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—Original Article—

Prepartum change in ventral tail base surface temperature in beef cattle: comparison with vaginal temperature and behavior indices, and effect of ambient temperature

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Abstract. Prediction of parturition is essential for sustainable production in beef and dairy cattle, yet the present methods are limited by their high invasiveness and low utility. Here we compared prepartum changes in ventral tail base surface temperature (ST) with changes in vaginal temperature (VT) and behavioral indices. We analyzed 22 parturitions from 22 beef cows. Changes in daily values of ST, VT, and behavioral indices over the 7 days before parturition were investigated. Hourly values were calculated as the actual values minus the mean values for the same hour over a 3-day period, and the changes in hourly values over the 48 h before parturition were investigated. To test the effect of ambient temperature, tested cows were assigned to two season-groups based on the ambient temperature to which they were exposed (warm: n = 13; cool: n = 9), and the daily and hourly values of the indices were compared between seasons. A decrease in ST occurred approximately 30 h before parturition, which was similar to the time of the decrease in VT and earlier than the increase of behavioral indices. In addition, a unique fluctuation of ST observed in the last few hours before parturition indicates that ST could provide a sign for parturition not only in the long-term like VT, but also in the short-term like behavioral indices. Although ST was more sensitive to ambient temperature than VT or the behavioral indices, the day of parturition could be predicted from ST in both the warm and cool seasons.

Key words: Beef cattle, Behavior, Body surface temperature, Parturition, Vaginal temperature

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Prediction of calving time is one of the essential aspects of reproduction management in order to realize sustainable production in beef and dairy cattle. In Japan, the rate of incidence of dystocia and stillbirth have been reported as 8.55% and 2.46% in beef cattle [1] and 1.40% and 5.06% in dairy cattle [2], respectively. The occurrence of dystocia not only contributes to an increase in the mortality of newborn offspring but also impairs the postpartum performance of calves and dams, resulting in an economic loss [3]. Precise prediction of calving time allows for assistance with the calving as necessary, which decreases the risk of dystocia, stillbirth and several other troubles associated with parturition. However, monitoring prepartum cows by direct observation has become more time-consuming and labor-intensive along with the expanding herd sizes, resulting in a decrease in labor time per cow.

To overcome this problem, several technologies for the automatic prediction of calving time have been developed and are already on the market (reviewed in [4]). There are various signs of parturition (e.g., relaxation of the pelvic ligaments, secretion of vaginal mucous, edema of the udder and vulva, filling of the teats and relaxation of the tail [5]), and these commercial devices mainly use behavioral or body temperature changes as indicators of calving prediction. Many behavioral changes occur around the time of parturition, including changes in the duration of lying, standing, feeding, drinking and ruminating, and in the frequency of steps, posture change and tail raising [6–10]. The calving prediction devices can quantify these behavioral changes by means of connected sensors (e.g., collar, pedometer, ear-tag, and tail-belt sensors). On the other hand, core body temperature significantly decreases before parturition, and this decrease is suggested to be mainly triggered by a change in the maternal plasma progesterone concentration [11, 12]. Vaginal temperature (VT) has been primary index used for calving prediction because it is relatively easy to measure and suitable for continuous measurements using an indwelling sensor which remains in the cow's vagina until the time of amniorrhexis and delivery [13]. However, VT measurement inevitably involves invasiveness and a risk of inflammation of the vaginal wall, which limits its use on commercial farms [14]. Therefore, alternative approaches are needed for the body-temperature measurement in calving prediction.

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Recently, a new type of wearable wireless sensor was developed to continuously measure the ventral tail base surface temperature (ST; [15, 16]), which is highly correlated with rectal temperature. Compared to the other measurements of body temperature, monitoring ST is less invasive and therefore probably less stressful to the animal, since the sensor can be easily attached to and detached from the body surface without paying special attention to hygiene issues.

Miura *et al*. [17] first reported the application of ST measurement for estrus detection in beef and dairy cattle, and then Koyama *et al*. [18] used the ST sensor to predict the calving time of dairy cattle. However, as an index for calving prediction, it remains unclear whether ST would compare favorably to other indices derived from conventional animal-attached sensors. Moreover, since combining several indices from different sensor data may maximize the performance of calving prediction in cattle [19], the pros and cons of using heterogenous sensors should be clarified.

The object of the present study was to compare ST with other animal-attached sensor measurements during the prepartum period in beef cattle. We investigated the following: (1) the relation between ST and VT, (2) the relation between ST and the behavioral indices derived from leg-attached device, and (3) the effect of ambient temperature on ST.

