HT (−457 vs. −260 g/d; interaction  $P = 0.43$ ). On the basis of daily measurements (Fig. 1), a severe depression in ADFI was observed on the day of the first LPS injection in pigs kept at TN and at HT (−85% and −77% of baseline, respectively;  $P < 0.01$ ). On the subsequent injections and irrespective of thermal condition, ADFI was also reduced ( $P < 0.05$ ), but the magnitude of the decrease was lower than that observed after the first injection.



**Figure 1.** Effect of lipopolysaccharide challenge at thermoneutrality (24°C) and at high ambient temperature (30°C) on (A) feed intake, (B) heat production, and (C) respiratory quotient in 65 kg BW growing pigs housed by pairs in respiration chambers. At baseline, each point is the mean  $\pm$  SEM of the 6-d period before the LPS challenge per ambient temperature (36 observations per ambient temperature). From d 1 to 10 and on fasting day, each point is the mean  $\pm$  SEM of 6 observations per ambient temperature. \*At 24°C, daily mean value is statistically different from baseline ( $P < 0.05$ ). †At 30°C, daily mean value is statistically different from baseline ( $P < 0.05$ ). Fasting day values were not included in the statistical analyses; values were compared using the contrast statement of the MIXED procedure (SAS Inst. Inc., Cary, NC).

Except for nitrogen digestibility, which tended to be greater in HT pigs than in TN pigs (88.0% vs. 86.1%;  $P = 0.07$ ), digestibility coefficients and the CH<sub>4</sub> energy losses were not affected by ambient temperature or by the LPS challenge. Whatever the temperature level was, the urinary energy losses as a percentage of DE supply were increased by LPS injection ( $P < 0.01$ ; not shown) with a subsequently lower ME:DE ratio during the LPS period (94.7 vs. 95.8 on average;  $P < 0.01$ ). The ambient temperature did not affect the ME:DE ratio, which averaged 95.2%.

### Nitrogen and Energy Utilization

Consistent with the changes in ADFI, nitrogen intake was lower in HT pigs than in TN pigs ( $P = 0.02$ ; Table 3) and was lower during the inflammatory challenge ( $P < 0.01$ ). Concomitantly, nitrogen retention was lower

**Table 2.** Effects of ambient temperature on performance, fecal digestibility, and metabolizability of DE in growing pigs before (baseline; 6 d) and during (LPS<sub>period</sub>; 10 d) a LPS challenge (least squares means of 6 observations of 2 pigs per ambient temperature and per period; data expressed per pig)<sup>1</sup>

Item	Ambient temperature				RSD <sup>2</sup>	P-value <sup>3</sup>				Contrast <sup>4</sup>	
	24°C		30°C			AT	P	AT × P	R	24 <sub>b</sub> ×LPS	30 <sub>b</sub> ×LPS
	Baseline	LPS <sub>period</sub>	Baseline	LPS <sub>period</sub>							
Mean BW, kg	64.5	67.9	63.6	66.1	1.4	0.46	<0.01	0.46	0.03	<0.01	0.01
ADFI, <sup>5</sup> g/d	2,003	1,499	1,500	1,221	250	0.02	<0.01	0.30	0.08	<0.01	0.08
ADG, g/d	684	227	449	189	293	0.03	0.01	0.43	0.01	0.02	0.16
Water consumption, <sup>6</sup> g/d	4,959	4,429	6,472	6,392	1,246	0.22	0.56	0.67	0.52	0.48	0.91
Water consumption, <sup>6</sup> g/g of feed	2.53	3.03	4.28	5.32	0.38	0.03	<0.01	0.11	0.59	0.04	<0.01
Digestibility coefficients, %											
Dry matter	85.9	86.0	86.8	86.9	1.2	0.23	0.82	0.96	0.91	0.85	0.90
Organic matter	88.5	88.4	89.3	89.3	1.0	0.21	0.85	0.99	0.94	0.89	0.90
Nitrogen	86.2	85.9	88.4	87.5	2.0	0.07	0.50	0.73	0.55	0.81	0.48
GE	86.6	86.5	87.7	87.6	1.2	0.20	0.90	0.96	0.91	0.91	0.96
CH <sub>4</sub> :DE, <sup>7</sup> %	0.88	0.94	0.97	1.02	0.13	0.28	0.34	0.93	0.18	0.46	0.53
ME:DE, <sup>8</sup> %	96.0	94.8	95.5	94.6	0.6	0.08	<0.01	0.55	0.01	<0.01	0.03

