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Circadian Temperature Rhythm of Laboratory Swine

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The circadian temperature rhythm (CTR) profile holds promise for monitoring the domestic pig's responses to stress and illness. In the present study we quantified the CTR profile of nine growing–finishing swine using a time-series, small-group design. Temperature was monitored using a probe implanted in the ear for 5 1/2 to 9 1/2 consecutive days while the unrestrained pigs were housed singly in pens. The dominant period of the temperature data was estimated with the autocorrelation function and then used in standard cosinor analysis to compute the amplitude (half of the distance between the highest and lowest value within the period), mesor (rhythm-adjusted mean), and acrophase (timing of the cosine maximum). To examine the effect of procedural stress on CTR, we compared data from the first 3 days with those from subsequent days. Eight of the nine (89%) pigs had CTR with a mean (± standard error) period of 23.6 (0.5) h, amplitude of 0.18 (0.02)°C, mesor of 38.7 (0.24)°C, and acrophase at 19:44 h. Mean mesor and acrophase were not different, but amplitude was lower (P = 0.03) during the first 3 days after instrumentation than during subsequent days. We conclude that: 1) laboratory-housed, unrestrained, growing–finishing swine have CTR; 2) our ear-based instrumentation protocol imposes acute stress as reflected in attenuated CTR amplitude during the first 3 days after instrumentation; and 3) CTR adaptation to stress appears to occur over time.

Despite extensive research on circadian temperature rhythm (CTR) in diverse species, literature on this biological rhythm in the domestic farm pig is sparse, and the available studies were published more than 30 years ago. Although diurnal differences in temperature were reported (7, 29, 30), porcine CTR was not well characterized. Advances in monitoring and analysis technologies now make it both possible and desirable to understand biological rhythms by the conventional parameters of period, amplitude, mesor, and phase. Here we report CTR parameters over a period of ≤ 9 1/2 days for unrestrained growing–finishing swine housed in a research laboratory to document CTR in the domestic farm pig. Description of the CTR profile is fundamental to its potential use as a future reference to assess porcine CTR under stress and during adaptation.

All living organisms have clock mechanisms that coordinate internal biological rhythms. Mammals have a central pacemaker (the biological clock), located in the hypothalamic suprachiasmatic nuclei, that is reset daily by such environmental synchronizers as the photoperiod, sleep–wake cycles, and daily routines (4, 51). Poor synchronization of the central pacemaker to the external environment is associated with impaired physical and mental performance (26, 52). Synchronization problems can arise from disruption in the central pacemaker from illness or injury or such environmental changes as thermal stress and nighttime wakefulness in a diurnally active organism.

Because study of the central pacemaker is difficult (23), overt rhythms regulated by the pacemaker have been used to understand biological time-keeping mechanisms. Core body temperature, cortisol, and melatonin are reliable biomarkers of circadian rhythm in humans (34, 51) and the domestic farm pig (19, 28, 35). CTR, however, has been less well characterized in domestic laboratory swine than have cortisol (5, 17, 18, 31, 33, 44, 45) and melatonin (2, 17, 18, 43, 48) rhythms. Monitoring of CTR has advantages over biochemical markers because it is less costly, less invasive, and more amenable to continuous measurement.

One reason for the paucity of research on porcine CTR may be logistical problems with longitudinal temperature monitoring. Biomarker sampling should be at frequent intervals for sufficient periods of time to capture 24-h variation and stability of the rhythm over time (23). Technical challenges of CTR measurement in unrestrained pigs can be met by the use of telemetry or portable data loggers. Telemetric systems include implantable and ingestible transmitters. Surgical implantation of a sensor is a stressor and requires several days to weeks of recovery before meaningful data can be obtained (15, 32). Issues with ingestible transmitters include rapid and variable gut transit time, missing data and artifact from feed intake, and associated cost (22). Hahn and colleagues (27) successfully have used portable data loggers with an ear-based thermal sensor in swine. Their "tympanic" approach eliminates the barriers to continuous temperature monitoring posed by telemetry systems of measurement.