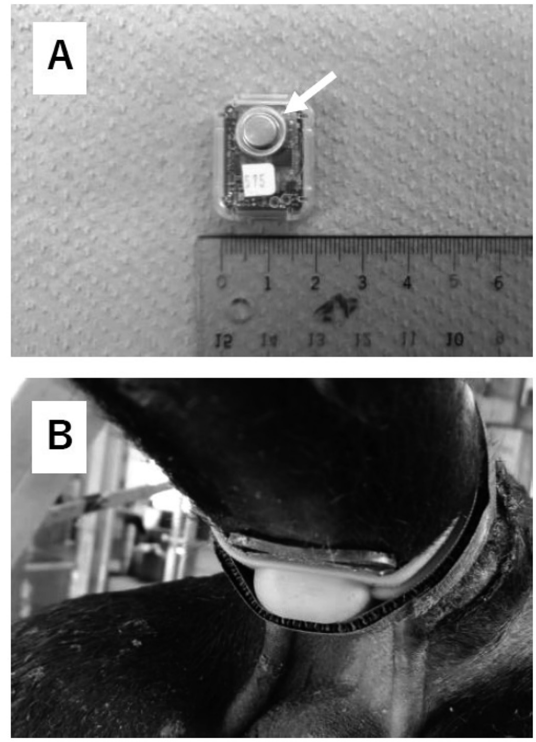

Fig. 1. Appearance of the surface temperature sensor. (A) Sensor enclosure with a thermistor. The sensor measures the surface temperature at the point of contact with the thermistor (white arrow). (B) The sensor is attached to the surface of the ventral tail base of the cow using a custom-made belt. The sensor enclosure is contained in the belt and the thermistor sits on the surface of the ventral tail base.

Materials and Methods

Animal care

All procedures in the present animal experiments were approved by the Animal Care and Use Committee of the Institute of Livestock and Grassland Science (NILGS), National Agriculture and Food Research Organization (NARO).

Animals

All experiments were conducted at NILGS, NARO (Nasushiobara, Tochigi, Japan; 36.9 N, 139.9 E) from December 2017 to November 2018. A total 24 parturitions from 24 beef cows were used for data collection. The tested cows were Japanese Black (JB; n = 18) and crossbred cows (Japanese Black bull × Holstein cows, F1; n = 6) (age: 67.3 ± 33.0 months; body weight: 606.7 ± 115.2 kg; parity: 2.1 ± 1.9 including 4 primiparous cows; means ± SD). Most of the JB cows were bred by artificial insemination (n = 17); the other JB (n = 1) and all F1 cows (n = 6) were bred by embryo transfer. Each cow was moved to a maternity pen (4 m × 4 m) by at least 7 days (d) prior to the due date; at the same time, multiple sensors were attached for the collection of ST, VT and behavioral index data and remained in place until delivery. Cows in the maternity pens were given hay (3.0–4.0 kg) and concentrate (1.0–1.5 kg) twice per day (0900 h and 1600 h) and had free access to water. Three cows received calving induction treatments (500 mg dexamethasone i.v., 4 mg estradiol i.m. (Kyoritsu Seiyaku, Tokyo, Japan), and 500 µg cloprostenol i.m. (Nippon Zenyaku Kogyo, Tokyo, Japan)) and seven deliveries were assisted.

Ventral tail base surface temperature

The ventral tail base surface temperature of the cows was measured using a modified version of the sensors described in previous reports [17, 18] (Fig. 1A). The sensor measured 25 × 20 × 10 mm, and weighed 5.87 g with a battery (CR1220) inserted. The thermistor of the sensor was placed on the surface of the ventral tail base, and the sensor was fixed to the tail using a custom-made silicon belt. The belt including the sensor was fastened securely with a hook and loop fastener and covered with an elastic medical bandage (Elatex 11885; ALCARE, Tokyo, Japan) to stabilize its position (Fig. 1B). Ventral tail base surface temperature was measured in increments of 0.01ºC every 2 min through August 2018 and then every 10 min from September 2018 due to an update of the sensor firmware. Recorded ST data were automatically transmitted to a receiver (920 MHz) and stored on a local PC or cloud server via 3G/LTE.

Vaginal temperature

For comparison with ST, the VT was recorded using a temperature data logger (Thermoclone type SL; KN Laboratories, Osaka, Japan) combined with a modified progesterone-free vaginal implant device (CIDR blank; InterAG, Hamilton, New Zealand) according to Sakatani *et al*. [20]. The device was soaked in 0.1% benzalkonium chloride solution before insertion. The vulva of the cow was washed, and then the device was inserted into cow's vagina using a CIDR applicator. The vaginal temperature was recorded in increments of 0.1ºC every 10 min. The device was spontaneously ejected from the vagina upon amniorrhexis. The data logger was then recovered, and the VT data were exported using the software (RhManager; KN Laboratories).