<sup>1</sup>Lipopolysaccharide (LPS) challenge consisted of 5 successive injections of *Escherichia coli* LPS performed at 2-d intervals, each during the LPS<sub>period</sub>.

<sup>2</sup>Residual SD.

<sup>3</sup>Data were analyzed using a linear MIXED model (SAS Inst. Inc., Cary, NC) including the fixed effects of ambient temperature (AT;  $n = 2$ ), period (P;  $n = 2$ ), the interaction between AT and P (AT × P), and replicate (R;  $n = 6$ ), and period was specified as a repeated effect.

<sup>4</sup>Linear contrasts were generated using the contrast statement of the MIXED procedure to compare baseline to LPS<sub>period</sub> at 24°C (24<sub>b</sub>×LPS) and baseline to LPS<sub>period</sub> at 30°C (30<sub>b</sub>×LPS).

<sup>5</sup>For an average DM content of 87.0%.

<sup>6</sup>Comprises water intake and spillage.

<sup>7</sup>Energy lost as CH<sub>4</sub> to DE ratio.

<sup>8</sup>ME to DE ratio; ME was calculated as the difference between DE and energy losses in urine and as CH<sub>4</sub>.

in pigs at HT than at TN (16.3 vs. 23.0 g/d;  $P = 0.02$ ) and decreased significantly in response to LPS in both TN and HT conditions (−12.6 and −6.5 g/d, respectively;  $P < 0.01$ ). Similar responses were found for ME intake, which decreased in HT conditions (−389 kJ·kg BW<sup>−0.60</sup>·d<sup>−1</sup> on average;  $P = 0.01$ ) and during the LPS challenge (−464 kJ·kg BW<sup>−0.60</sup>·d<sup>−1</sup> on average;  $P < 0.01$ ). Total HP also decreased in response to the increased temperature (−176 kJ·kg BW<sup>−0.60</sup>·d<sup>−1</sup> on average;  $P < 0.01$ ), mainly associated with a lower RHP and lower TEF<sub>st</sub> ( $P < 0.01$ ). In response to the LPS challenge, total HP was reduced by 14% and 9% in pigs at TN and HT conditions, respectively ( $P < 0.01$ ). Irrespective of thermal conditions, the reduction in total HP during the inflammation challenge was mainly related to a lower RHP ( $P < 0.01$ ) and TEF<sub>st</sub> ( $P = 0.01$ ). With regard to daily effects of LPS on total HP, the repeated injections of LPS induced a permanent decrease in total HP in pigs at TN. In pigs at HT, total HP also decreased, but a transient recovery was observed on d 5, 6, and 10. When expressed as a percentage of ME intake, AHP was greater in pigs at HT than in those at TN (13.7% vs. 9.7% of ME on average;  $P < 0.01$ ) and was greater during than before the LPS challenge (12.9% vs. 10.5% of ME on average;  $P = 0.02$ ). Additionally, TEF<sub>st</sub> was lower at HT than at TN (7.1% vs. 8.0% of ME;  $P = 0.04$ ), whereas it was not

affected by LPS. When compared to the value calculated at TN, the estimated ME<sub>m</sub> decreased at HT (−90 kJ·kg BW<sup>−0.60</sup>·d<sup>−1</sup> on average;  $P < 0.01$ ), but whatever the thermal conditions, it did not differ between the baseline and LPS periods (961 kJ·kg BW<sup>−0.60</sup>·d<sup>−1</sup> on average).