The CTR profile holds promise as a means of monitoring porcine stress and adaptation. Data suggest that stress tolerance and treatment responses vary at different times of the circadian cycle (19, 44). It is well established that stress and disease affect the circadian pattern of physiologic functions, albeit the accumulated evidence is mostly from human studies. The purpose of the present study was to characterize CTR over time in unrestrained growing–finishing swine that were housed in a research laboratory. We used an automated ear-based method to monitor body temperature for multiple consecutive days. Our method was modified from that used in swine and cattle by Hahn and colleagues (27); notably we inserted the ear-based sensor under general anesthesia. We compared the CTR profile during the first

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3 days after the procedure with that from subsequent days to determine whether our protocol imposed acute stress from transport and anesthesia and, if so, subsequent adaptation of the CTR. In addition, we intermittently monitored ambient temperature and humidity to explore ambient temperature synchronization of CTR and to ensure that ambient conditions were within the range recommended for laboratory swine.

Materials and Methods

A time-series, small-group design was used to examine CTR in the unrestrained domestic farm pig (*Sus scrofa*). The institutional animal care and use committee reviewed and approved the study. Care, handling, and procedures conformed to the U. S. Public Health Service guidelines for humane care and use of laboratory animals (38, 39), and the research facility was accredited by the Association for Assessment and Accreditation of Laboratory Animal Care, International.

Animals. Nine domestic farm pigs (six male, three female) between the ages of 4 1/2 and 5 months, weighing 53 to 80 kg (117 to 176 lb), underwent ear-based temperature monitoring for 5 1/2 to 9 1/2 consecutive days while they were individually penned in the animal care center housing room. The first seven subjects were Yorkshire × Blue Butt pigs from an open herd (Russell Barham, Midway, Tex.), and the last two subjects were Yorkshire × Duroc pigs from a closed herd (HDH Swine Farm, Boerne, Tex.).

The pigs, which were pairs of same-sex littermates except for pig 1, were transported from the farm to the animal care center in pairs and acclimated for 10 to 14 days before the study. Farm pigs from the closed herd were vaccinated at weaning (5 weeks of age) for atrophic rhinitis associated with *Bordetella bronchiseptica*, erysipelas caused by *Erysipelothrix rhusiopathiae*, and *Pasteurella multocida*. Since 1980, the vendor has introduced all blood lines through artificial insemination, thereby maintaining a closed herd, and all breeding animals have tested normal for malignant hyperthermia. On admission to the research facility (17 weeks), all subjects were dewormed and given oxytetracycline. Daily routine consisted of one midday meal of Lab Porcine Diet Grower (LabDiet, Formula 5084, PMI Nutrition International, Inc., Brentwood, Mo.), exercise while the pens were cleaned (10 to 20 min daily), examination by the clinical veterinarian (G.C.), and daily visits from the first author (S.K.H.). Water was available ad libitum.

The housing room was dark (except for a night light) between 1900 and 0700 and lighted between 0700 and 1900 with an automatic room timer. Air changes were 12 to 15 per hour. The pens were cleaned daily with biocide foam disinfectant and then rinsed with lukewarm water.

Instruments. A noninvasive tympanic membrane temperature sensor was fashioned from a YSI 400 series skin surface thermistor probe (Steri-probe, Cincinnati Sub-Zero Products, Cincinnati, Ohio) for connection to a portable physiologic data logger (MiniLogger Series 2000, Mini Mitter, Bend, Oreg.) programmed to measure temperature between 32 and 45°C with resolution of 0.05°C. Bias and precision of the thermistor probe across the temperature range of 34 to 42°C, with and without the logger, were tested in our laboratory with a laboratory-certified digital thermometer and a precision-controlled water bath (24). Bias and precision were 0 ± 0 for the probe alone and 0°C ± 0.2°C for the logger and probe together. Accuracy of the logger was within ± 0.08°C and remained stable during the data collection period, as evidenced by pre- and postcalibration certification performed by the manufacturer.

Ambient temperature and relative humidity in the housing area were monitored every min for 1 to 5 days during each subject's body temperature monitoring period by using the Q-Trak indoor air quality monitor (Model 8550, TSI Incorporated, St. Paul, Minn.). The monitor measures temperature from 0 to 50°C by a thermistor sensor with 0.1°C resolution, accuracy of ± 0.6°C, and response time of 30 sec at an air velocity of 2 m/sec. It measures relative humidity from 5 to 95% by a thin-film capacitive sensor with 0.1% resolution, accuracy of ± 3%, and response time of 20 sec at an air velocity of 2 m/sec.