Behavioral data

Behavioral data were recorded using a leg-attached device (IceTag sensor; IceRobotics, West Lothian, UK). The device uses a 3-axis accelerometer and provides the following indices at 1-min intervals: the motion index, step count, number of lying bouts, and duration of standing and lying (%). The motion index is a proprietary index defined by IceRobotics and is considered to reflect the overall activity level of animal. Lying bouts were defined as posture transitions from standing to lying. Although the methods used to calculate these indices were not disclosed, the device has previously been validated with observation data [21–23], and consequently these indices were considered sufficiently accurate for behavioral assessment. The device was attached at the left hind leg of the tested animal with a strap. The recorded behavioral data were downloaded wirelessly to a PC via IceReader (IceRobotics, West Lothian, UK) and processed using the provided software (IceManager; IceRobotics). Of the indices noted above, the motion index, step count, number of lying bouts, and standing duration were adopted as behavioral indices in the present study; lying duration was not used because it was merely the inverse of standing duration, and thus the results for this index could be inferred from the standing duration.

Throughout the experiments, the tested cows were visually recorded with an infrared video camera (WTW-AR27HJP; Panasonic, Osaka, Japan) installed at a maternity pen, and the calving time was determined from the video data. A calving was defined as the complete expulsion of the fetus body from the vagina.

Meteorological data

Ambient temperature was recorded in increments of 0.1ºC every 10 min by a data logger (TR-72wf; T&D Corporation, Matsumoto, Nagano, Japan). After the experiment, the data were transmitted to a computer (T&D Recorder; T&D Corporation).

Data processing

All sensor data (ST, VT, and behavioral indices) over the last 7 d before parturition in each cow were synchronized with the relative time assuming the time of parturition was zero, and then summarized in the daily and hourly representative values based on the relative time (i.e., hours or days before parturition). Finally, the data for all cows were entered onto the timeline used in the previous studies [7, 24] and applied to the following statistical analysis.

Before the synchronization, ST data above 39.0ºC and below 35.0ºC were removed as outliers. Standing duration was converted from % to min. In ST and VT, hourly maximum values were calculated as hourly representative values, and hourly integrated values in behavioral indices were calculated. Daily representative values were also calculated from the hourly representative values; the average values for each hour throughout the day were used for ST and VT, and the combined values for an entire day were used for behavioral indices. The meteorological indices per each 10 min interval were also converted to hourly and daily average values. The values at –1 h correspond to those from –60 to –1 min before parturition, and the values at –1 day refer to those from –24 h to –1 h until parturition. To remove the effect of the circadian variations, changes in the hourly values of the indices were expressed as actual values minus average values for the same hour over the preceding 3 days [7, 17, 18, 25].

Finally, the daily values of behavioral indices (motion index, step count, number of lying bouts, and standing duration) were transformed into a natural logarithm for normalization using the additive constant one due to the zeros obtained and were used for the following statistical analysis. The daily values of body temperature (ST and VT) and meteorological indices, hourly residual values of the body temperature and behavioral indices, and the hourly meteorological index were used in the following statistical analysis without transformation.

Statistical analysis

To investigate the relationship between the body temperature and behavioral indices, we first calculated Pearson's correlations among body temperature and behavioral indices using hourly data over the last 7 days before parturition. In addition, we evaluated the daily change of the body temperature and behavioral indices through the last 7 days before parturition, using the following mixed linear model (model 1): $Y_{ijk} = \mu + Day_i + id_j + e_{ijk}$, where Y_{ijk} are the daily representative values of the body temperature and behavioral indices, μ is the overall mean, Day_i is the fixed effect of the day before parturition (–7 to –1 day), id_j is the random effect of individuals, and e_{ijk} are the residual errors. The differences between the days before parturition were analyzed using the least squares means with the Tukey–Kramer adjustment. We also evaluated the hourly change of the body temperature and behavioral indices through the last 48 h before parturition using the following mixed linear model (model 2): $Y_{ijk} = \mu + Hour_i + id_j + e_{ijk}$, where Y_{ijk} are the hourly residual values of the body temperature and behavioral indices, μ is the overall mean, $Hour_i$ is the fixed effect of the hour before parturition (–48 to –1 h), id_j is the random effect of individuals, and e_{ijk} are the residual errors. In addition, the change in residual values was investigated using the least squares means with one-sample *t*-test, on the assumption that the residual values of the body temperature and behavioral indices are almost zero when parturition does not come. When a significant increase or decrease in an index was seen in two consecutive hours, the first hour was regarded as the turning point when the indices began to change.