The RQ tended to be lower in pigs kept at HT than in those kept at TN (1.025 vs. 1.050;  $P = 0.08$ ), and it was significantly greater during the baseline period than during the LPS<sub>period</sub> (1.065 vs. 1.009;  $P < 0.01$ ). Concerning the daily pattern (Fig. 1), the LPS challenge induced a permanent decrease in the RQ in pigs kept at TN, except on d 6. In pigs kept at HT, RQ values were lower than at baseline only on d 1, 2, 3, 7, and 9. Finally, pigs kept at HT had lower protein (101 vs. 143 g/d on average;  $P = 0.02$ ) and fat deposition (60 vs. 102 g/d;  $P = 0.04$ ) than those at TN. Relative to baseline, LPS induced a reduction in protein deposition of 79 and 41 g/d in pigs at TN and at HT, respectively ( $P < 0.01$ ), and in fat deposition of 73 and 44 g/d in pigs at TN and at HT, respectively ( $P = 0.01$ ).



**Table 3.** Effects of ambient temperature on nitrogen utilization, energy balance, components of heat production, respiratory quotient, and nutrient deposition in growing pigs before (baseline; 6 d) and during (LPS<sub>period</sub>; 10 d) a LPS challenge (least squares means of 6 observations of 2 pigs per ambient temperature and per period; nitrogen and energy balance data expressed per pig or per kg BW<sup>0.60</sup>)<sup>1</sup>

Item	Ambient temperature				RSD <sup>2</sup>	P-value <sup>3</sup>				Contrast <sup>4</sup>	
	24°C		30°C			AT	P	AT × P	R	24 <sub>b×LPS</sub>	30 <sub>b×LPS</sub>
	Baseline	LPS <sub>period</sub>	Baseline	LPS <sub>period</sub>							
Nitrogen balance											
Intake, g/d	57.1	43.4	42.8	35.4	7.3	0.02	<0.01	0.31	0.08	<0.01	0.11
Excretion, g/d											
In feces	7.9	6.1	4.9	4.3	1.1	0.02	0.02	0.19	0.35	0.01	0.36
In urine and evaporated	19.9	20.7	18.4	18.0	2.4	0.23	0.84	0.58	0.45	0.60	0.80
Total	27.9	26.7	23.3	22.4	3.0	0.09	0.42	0.95	0.47	0.53	0.59
Retained, g/d	29.3	16.7	19.5	13.0	5.6	0.02	<0.01	0.21	0.04	<0.01	0.07
Retained, % of nitrogen absorbed	59.2	42.4	51.1	38.1	8.5	0.12	<0.01	0.59	0.07	<0.01	0.02
Energy balance, kJ·kg BW <sup>-0.60</sup> ·d <sup>-1</sup>											
ME intake	2,170	1,576	1,651	1,316	266	0.01	<0.01	0.26	0.05	<0.01	0.05
Heat production <sup>5</sup>											
RHP	1,012	880	809	766	35	<0.01	<0.01	0.01	0.11	<0.01	0.05
AHP	174	169	215	183	29	0.21	0.15	0.27	0.61	0.79	0.08
TEF, short-term component	180	126	122	93	33	<0.01	0.01	0.37	<0.01	0.02	0.16
Total	1,365	1,175	1,146	1,042	66	<0.01	<0.01	0.14	0.10	<0.01	0.02
Total adjusted <sup>6</sup>	1,237	1,202	1,154	1,137	28	<0.01	0.18	0.47	0.18	0.18	0.40
Retained energy											
As protein	354	195	238	154	62	0.01	<0.01	0.16	0.02	<0.01	0.04
As fat	450	206	267	120	151	0.04	<0.01	0.45	0.06	0.02	0.12
Total	804	400	505	274	206	0.02	<0.01	0.33	0.04	<0.01	0.08
ME utilization, % ME											
AHP	8.0	11.4	13.0	14.3	2.1	<0.01	0.02	0.26	0.12	0.02	0.32
TEF, short-term component	8.3	7.8	7.3	6.8	1.0	0.04	0.25	0.95	0.03	0.43	0.39
Estimated ME <sub>m</sub> <sup>5</sup> , kJ·kg BW <sup>-0.60</sup> ·d <sup>-1</sup>	1,017	994	921	910	38	<0.01	0.29	0.72	0.40	0.32	0.60
Respiratory quotient	1.081	1.019	1.050	0.999	0.034	0.08	<0.01	0.73	0.09	0.01	0.02
Nutrient deposition, g/d											
As protein	183	104	122	81	35	0.02	<0.01	0.21	0.04	<0.01	0.07
As fat	139	66	82	38	48	0.04	0.01	0.47	0.07	0.03	0.15