Experimental protocol. The pigs were fasted 12 h before insertion of the temperature probe. After sedation with an intramuscular injection of atropine sulfate (0.5 mg/kg), acepromazine (0.11 to 0.22 mg/kg), and ketamine HCl (20 mg/kg), the pig was transported to the procedure room. Isoflurane (0.5 to 2%) was given by face cone to effect a light to moderate level of anesthesia; the therapeutic end-point was absence of ear twitch to manual stimulation. Electrocardiography leads were applied to monitor heart rate and rhythm.

After removing the adhesive and thermal shield from the thermistor, we advanced the thermistor tip through a 14-French suction catheter (Regu-Vac, Kendall Healthcare Products Company, Mansfield, Mass.) so that the thermistor tip lay 0.5 to 1.0 mm proximal to the suction catheter tip (a semirigid tube is needed to advance the sensor tip into the pig's ear canal to avoid curling of the flexible probe in the external meatus). The distal end of the probe was secured to the distal end of the suction catheter with an antiseptic adhesive solution (Benzoin Tincture Compound, Century Pharmaceuticals, Indianapolis, Ind.) and thin strips of adhesive tape. We applied tension to the distal end of the probe prior to insertion to verify that there was no discernible backward movement of the thermistor tip into the suction catheter.

A vest with pockets and lacing to adjust fit (Alice King Chatman Medical Arts, Hawthorne, Calif.) was put on the pig. The laces were adjusted to allow a weight gain of 0.5 kg daily for 10 days. The outer ear was prepped with an aqueous solution of 10% povidone–iodine (Betadine, Purdue Pharma L.P., Stamford, Conn.). With the animal in the lateral recumbent position, the probe was blindly, slowly, and carefully advanced in an anterior caudal direction into the external meatus of the ear until resistance was met. The insertion distance was variable, but in no case was it greater than 3 cm.

To ensure proper placement of the probe, we observed temperature logging with the data logger connected to a laptop computer. The temperature rose steadily from 31.4°C (the bottom end of our data logger scale) to a stable reading greater than 36°C. At that point, and while still watching temperature being recorded every sec, we sutured the suction catheter to the auricular cartilage at five sites. We then packed the ear loosely with 4 × 4 in. (ca. 10 × 10 cm) gauze pads, and wrapped the ear with tape to secure the probe.

Upon stabilization of the temperature and after securing the probe and logger, the MiniLogger was programmed with the laptop computer to measure temperature at 1-min intervals. The portable data logger was disconnected from the laptop, wrapped in a plastic bag, packed in gauze, and placed in a vest pocket. We returned the pig to the pen for recovery, observing the pig every

Table 1. Descriptive data for porcine subjects

Subject number	Sex	Age (months)	Weight (kg)	Ambulatory monitoring time (days)	Temperature (°C)	White blood cells ($\times 10^3$)
1	Female	4.5	64	6.5	38.09 ± 0.43	NA
2	Male	5	69	6.7	38.43 ± 0.54	NA
3	Male	5	77	6.0	38.61 ± 0.58	NA
4	Male	4.5	62	8.5	40.16 ± 1.0	15.4
5	Male	4.5	53	7.3	38.13 ± 1.0	16.5
6	Female	4.5	77	6.0	37.63 ± 0.74	14.7
7	Female	4.5	78	5.5	39.12 ± 0.85	24.2
8	Male[a]	5	80	9.0	38.39 ± 0.48	17.0
9	Male[a]	5	75	9.5	38.06 ± 0.42	14.1

NA, not available.
Normal is 10.3 – 20.7 K/mm^3 (36).
Temperature data are given as mean ± standard deviation.
[a]Subjects from closed herd.

30 min to 1 h throughout recovery and every 6 h thereafter until the probe was removed.

After examining the temperature data of the first three subjects, we added a blood draw to the protocol. In the subsequent six pigs, we drew blood from an ear vein before probe insertion, while the pig was anesthetized, to measure white blood cell (WBC) count as a screening indicator of subclinical infection.