To investigate the effect of ambient temperature on the body temperature and behavioral indices, we first calculated Pearson's correlations of body temperature and behavioral indices with ambient temperature using daily data over the last 7 days before parturition. For a detailed analysis, the tested animals were divided into two season-groups based on the average ambient temperature to which they were exposed over the last 7 days before parturition. The temperature threshold followed Koyama *et al*. [18]; the periods during which the average ambient temperature ≥ 10ºC and < 10ºC were defined as a warm (n = 13; 18.3 ± 3.2ºC, means ± SD) and cool (n = 9; 3.2 ± 2.8ºC, means ± SD) season, respectively. We evaluated the daily change of the body temperature and behavioral indices through the last 7 days before parturition in different seasons using the following mixed linear model (model 3): $Y_{ijkl} = \mu + Day_i + Season_j + (Day \times Season)_{ij} + id_k + e_{ijkl}$, where Y_{ijkl} are the daily representative values of the body temperature and behavioral indices, μ is the overall mean, Day_i is the fixed effect of the day before parturition (–7 to –1 h), $Season_j$ is the fixed effect of the season (warm, cool), $(Day \times Season)_{ij}$ is the interaction between the day before parturition and season, id_k is the random effect of individuals, and e_{ijkl} are the residual errors. We also evaluated the hourly change of the body temperature and behavioral indices through the last 48 h before parturition in different seasons using the following mixed linear model (model 4): $Y_{ijkl} = \mu + Hour_i + Season_j + (Hour \times Season)_{ij} + id_k + e_{ijkl}$, where Y_{ijkl} are the hourly residual values of the body temperature and behavioral indices, μ is the overall mean, $Hour_i$ is the fixed effect of the hour before parturition (–48 to –1 h), $Season_j$ is the fixed effect of the season (warm, cool), $(Hour \times Season)_{ij}$ is the interaction between the day before parturition and season, id_k is the random effect of individuals, and e_{ijkl} are the residual errors. The change in residual values through 48 h in each season was also investigated using the least squares means with one-sample *t*-test as well as model 2.

The Pearson's correlation coefficients were calculated with PROC CORR procedure and statistical analyses with mixed linear model were performed using the MIXED procedure of SAS 9.4 (SAS Institute, Cary, NC). Differences were considered significant when $P < 0.05$, and nonsignificant trends were reported when $0.05 \leq P < 0.10$.

Results

A total of 24 parturitions were observed during the experiments. The data from two cows were excluded from further analysis because the calving times were indeterminate due to a failure in video recording. In addition, some device data were also excluded from the subsequent analyses because they were incomplete. Finally, the data from 22 parturitions were used in the analysis (ST: n = 22; VT: n = 17; behavioral data: n = 17). The reception rate of hourly ST data was 85.3 ± 17.4% (means ± SD) throughout the whole experiments. There was no dropout or failure of the ST sensor throughout the 7 days before parturition, and the low data reception rate of hourly ST data was attributed to the low performance of the receiver and the distance between the sensor and the receiver.

During the experiments, calving was induced in three cows and seven deliveries were assisted. All deliveries had a single birth, and two were stillbirths. The gestation periods and calf sizes were 287.4 ± 4.6 days and 34.8 ± 5.3 kg, respectively (means ± SD). All data related to calving induction, assisted and still birth deliveries were included in the analyses.

Change in body temperature and behavior indices before parturition

The Pearson's correlation coefficients among body temperature and behavioral indices over the last 7 days before parturition are shown in Table 1. Through the last 7 days before parturition, a moderate or high positive correlation were only found between body temperature indices or between behavioral indices.

Daily changes in body temperature and behavior indices through the last 7 days before parturition are shown in Table 2. The body temperature and behavioral indices exhibited significant change on each of the 7 days before parturition. ($P < 0.05$). Although the daily values of ST were always lower than those of VT throughout the 7 days before parturition (ST: 37.9 ± 0.4ºC; VT: 38.9 ± 0.2ºC; means

Table 1. Pearson's correlations among hourly values of body temperature and behavioral indices before parturition

Index	VT	Motion index	Step count	Lying bouts	Standing duration
ST	0.56 *	–0.09 *	–0.06 *	–0.08	–0.01
VT		–0.07 *	–0.06 *	0.01 *	–0.03
Motion index			0.93 *	0.47 *	0.84 *
Step count				0.45 *	0.94 *
Lying bouts					0.37 *

ST: ventral tail base surface temperature; VT: vaginal temperature. The results of statistical analyses in behavior indices are obtained using the values coverted by a natural logarithm (ln(x + 1)) for normalization. * $P < 0.05$.