<sup>1</sup>Lipopolysaccharide (LPS) challenge consisted of 5 successive injections of *Escherichia coli* LPS performed at 2-d intervals, each during the LPS<sub>period</sub>. Data from d 1 were not considered in the energy balance calculations.

<sup>2</sup>Residual SD.

<sup>3</sup>Data were analyzed using a linear MIXED model (SAS Inst. Inc., Cary, NC) including the fixed effects of ambient temperature (AT;  $n = 2$ ), period (P;  $n = 2$ ), the interaction between AT and P (AT × P), and replicate (R;  $n = 6$ ), and period was specified as a repeated effect.

<sup>4</sup>Linear contrasts were generated using the contrast statement of the MIXED procedure to compare baseline to LPS<sub>period</sub> at 24°C (24<sub>b×LPS</sub>) and baseline to LPS<sub>period</sub> at 30°C (30<sub>b×LPS</sub>).

<sup>5</sup>RHP: resting heat production; AHP: activity heat production; TEF: thermic effect of feeding; ME<sub>m</sub>: maintenance energy requirements calculated as ME intake – (RE<sub>p</sub>/0.60 + RE<sub>f</sub>/0.80), where RE<sub>p</sub> and RE<sub>f</sub> are energy retained as protein and fat, respectively (kJ·kg BW<sup>-0.60</sup>·d<sup>-1</sup>; Noblet et al., 1999).

<sup>6</sup>Adjusted for a ME intake of 1,557 kJ·kg BW<sup>-0.60</sup>·d<sup>-1</sup> (mean value of the experiment).

## DISCUSSION

### Effect of High Ambient Temperature in Healthy Growing Pigs

Before the LPS challenge, pigs seemed to be in good health and adapted to experimental conditions. Therefore, when they existed, differences in performance, nitrogen, and energy utilization between TN and HT pigs were specifically associated with an effect of ambient temperature. In agreement with literature studies dem-

onstrating a negative relationship between the increase of ambient temperature above the upper limit of the thermoneutral zone and voluntary feed intake, pigs kept at 30°C had a lower feed intake than those kept at 24°C (–80 g·d<sup>-1</sup>·°C<sup>-1</sup> on average). Our result agrees with the 70 g·d<sup>-1</sup>·°C<sup>-1</sup> reduction in feed intake estimated from the equation proposed by Quiniou et al. (2000) using data obtained in 30 to 90 kg BW castrated male pigs housed in the same respiration chambers and in groups of 3 or 4 pigs. In addition, Huynh et al. (2005) reported that each degree increase between 25°C and 32°C, at a relative

humidity of 65%, resulted in a 99 g/d decrease in feed intake in 60 to 70 kg BW group-housed pigs (10 pigs per group) in respiration chambers. This slightly greater reduction compared to our study may be associated with their greater group size and no prior acclimation and also to their greater range of ambient temperature increase, with the decrease in feed intake being as important when temperature is high (Quiniou et al., 2000; Renaudeau et al., 2010). Apart from a tendency of greater fecal digestibility of nitrogen at HT than at TN conditions in the present trial because of some possible effects of feed intake change with ambient temperature on fecal digestibility, high ambient temperature has negligible effects on fecal digestibility of nutrients. Similarly, Renaudeau et al. (2008) reported no effect of high temperatures (i.e., 28°C, 32°C, or 36°C vs. 24°C) on nitrogen fecal digestibility in 50 kg BW pigs fed ad libitum.