Data management and analysis. We used a longitudinal analytic technique—cosinor analysis—to identify a systematic pattern in a sequence of observations of temperature. Cosinor analysis uses least-squares approximation of the time series with a cosine function of known period. Modeling of experimental data in this way is commonly used for variables governed by circadian and other biological rhythms (23, 40). The cosinor method is based on a Fourier series with a unique harmonic, and it considers that the variable of interest shows sinusoidal time dependence. The model provides three parameters: mean estimated statistic over rhythm (mesor, the rhythm-adjusted mean), amplitude of the oscillation (half of the distance between the highest and lowest value within the period), and the acrophase (time at which the variable reaches its maximum on the fitted curve). Limitations of the cosinor method are the assumptions that the circadian rhythm fits to a cosine wave with a period of 24 h and that there is no serial dependence in the observational errors (1).

Data were downloaded and examined for outliers, artifact, and length of the time series. The data required minimal editing. For all nine porcine temperature files, 2937 of 397,866 (0.007%) data points were deleted with editing. After editing, a 6-min windowed mean was applied to the data, resulting in 10 data points per hour. The resulting time series was detrended using a moving median with a width of 24.1 h to accurately estimate circadian parameters. The detrending procedure was used because our analysis approach assumes time-independence of data and temperature data are strongly time-dependent.

Porcine CTR profile. The autocorrelation function (ACF) was used to estimate the dominant period of the temperature data (46) and that period was used in standard cosinor analysis to compute the amplitude and mesor. Unlike methods assuming that the data fit a cosine wave, such as spectral analysis, the nonlinear-regression method (49), and the maximum-likelihood method (16), ACF analysis finds the dominant period of a time series without the assumption of cosine wave fit. Therefore, the ACF can reliably estimate the dominant period of temperature data, which often deviates dramatically from the cosine form.

ACF was computed for each time series by using progressive lag times of 0.1, 0.2, …, 96.0 h or until the end of the time series. The dominant period was defined to be the lag time at which the ACF reached its maximum value. Because the dominant rhythm found is an estimate, random fluctuations prevent it from being exactly equal to 24 h, even if the true rhythm is 24.0 h. Consequently, we assume that estimates of the period between 22 and 26 h reflect a circadian rhythm in the temperature.

Using the dominant period found by the ACF, standard (linear-regression) cosinor analysis (40) was performed to estimate the amplitude, mesor, and phase parameters for each processed time series. The estimates of phase are not reported because they are not useful for comparison across subjects with different periods. In addition, the lower the amplitude, as is the case with CTR, the less accurately the phase can be determined (50).

Procedural stress analysis. To determine the effect of procedural stress on CTR, we separated each subject's time series into two subsets: the first 3 days after instrumentation and subsequent days. The standard cosinor analysis, assuming a 24-h period, was performed on the subsets of data to compare differences in the CTR parameters. The closeness of the data to the assumption of a 24-h rhythm was evaluated by the "percentage of rhythm," a goodness-of-fit index (GFI) that can be computed with the standard cosinor analysis procedure. Although others (25) used 10% as the criterion for the GFI for CTR, the criterion appears to be subjective. The lower the percentage of the rhythm that fits the superimposed cosine curve, the less valid is the assumption of a 24-h period and, conversely, the higher the percentage of the rhythm that fits the cosine curve, the greater is the validity of the assumption. Thus, a GFI of 0 would be interpreted as no fit between the data and the cosine curve, whereas a GFI of 1.0 would indicate a perfect fit. Using SPSS Version 12.0 for Windows (SPSS Inc., Chicago, Ill), the paired (two-tailed) t test was used to compare differences in amplitude, mesor, and GFI at $\alpha = 0.05$.

Ambient temperature analysis. Ambient temperature was analyzed using the same procedure described for porcine CTR profile.

Results

Descriptive data for the sample are shown in Table 1 for the nine growing–finishing domestic farm pigs. The temperature (mean ± standard deviation [S.D.]) for individual subjects varied from 37.63 ± 0.74°C to 40.16 ± 1°C. The sample mean temperature was 38.5 ± 0.71°C. All WBC counts were normal with the exception of that for subject 7, which was elevated.