Table 2. Change in daily values of body temperature and behavioral indices before parturition

Index	n	Days before parturition							P-value
		–7	–6	–5	–4	–3	–2	–1	
ST (°C)	22	37.9 ± 0.1 [ab]	38.0 ± 0.1 [ab]	38.0 ± 0.1 [ab]	38.0 ± 0.1 [ab]	38.0 ± 0.1 [a]	37.8 ± 0.1 [b]	37.4 ± 0.1 [c]	< 0.05
VT (°C)	17	39.0 ± 0.0 [a]	39.0 ± 0.0 [a]	39.0 ± 0.0 [a]	39.1 ± 0.0 [a]	39.1 ± 0.0 [a]	38.1 ± 0.0 [b]	38.5 ± 0.0 [c]	< 0.05
Motion index	17	1721 ± 340 [b]	1768 ± 333 [b]	1883 ± 333 [b]	1668 ± 333 [b]	1559 ± 333 [b]	1772 ± 333 [b]	5354 ± 333 [a]	< 0.05
Step count	17	702 ± 106 [b]	717 ± 104 [b]	712 ± 104 [b]	671 ± 104 [b]	647 ± 104 [b]	733 ± 104 [b]	2009 ± 104 [a]	< 0.05
Lying bouts	17	24 ± 7 [b]	26 ± 7 [b]	25 ± 7 [b]	22 ± 7 [b]	24 ± 7 [b]	22 ± 7 [b]	70 ± 7 [a]	< 0.05
Standing duration (min)	17	602 ± 30 [b]	610 ± 29 [b]	596 ± 29 [b]	605 ± 29 [b]	588 ± 29 [b]	649 ± 29 [b]	853 ± 29 [a]	< 0.05

ST: ventral tail base surface temperature; VT: vaginal tempearature. Values are expressed as least square means ± SEM. The results of statistical analyses in behavior indices are obtained using the values coverted by a natural logarithm (ln(x + 1)) for normalization. [a,b,c] Different letters in the same row indicate significant differences ($P < 0.05$).

± SD), both began to decrease 2 days before parturition and then fell rapidly by 0.4ºC on the day of parturition ($P < 0.05$). On the other hand, all behavioral indices remained constant until 2 days before parturition and clearly increased on the day of parturition ($P < 0.05$).

Hourly changes in body temperature and behavior indices through the last 48 h before parturition are shown in Fig. 2. The residual values of each index exhibited significant change on each of the last 48 h before parturition ($P < 0.05$). Residual values of ST and VT began to decrease significantly at approximately 30 h before parturition (ST: –32 h; VT: –36 h; Fig. 2A, B). Then, the residual values of VT turned upward at approximately 20 h (Fig. 2B), while those of ST remained almost constant (Fig. 2A). In addition, the residual values of ST decreased dramatically at approximately 4 h before parturition (Fig. 2A).

The residual values of the behavioral indices began to increase significantly at around 15 h before parturition (motion index: –14 h; step count: –14 h; lying bouts: –13 h; standing duration: –16 h; Fig. 2C, D, E, F). However, the changes in the values of standing duration were unstable until parturition (Fig. 2F). In addition, the residual values of motion index, step count, and lying bouts decreased at least a few hours before parturition (motion index: –3 h; step count: –3 h, lying bouts: –2 h; Fig. 2C, D, E).

The effect of ambient temperature

The Pearson's correlation coefficients of body temperature and behavioral indices with ambient temperature over the last 7 days before parturition are shown in Table 3. Ventral tail base surface temperature, motion index and step count were significantly correlated with ambient temperature ($P < 0.05$), while VT, lying bouts and standing duration were not.

For a detailed analysis, the tested animals were divided into two season-groups based on the average ambient temperature to which they were exposed over the last 7 days before parturition; a warm (n = 13; age: 56.4 ± 14.6 months; body weight: 547.4 ± 76.9 kg; parity: 1.5 ± 0.7 including two primiparous cows; means ± SD) and cool (n = 9; age: 72.7 ± 32.5 months; body weight: 680.1 ± 118.8 kg; parity: 2.4 ± 2.1 including two primiparous cows; means ± SD) season. Five and two delivers were assisted in warm and cool season, respectively. Changes in daily values of the body temperature and behavioral indices before parturition for the two different seasons are shown in Fig. 3. The values for all body temperature and behavioral indices showed significant change on each of the 7 days before parturition ($P < 0.05$) as seen in Table 2. In regard to the body temperature indices, VT was not significantly different between the seasons, while ST tended to be lower in the cool season ($P < 0.10$) and showed a significant interaction between day and season ($P < 0.05$). With respect to the behavioral indices, only step count was significantly lower in the cool season ($P < 0.05$).