In connection with the lower ME intake ( $-519 \text{ kJ}\cdot\text{kg BW}^{-0.60}\cdot\text{d}^{-1}$ ),  $\text{TEF}_{\text{st}}$ , RHP, and total HP were also reduced in pigs kept at HT ( $-58$ ,  $-203$  and  $-219 \text{ kJ}\cdot\text{kg BW}^{-0.60}\cdot\text{d}^{-1}$ ). The reduction of these traits in hot conditions was previously reported in 60 kg BW pigs individually housed in the same respiration chamber between 24°C and 32°C ( $-100$ ,  $-300$ , and  $-360 \text{ kJ}\cdot\text{kg BW}^{-0.60}\cdot\text{d}^{-1}$ , respectively; Renaudeau et al., 2013). The lower effects observed in our study might be similarly explained by the prior acclimation phase and the lower temperature increase. In the current study, the reduction in total HP is also associated with a reduction in the HP related to maintenance requirements ( $-96 \text{ kJ}\cdot\text{kg BW}^{-0.60}\cdot\text{d}^{-1}$ ). Expressed relative to the decrease of ME intake, the decrease in  $\text{ME}_m$  in our experiment equaled  $-0.19 \text{ kJ ME}_m/\text{kJ ME intake}$ . This decrease is quite similar to that reported by Labussière et al. (2011;  $-0.17 \text{ kJ ME}_m/\text{kJ ME intake}$ ), suggesting that the change in  $\text{ME}_m$  between both temperatures in the present trial most likely resulted from the lower feed intake rather than from a direct effect of the high ambient temperature. The decrease in feed intake in hot conditions has a beneficial effect in reducing HP associated with digestive and metabolic processes, as is evidenced by the 32% reduction in  $\text{TEF}_{\text{st}}$  in pigs kept at HT relative to those at TN.

In agreement with the direct effect of energy intake on protein and fat deposition (Quiniou et al., 1995) and the direct negative effect of high temperature on protein deposition (Le Bellego et al., 2002), protein and fat deposition were reduced in pigs kept at HT ( $-33\%$  and  $-41\%$ , respectively) with a subsequent decrease in weight gain ( $-235 \text{ g/d}$ ), even if the latter value measured over a short period has a rather low accuracy. Similarly, Campos et al. (2014b) reported a 290 g/d reduction in weight gain in pigs exposed to 30°C without prior acclimation (60 kg BW pigs from 2 lines selected for high or low residual feed intake). However, the weight gain

reduction observed in our study was greater than that estimated using the equation from the meta-analysis study of Renaudeau et al. (2011;  $-170 \text{ g/d}$ , considering a 24°C to 30°C temperature increase, 18% CP in the diet, and 65 kg BW). This discrepancy may be partially explained by the fact that our pigs were pair housed in respiration chambers in which the temperature of the floor and walls was similar to the ambient temperature, whereas most meta-analysis studies were performed with pigs housed individually and in temperature-controlled rooms equipped with cooler and conductive floors.

### ***Effect of Ambient Temperature on the Pig's Ability to Cope with a LPS Inflammatory Challenge***

The model of repeated injections of LPS was used to mimic the long-lasting metabolic effects of inflammatory responses in pigs exposed to pathogenic organisms (Rakhshandeh and de Lange, 2012). In the previous study of Campos et al. (2014a), this model was similarly implemented in pigs exposed to TN and HT conditions, and results indicated that pigs were subjected to a similar inflammatory stimulus irrespective of their thermal condition. In the present study, on the basis of a long-lasting reduction in feed intake, HP, and RQ, the LPS model also successfully induced an inflammatory state in pigs. However, irrespective of thermal conditions, the magnitude of the LPS effect was much greater after the first LPS injection than after the subsequent administrations, evidencing the development of an immune tolerance to the repeated stimulus despite the increased amount of LPS at each subsequent injection. Similar conclusions were reported in previous studies (de Ridder et al., 2012; Litvak et al., 2013). This fact may be associated with the synthesis of anti-inflammatory factors such as IL-10 and glucocorticoids that modulate the production of proinflammatory cytokines during a repeated stimulus to protect the organism against an excessive inflammatory response (Ziegler-Heitbrock, 1995; Rearte et al., 2010).