Porcine CTR profile. Ear-based temperature was measured

Table 2. Circadian temperature rhythm parameters for growing–finishing domestic pigs

Subject	Amplitude (°)	Mesor (°)	Period (h)	Acrophase (military time)
1	0.18	38.15	24.4	17:50
2	0.14	38.55	24.5	21:18
3	0.13	38.77	23.1	17:22
4	0.08	40.33	24.1	22:06
5	0.22	38.55	25.9	21:18
6[a]	0.31[a]	37.83[a]	20.5[a]	22:25[a]
7	0.14	39.13	22.9	20:33
8	0.21	38.48	22.7	16:05
9	0.25	38.17	23.9	18:32
Mean (SE)	0.18 (0.02)	38.66 (0.24)	23.6 (0.50)	19:44

[a]indicates the pig with noncircadian period.

on consecutive days to characterize CTR. The amplitude, mesor, period, and acrophase of the temperature rhythm for each pig is shown in Table 2. The amplitude varied from 0.08 to 0.31 with a sample mean ± standard error (SE) of 0.18 ± 0.02°C. The mesor varied from 37.83 to 40.33°C with a sample mean of 38.66 ± 0.24°C. The period varied from 20.5 to 25.9 h with a sample mean of 23.6 ± 0.5 h. The acrophase, expressed in military clock time, varied from 16:05 to 22:25, with a mean at 19:44.

Procedural stress analysis. The data were divided into subsets for each farm pig, representing the first 3 days of monitoring and subsequent monitoring days, to determine the effect of procedural stress on CTR. Table 3 shows the data to compare the CTR parameters and GFI for each pig in the first 3 days after instrumentation under general anesthesia with subsequent days of temperature monitoring. For seven of the nine (78%) pigs, amplitude was lower during the first 3 days after the procedure than during subsequent days. The sample mean was 0.14 ± 0.02°C and 0.22 ± 0.04°C respectively; the difference between the amplitudes was statistically significant ($t = -2.5$, $df = 8$, $P = 0.03$). The mean mesor was the same and GFI was not statistically different between data subsets. The correlation between amplitude and GFI was moderate for the 3-day adaptation period ($r = 0.56$, $n = 9$, $P > 0.05$) and strong for the subsequent monitoring days ($r = 0.70$, $n = 9$, $P = 0.035$). The mean acrophase was not materially different between data subsets, but there was large intraindividual variability in five (56%) pigs. In our data sets, GFI ≥ 20% is associated with a 24-h period based on visual inspection of a cosine curve superimposed on the raw data. Four (44%) of the pigs had a GFI < 20% during the first 3 days after the instrumentation procedure, and two of those pigs also had GFI < 20% in the subsequent data set.

Ambient temperature characteristics. We analyzed 20 days of ambient data to determine whether the ambient environment synchronized porcine CTR. Ambient temperature varied from 21.6 to 24.9°C, with mean (± S.D.) of 23 ± 0.6°C. Relative humidity varied from 16% to 68% (mean, 41% ± 11%). Times of minimum and maximum values varied widely across the ambient data sets for both temperature and relative humidity. Only one data set (4-day duration) had evidence of CTR, with a period length of 24.4 h, amplitude of 0.09°C, mesor of 23.3°C and acrophase of 12:49. This data set was collected during the temperature monitoring of pigs 6 and 7, which had CTR periods of 20.5 h and 22.9 h respectively.

We examined the ambient data sets for changes in temperature and humidity during cleaning of the housing room and pens. The relative humidity rose and ambient temperature dropped or rose transiently during the cleaning times. The mean increase in relative humidity was 20.7%, and the mean change in ambient temperature was 0.3°C; the average time of change was 37 min, with the humidity gradually and steadily declining over the subsequent 45 min. Cleaning time was variable from day to day, between 08:30 and 15:30.

Discussion

To our knowledge, these findings represent the most detailed characterization of CTR in laboratory swine published to date. Eight of the nine subjects (89%) had a circadian rhythm in body temperature; one subject had an ultradian rhythm (< 22 h). Heretofore, porcine CTR investigators have determined circadianicity from graphic display of raw data and differences between minimum and maximum values in a 24-h period (7, 29, 30). Bond and colleagues (7) reported rectal temperature data for swine with an average weight of 53 kg housed in a controlled-temperature laboratory. The mean diurnal change varied from 0.7 to 1.6°C, with greater diurnal variability at higher ambient temperatures. Although Ingram and Mount (30) did not report temperature values, graphic recordings of core body temperature in one pig showed equivalent diurnal changes across three different feeding patterns. Diurnal variability is critical to CTR; however, it may be independent of the rhythm period and, thus, is insufficient for characterizing CTR.