Residual hourly values of the body temperature and behavioral indices in the last 48 h before parturition in the two different seasons are shown in Fig. 4. For all indices, the residual values showed significant change on each of the last 48 h before parturition ($P < 0.05$) as seen in Fig. 2. In body temperature indices, only the residual values of ST exhibited a significant effect of season ($P < 0.05$) and showed a tendency of interaction between hour and season ($P < 0.10$). Decreases in the residual values of ST and VT occurred simultaneously in the warm season (ST: –31 h; VT: –31 h; Fig. 4A, B), while those in the cool season had a temporal difference (ST: –26 h; VT: –36 h; Fig. 4A, B). Residual values of ST in the warm season were kept consistently lower until parturition. However, those in the cool season showed large variation and therefore were unstable until parturition.

With respect to the behavioral indices, the effect of season was not significant in any indices, and the interaction between hour and season was significant for motion index, step count, and lying bouts ($P < 0.05$) and exhibited a nonsignificant trend for standing duration ($P < 0.10$). Although the turning points at which the indices began to change (motion index: –11 h and –14 h; step count –11 h and –16 h; lying bouts: –5 h and –14 h; standing duration: –21 h and –16 h; Fig. 4C, D, E, F) and the degree of change in the indices in the last several hours before parturition differed between the warm and cool seasons, there was a consistent pattern of the indices increasing within the last day before parturition.

Discussion

Changes in body temperature indices before parturition

In the present study, we compared ST, VT and behavior indices and the effect of ambient temperature on these indices during the prepartum stage in beef cattle to test the potential of ST as an index for calving prediction. Although ST was approximately 1.0ºC lower than VT throughout the prepartum days, ST significantly correlated with VT ($r = 0.56$; Table 1), and the general pattern of change, the points where the decrease started, and the degree of decrease in both ST and VT before parturition were almost identical (Table 2, Fig. 2A, B). In their experiments in calves, Nogami *et al*. [15] demonstrated that ST was highly correlated with rectal temperature, while ST was 2–3ºC lower than rectal temperature. It has been reported that VT is correlated with rectal temperature [26, 27], and therefore our results indicated that ST would reflect the core body temperature in adult cows from prepartum to parturition as well as VT.

Maternal body temperature starts to decrease approximately 48 h before parturition [28, 29]. A previous report in dairy cows [18] showed that the values of ST began to decline approximately 36 h before calving, became constant approximately 18 h before calving and then declined again approximately 6 h before calving. The previously reported prepartum fluctuations in ST —an initial decrease, followed by temporary stagnation, and finally a second decrease in the last several hours— were also observed in our present experiments (Fig. 2A), suggesting the presence of a unique change in ST in both dairy and beef cows. In addition, the pattern of prepartum change in ST was partially different from that in VT; the value of VT decreased at approximately 30 h before parturition just as ST did, but then turned upward at approximately 20 h before parturition (Fig. 2B), while that of ST remained relatively constant over several hours and declined again in the last few hours before parturition (Fig. 2A). These differences between ST and VT may have been due to the difference of factors affecting both body temperature indices. Costa *et al*. [11] explained the mechanism of the drop of body temperature before calving as the result of a regulation of the obligatory and facultative thermogenesis, which was controlled by thyroid hormones, catecholamines and sexual hormones. Their report mentioned that the progressive increase in estrogen concentration in the final third of gestation stimulates