In the current study, the LPS challenge induced a reduction in feed intake of 500 and 280 g/d in pigs kept at TN and at HT conditions, respectively. These results are in close agreement with the 510 and 270 g/d feed intake reduction in TN and HT LPS-challenged pigs, respectively, reported in the study of Campos et al. (2014a). Feed intake depression caused by LPS has been similarly reported in young pigs (Daiwen et al., 2008), broiler chickens (Tan et al., 2014), and growing beef steers (Waggoner et al., 2009). This response is presumably mediated by the release of proinflammatory cytokines by immune cells in response to an inflammatory stimulus such as IL-1 that acts through both peripheral and central nervous system mechanisms to induce anorexia (Plata-Salamán, 1999; Konsman et al., 2002). Even

though our present results and those of Campos et al. (2014a) show a quite similar effect of the LPS model on feed intake, the responses of pigs to immune challenges are highly influenced by the type of challenge (i.e., duration, pathogen pressure, dose) and interindividual differences in immune responsiveness and status (Pastorelli et al., 2012). Pastorelli et al. demonstrated through a meta-analytic approach considering 6 different sanitary challenges that all of them induced a significant reduction in feed intake, but the magnitude of the reduction differed between challenges. For example, they reported a reduction in feed intake of about 8% for digestive bacterial infections, 4% for poor housing conditions, 10% for LPS challenges, 23% for mycotoxicoses, 3% for parasitic infections, and 16% for respiratory diseases. The greater LPS-induced reductions in feed intake observed in our study (−25% and −19% in TN and HT pigs, respectively) relative to that reported in the meta-analysis could be related to the fact that most of their database results were obtained for younger pigs (13 kg of initial BW on average) that received a fewer LPS injections.

Fecal digestibility of nutrients was not affected by LPS in pigs kept at either TN or HT. Similarly, Rakhshandeh et al. (2010) found no effect of chronic LPS administration on apparent ileal digestibility of amino acids and energy in growing pigs. Taken together, both studies suggest that chronic parenteral administration of LPS does not affect gastrointestinal digestive efficiency. These findings are in contrast to those of Rakhshandeh et al. (2012) that demonstrate a negative effect of LPS on apparent ileal digestibility and apparent fecal digestibility of CP in pigs, which was partially associated with increased endogenous nutrient losses. Furthermore, there is evidence that LPS affects intestinal health and absorptive function (Kanno et al., 1996; Albin et al., 2007) and, subsequently, nutrient digestion and absorption. This discrepancy between studies could be related to differences in the LPS model (e.g., dose, single or repeated LPS injections), experimental diet composition, and distribution (e.g., dietary fiber content, ad libitum or restricted feeding).

During the LPS challenge, total HP was reduced by 190 and 104 kJ·kg BW<sup>−0.60</sup>·d<sup>−1</sup> in pigs at TN and HT conditions, respectively. Similarly, Steiger et al. (1999) also reported a decrease in total HP in ad libitum-fed heifers receiving a single LPS administration compared to those receiving a saline solution (2 µg/kg BW; approximately −0.20 MJ/h between 6 and 10 h after the challenge). In our study and under both thermal conditions, the reduced total HP was mainly associated with a lower TEF<sub>st</sub> and a decreased RHP. The lower TEF<sub>st</sub> clearly reflects the associated effect of feed intake depression on HP. In contrast, the reduction in RHP has to be interpreted cautiously. According to the modeling approach for partitioning HP (van Milgen and Noblet, 2000), RHP is