Fully automated sampling methods, advances in computational ability, and new analytic approaches now permit more refined analysis of dense data and long data sets than was possible 35 years ago. Hahn and colleagues have used modern techniques to contribute substantively to our knowledge of ambient temperature influences on animal performance but have not focused directly on the pig's CTR (20). Therefore, our present results appear to represent the first published data characterizing CTR in the domestic farm pig since the work of Ingram and colleagues in the 1970s. As such, our findings provide preliminary normative CTR data for laboratory-housed, unrestrained, growing–finishing swine.

For 10 to 14 days before the experiment, all of our subjects were synchronized to a 12:12-h light:dark cycle, one feeding at midday, and a controlled environment and were expected to have a circadian rhythm in body temperature. Yet, one pig (11%) had an altered rhythm with a period that exceeded the boundaries of CTR. Nagayama (37) recommended that strict conditions be maintained when measuring circadian rhythm in animals, including constant ambient temperature and humidity, muted noise, and random cleaning times under dim red light. Whereas ambient temperature was well controlled in our experiment, relative humidity was variable. Furthermore, we did not attempt to mute noise or randomize cleaning time of the housing room. It is unlikely that such deviations from "strict" conditions explain the altered rhythm in subject 6 because the littermate of this pig (subject 7) was monitored at the same time in an adjacent pen. Monitoring start and end times varied by < 24 h for this littermate pair. Therefore, within-subject factors would seem to offer a more plausible explanation of the findings in subject 6 than environmental factors. Halberg and colleagues (21) suggested that alterations in CTR amplitude or period may precede elevation of mesor, thereby serving as a warning signal of illness or the sole sign of subclinical illness. Future research would benefit from linking the CTR profile with health status.

Table 3. Circadian temperature rhythm parameters and goodness-of-fit index (GFI) of nine growing–finishing domestic pigs for the first 3 days after the instrumentation procedure and subsequent monitoring days

	First 3 days				Subsequent days			
Pig	Amplitude (°C)	Mesor (°C)	Acrophase (military time)	GFI	Amplitude (°C)	Mesor (°C)	Acrophase (military time)	GFI
1	0.08	38.14	18:50	15%	0.2	38.14	18:04	34%
2	0.16	38.54	0:02	49%	0.1	38.54	18:10	23%
3	0.04	38.78	17:54	2%	0.08	38.77	16:08	9%
4	0.08	40.33	20:09	16%	0.05	40.33	23:41	5%
5	0.16	38.53	19:11	34%	0.24	38.57	22:42	38%
6[a]	0.24[a]	37.8[a]	1:11[a]	18%[a]	0.45[a]	37.85[a]	20:26[a]	35%[a]
7	0.13	39.2	20:07	31%	0.3	39.18	5:22	34%
8	0.16	38.44	16:56	36%	0.26	38.48	15:17	55%
9	0.23	38.17	18:14	34%	0.27	38.18	18:14	38%
Mean	0.14	38.7	20:04	26%	0.22	38.7	20:14	30%
(95% CI)	(0.10, 0.18)	(38.2, 39.2)			(0.14, 0.30)	(38.2. 39.2)		

CI, confidence interval.
[a]indicates the pig with a noncircadian period.

We used a blind approach to thermistor placement, and it is possible that the sensor was improperly positioned in subject 6. Hahn and colleagues (27) inserted foam into the ear to secure the sensor and we did not. Considering that our one pig with a noncircadian period had the lowest mesor recorded across subjects, dislodgment of the sensor is a plausible explanation. The thermal gradient along the ear canal in pigs is unknown, but in humans the gradient can be as much as 0.8°C between the external opening and tympanic membrane (10). Therefore, the thermistor tip could have been located too distant from the tympanic membrane, and unreliable measurement from sensor movement could have resulted in an altered CTR profile.

On the basis of the dominant period finding in subject 3, we incorporated a WBC count into our protocol to rule out a subclinical infectious process. The WBC was normal in subject 6 at the time of instrumentation, but that finding does not exclude the development of subclinical illness during the ensuing 6 days. As shown in Table 1, only subject 7 had an abnormal WBC, and its temperature rhythm was circadian. Subject 4 had the lowest amplitude and highest mesor in the sample—a constellation of parameters that theoretically suggests disease—yet, its CTR period and WBC were normal. The inconsistent relation between WBC and CTR amplitude in this study is not readily explainable, and further research is recommended with more frequently monitored and sensitive measures of inflammation and infection.