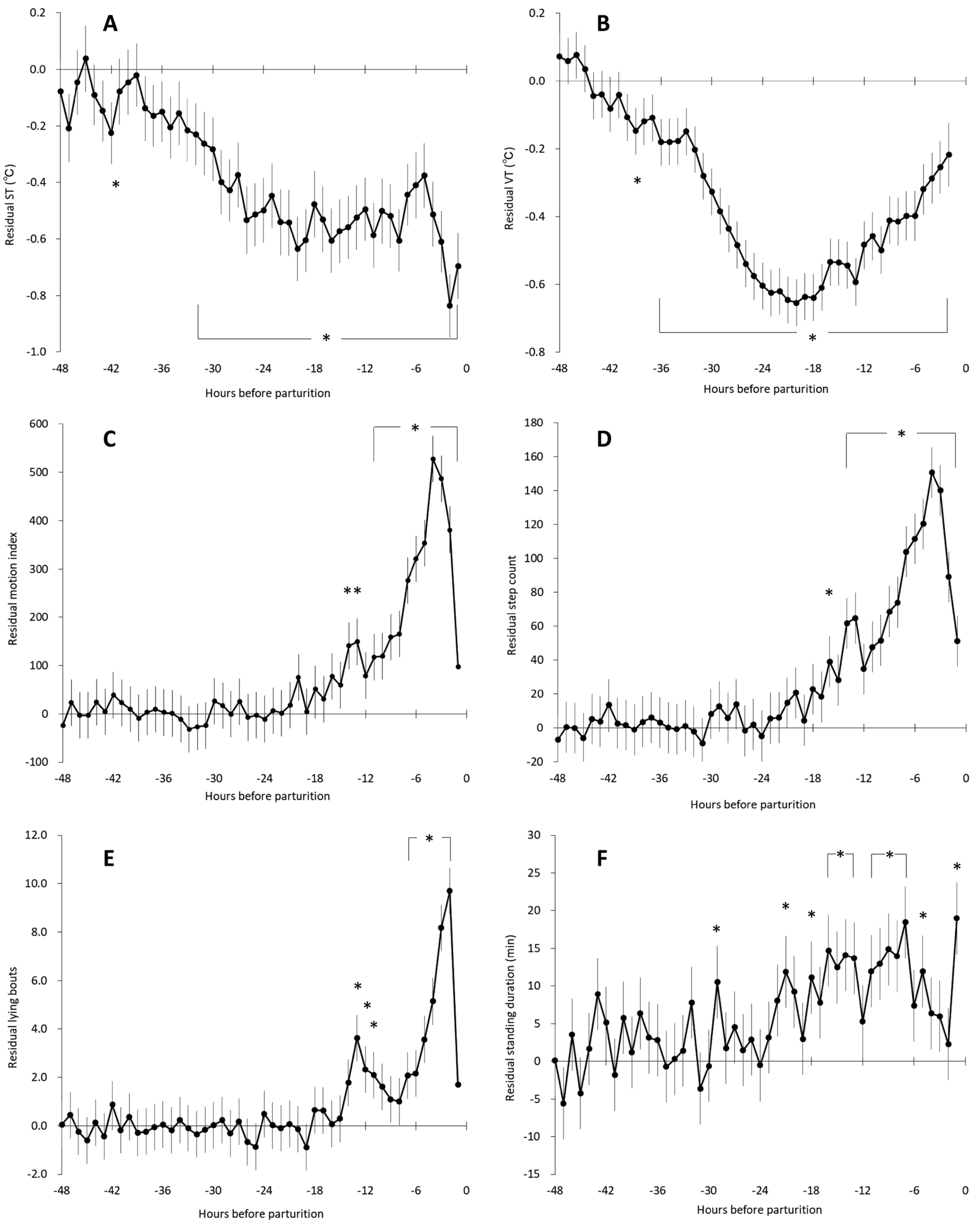

Fig. 2. Residual hourly values of body temperature and behavioral indices in the 48 h before parturition. (A) Ventral tail base surface temperature (ST; n = 22), (B) vaginal temperature (VT; n = 17) and the behavioral indices (C) motion index (n = 17), (D) step count, (E) lying bouts and (F) standing duration are shown. Values are expressed as least square means (± SEM). * Indicates a significant difference compared with zero ($P < 0.05$).

Table 3. Pearson's correlations among daily values of body temperature and behavioral indices with ambient temperature before parturition

	ST	VT	Motion index	Step count	Lying bouts	Standing duration
r	0.35 *	0.00	0.20 *	0.20 *	–0.02	0.08

ST: ventral tail base surface temperature; VT: vaginal tempearature. The results of statistical analyses in behavior indices are obtained using the values coverted by a natural logarithm (ln(x + 1)) for normalization. * $P < 0.05$.

heat-sensitive cells in the regulation of progesterone thermogenic modulation, favoring inhibition of cold-sensitive cells [30–32], which can cause a drop in body temperature. Vaginal temperature would mainly follow this mechanism, because it was widely thought to be equivalent to the core temperature [33]. On the other hand, ST is classified as peripheral temperature, which depends on the core temperature, environmental conditions, and peripheral blood system regulation [33]. Koyama *et al*. [18] assumed that the rapid decrease in ST in the last few hours before parturition resulted from a decrease in blood flow to the tail due to the foetus placing pressure on the blood vessels. Therefore, ST can be affected by blood flow around the tail in addition to changes in core temperature regulated by plasma hormonal concentration, which would result in a unique fluctuation of ST before parturition different from that of VT. In terms of actual use as body temperature indices, both ST and VT can be reliable substitutes for core body temperature, yet the differences in their own prepartum pattern in the last hours before parturition should be considered separately.