the sum of HP relative to the basal metabolic rate and the long-term thermic effect of feeding. Thus, the effects of LPS on basal metabolic rate and the long-term effect of feeding are confounded. In addition, LPS is likely to affect these traits in a divergent way. On the one hand, it induces anorexia and therefore would induce a reduction in the thermic effect of feeding. On the other hand, it presumably induces an increase in metabolism to support the synthesis of immune system compounds (Johnson, 1997) and other actions of the activated immune system such as the increase in body temperature (Rauw, 2012; Singh and Hasday, 2013). Indeed, Campos et al. (2014a) reported an increase in circulating proinflammatory cytokines (i.e., IL-1β, IL-6, and INF-γ) and haptoglobin and increased rectal temperature (+1.1°C, on average) in TN and HT pigs challenged with LPS. Moreover, the febrile response by itself is a high-energy intensive process (Carroll and Forsberg, 2007; Rauw, 2012). In agreement with this presumed high metabolism during immune system activation, our results indicate that ME<sub>m</sub> did not decrease during the LPS challenge despite the significant reduction in ME intake caused by LPS. In fact, ME<sub>m</sub> is supposed to decrease when ME intake is reduced (−0.17 kJ/decreased kJ ME intake; Labussière et al., 2011); however, this decrease was not observed in our study. From the relationship reported by Labussière et al. (2011), the reduction in ME intake should have induced a decrease in ME<sub>m</sub> of 101 and 57 kJ ME<sub>m</sub>·kg BW<sup>−0.60</sup>·d<sup>−1</sup> in TN and HT pigs, respectively. However, because the decrease was not observed, the result might reflect or be equivalent to the additional energy requirement of the inflammatory response under our experimental conditions. Accordingly, increased energy expenditure has been reported in humans with severe sepsis resulting from peritonitis (Plank et al., 1998) and in mice experimentally challenged with a single-administration keyhole limpet hemocyanin (150 µg; Demas et al., 1997).

The increase in maintenance requirements in LPS-challenged pigs seems to be, at least in part, responsible for the reduction in growth in LPS-challenged pigs (van Heugten et al., 1994). Accordingly, Daiwen et al. (2008) observed lower weight gain and feed efficiency in pigs receiving 2 successive doses of LPS (200 µg/kg BW in a 48-h interval) compared with pair-fed control pigs administered a saline solution. According to our results and in connection with the decrease in ME intake and the relative increase in ME<sub>m</sub>, ME available for growth (ME<sub>g</sub> = ME intake − ME<sub>m</sub>) was decreased between the baseline and the LPS challenge period (from 14.0 to 7.3 MJ/d in TN pigs and from 8.82 to 5.02 MJ/d in HT pigs). Also, protein and fat deposition were reduced by 40% and 50%, respectively, in pigs at TN. In pigs kept at HT, the corresponding values were 30% and 50%. Through regression analyses, Quiniou et al. (1995)

reported that each MJ decrease in  $ME_g$  was associated with a reduction in protein and fat deposition ( $-12.8$  and  $-11.4$  g, respectively, in 65 kg BW boars fed ad libitum). Applying a similar approach and assuming an efficiency of ME utilization of 0.60 and 0.80, respectively, for protein and fat deposition (Noblet et al., 1999), our data indicate a corresponding slope of  $-10.4$  and  $-11.9$  g/decreased MJ  $ME_g$  (not influenced by the thermal condition;  $P = 0.30$  and  $P = 0.31$ , respectively; results not shown). It should be noted that these findings did not consider data from the first LPS injection and thus may not represent the acute response to endotoxemia. On the other hand, they reflect responses during a chronic inflammatory challenge and therefore may better represent the disturbances occurring in pigs exposed to pathogenic organisms and under practical commercial conditions.