In our sample, the rhythm-adjusted mean (mesor) was 38.7°C, equivalent to the 39°C reported as the normal temperature of the pig both in its natural environment and in the research laboratory with one daily feed (30). Likewise, the low mean CTR amplitude we identified is consistent with low amplitudes in the pig reported by others for melatonin and cortisol circadian rhythms (31). Despite the low amplitudes, body temperature, melatonin, and cortisol appear to be strong oscillators in swine (31, 48, 50).

Chronobiology experts report that amplitude is attenuated in stress and illness (47). We chose a noninvasive method of temperature measurement because surgery disrupts CTR (12, 13). Gegout-Pottie and colleagues (15) reported that a 3-day adaptation period is required for re-synchronization of CTR after surgical implantation of a transmitter. We note that, whereas mesor was identical in our split data sets, amplitude was lower during the first 3 days after placement of the ear-based sensor. This finding suggests that the animal was stressed by transportation to and from the procedure room, anesthesia, or the novelty of the probe and vest. Therefore, our noninvasive ear-based method offers no advantage over transmitter implantation in terms of adaptation time. Time of CTR adaptation is of interest for many reasons related to pig husbandry and both clinical and research issues, and more study is needed to precisely identify adaptation responses to various stressors. Nonetheless, it appears to be important to monitor CTR with a longitudinal time-series design to allow for adaptation to stress. CTR profiles measured without an adaptation window or a sufficiently long time-series to distribute the effects of stress are likely to have low validity. This point is illustrated by comparing the amplitudes in Tables 2 and 3, where the mean amplitude of the entire dataset is between the mean amplitude of the subsets of data.

Ambient temperature was within the recommended range for housing farm animals (39) and growing–finishing swine (8, 42). However, we measured temperature of the macroenvironment, and variation between the microenvironment (i.e., individual subject's pen) and housing room conditions can be considerable (39, 41). Only one of our ambient temperature data sets had a 24-h rhythm. Because our ambient temperature values were stable across time within a 3.3°C range, the variation may have been insufficient to serve as a synchronizer of CTR in our subjects. Entrainment of CTR requires large-amplitude variation in intensity of the synchronizer (11, 23), and the amplitudes we calculated were small. Bruce (9) suggested that a total range of 5°C ordinarily will entrain CTR. Compared with those housed indoors, animals living outdoors would be subjected to a more variable, and thus higher-amplitude, ambient temperature rhythm. However, in free-ranging eland, Fuller and colleagues (14) did not find a consistent relation between daily variation in microenvironment ambient temperature and body temperature. In the housing conditions of our research laboratory, there was no evidence that circadian rhythm in the macro-environment temperature synchronized pig temperature to a circadian profile. Measurement of the microenvironment temperature may clarify the role of ambient conditions as a synchronizer of CTR in laboratory swine.

Whereas the average relative humidity was within the range recommended for laboratory swine (39, 42), the variation was greater than recommended, with the minimum value lower than the recommended 30% minimum. In terms of animal welfare, wide variation in relative humidity is less important at the observed ambient temperatures than at higher ones where evaporation is the only mechanism available to the animal for heat loss (6). Although poor control of humidity may have affected our CTR findings, relative humidity is a less effective synchronizer than ambient temperature in homeotherms (3, 50).

We conclude that domestic farm pigs between 4 1/2 and 5 months of age that are housed in a research laboratory have circadian rhythm in body temperature. The CTR is characterized by a low amplitude, mesor that is equivalent to the normal temperature of swine, and acrophase in the evening. Explanations for absence of CTR should be sought and might include illness and alterations in sleep–wake cycles and other synchronizers of CTR. Even a noninvasive instrumentation protocol appears to cause stress, and further research is needed to more precisely define the time frame for adaptation to stress. Ambient temperature that varies by ≤ 3.3°C within a 24-h period does not synchronize porcine CTR. Future research will establish the value of CTR to monitor porcine stress and adaptation patterns, and the findings presented here provide a beginning foundation for doing so.

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