Overall, our results demonstrate a numerically greater effect of the LPS challenge in pigs kept at TN than in those kept at HT. However, the effect of the interaction between ambient temperature and period was above the 5% significance threshold for most traits evaluated. This absence of significance might be attributed to the high interindividual variability. Indeed, through a complementary analysis (not shown), we observed that the CV of most traits considerably increased between the baseline and the LPS period. For example, the CV of ME intake per kilogram of metabolic body size increased from 14% to 22% in pigs at TN and from 11% to 37% in pigs kept at HT. The same pattern of response was observed for other traits, such as protein deposition, whose CV between the baseline and the LPS period increased from 23% to 48% in pigs kept at TN and from 22% to 70% in those kept at HT. Accordingly, when LPS-induced responses are compared with those at baseline within each thermal condition, i.e., through linear contrasts, a different pattern of response between pigs at TN and those at HT is evidenced. For example, by using this analysis, it is demonstrated that the LPS challenge induced a significant decrease in ADFI, protein, and fat deposition in pigs kept at TN but not in those kept at HT. Furthermore, from daily measurements, it is shown that LPS induced a permanent decrease in daily feed intake and RQ in pigs kept at TN, whereas in those kept at HT, these traits mostly decreased on the days of LPS administration. These findings suggest that the effects of LPS on metabolism were moderated in pigs previously acclimated to HT compared to those kept at TN and are in close agreement with the findings of Campos et al. (2014a) demonstrating that LPS caused greater feed intake and growth depression (along with increased proinflammatory cytokines, haptoglobin, and cortisol release) in pigs kept at TN than in those kept at HT. Unfortunately, little is known about the potential effects of the thermal environment on the responses of

pigs subjected to an inflammatory challenge, and the few studies in this area, including those of Klir et al. (1997) and Frank et al. (2003), essentially focused on the effects of a low ambient temperature in either neonatal or weaning pigs. However, it seems that prior exposure of pigs to a high ambient temperature might enhance their ability to cope with a subsequent challenge and that heat shock proteins are presumably involved in this process. In fact, the expression of heat shock proteins is increased in response to high ambient temperature (Pearce et al., 2013a; Rhoads et al., 2013), and these proteins have a protective function against collateral damage associated with an excessive immune response during inflammation (Ozveri et al., 1999; Karrow, 2006; Launey et al., 2011), which may therefore explain the relatively greater capacity of pigs kept at HT compared to those kept at TN to limit the disturbances caused by the LPS challenge observed in our study. Moreover, the presumed negative effects of heat stress on intestinal barrier integrity that would result in greater endotoxemia and inflammation in pigs kept at HT (Pearce et al., 2013b,c) might have been attenuated in our experimental conditions because pigs were acclimated to their thermal conditions before the beginning of the experimental period. Furthermore, the recent body of evidence suggests that exposing cells to different types of environmental stress leads to epigenetic modifications of the expression of specific genes and biochemical mechanisms that because of their common ground, presumably improve resistance to a wide range of stressors. Therefore, besides minimizing immediate cellular damage, such responses may improve the overall capacity of the organism to resist and to cope with environmental challenges (Horowitz, 2001; Elsasser et al., 2012). On the other hand, it may also be hypothesized that the greater productivity level of pigs kept at TN compared with those kept at HT (i.e., greater ADFI, ADG, HP) may contribute to the greater extent of LPS effects in pigs at TN than in those at HT.

In conclusion, our study shows that high ambient temperature reduces voluntary feed intake, growth performance, and HP but does not affect fecal digestibility of nutrients in growing pigs preacclimated for 1 wk to such conditions. Moreover, irrespective of thermal conditions, the model of repeated injections of LPS induced a reduction in ME available for growth as a consequence of a decreased ME intake and increased  $ME_m$ . Finally, this study provides evidence that LPS-induced effects on nutrient utilization are attenuated in pigs previously acclimated to high ambient temperature compared with those at thermoneutrality. This knowledge should be considered in the development and application of strategies to mitigate the negative effects of thermal and sanitary challenges in animal production.

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