It should be noted that there is another difference between ST and VT in terms of the method of amniorrhexis detection. The occurrence of amniorrhexis is one of last events to be determined before parturition, and it is possible to infer delivery difficulty and the sex and weight of the fetus from the elapsed time from amniorrhexis to delivery [13]. Measurement of VT can provide information on when amniorrhexis will occur as the time when the vaginal-indwelling sensor is pushed out from the cow's vagina by the foetus. Although measurement of ST cannot detect amniorrhexis in the same manner, since the ST sensor is fixed on the tail and does not directly interfere with expulsion of the foetus, it could provide an urgent sign of parturition as the second decline in ST at the last hours before calving, as noted above (Fig. 2A). Therefore, like VT, measurement of ST would also provide both long-term (approximately 30 h before parturition) and short-term (approximately 4 h before parturition) prediction for calving time.

Furhtermore, unlike the vaginal-indwelling sensor, the ST sensor can be used in postpartum cows whose vaginas are recovering, and thus could be used to monitor maternal body temperature throughout the entire stage of reproduction. This approach would be advantageous for detecting the puerperium diseases associated with body temperature change (e.g., mastitis: [34, 35]; metritis: [36]; retained placenta: [37]).

Changes in behavior indices before parturition

Daily values of behavior indices —i.e., motion index, step count, lying bouts, and standing duration— obtained from the leg-attached device in the present study increased significantly on the day of calving (Table 2), just as the body temperature indices did. On the other hand, analysis of hourly data indicated that the prepartum changes in behavior indices occurred later than those in body temperature indices (Fig. 2); the residual values of behavioral indices started to increase at around 15 h before parturition (Fig. 2C, D, E, F) while those of the body temperature indices did so approximately 30 h before parturition (Fig. 2A, B). These results generally accorded with the previous reports using the same device (dairy cows: [7, 8]; beef cows: [38]). Most of the changes in behavior are observed on the actual day of calving [7, 8, 38], while changes in body temperature indices can be observed ≥ 24 h before parturition [28, 39].

In addition, the residual values of all the behavioral indices except standing duration rose at around 15 h before parturition, but then fall sharply in the last few hours before parturition (Fig. 2C, D, E). This change in the last several hours has been reported previously: an increase in lying bouts and standing time (equivalent to a decrease in lying time) in the final 2 h and the 2–4 h period before parturition were reported, respectively [4]. The calving process is generally divided into three stages: stage I (dilation of the cervix), stage II (delivery of the calf), and stage III (delivery of the placenta), and different behavioral signs were observed in each stage. Stage I is characterized by olfactory ground checks, nest-building-like behavior, self-body-licking, vocalization, discharge of feces, restlessness, and tail raising [40–42]. While some of these behavioral signs, such as tail raising, restlessness, and vocalization, extend into stage II, at the onset of stage II cows usually lie down as the calf enters the birth canal [41] to prepare for delivery of the fetus. In this context, the different behavior patterns in the first and second halves of the day of parturition can be attributed to a shift from stage I to II. Interestingly, as noted above, ST also showed further change in the last few hours before parturition (Fig. 2A). This phenomenon may have partly resulted from the tail raising behavior around parturition. Walls and Jacobson [43] reported that an increase in tail-skin temperature was often observed when the cows in their experiments were lying down with their tails beneath them; inversely, tail-skin temperature is expected to decrease when the tail remains elevated. The frequencies of both lying and tail raising behavior increase as parturition approaches and the cow prepares to expel the fetus body [40, 44], which may involve a decrease in ST in the last few hours before parturition. In any case, these changes in both indices in the last few hours before parturition can be good indicators of imminent calving.

The hourly residual values of standing duration were less stable than the other behavioral indices over the last 48 h prior to parturition, and did not exhibit any consistent trend (Fig. 2F). This result was similar to the finding of Jensen [25] that lying duration showed no specific trend over the hours before calving while lying bouts significantly changed during the final 12 h. Most cows lie down when undergoing parturition, but some cows remain standing during labor until parturition, and therefore it is reasonable to judge the imminence of parturition based on postural change (i.e., lying bouts) rather than the posture itself (i.e., standing or lying duration). In

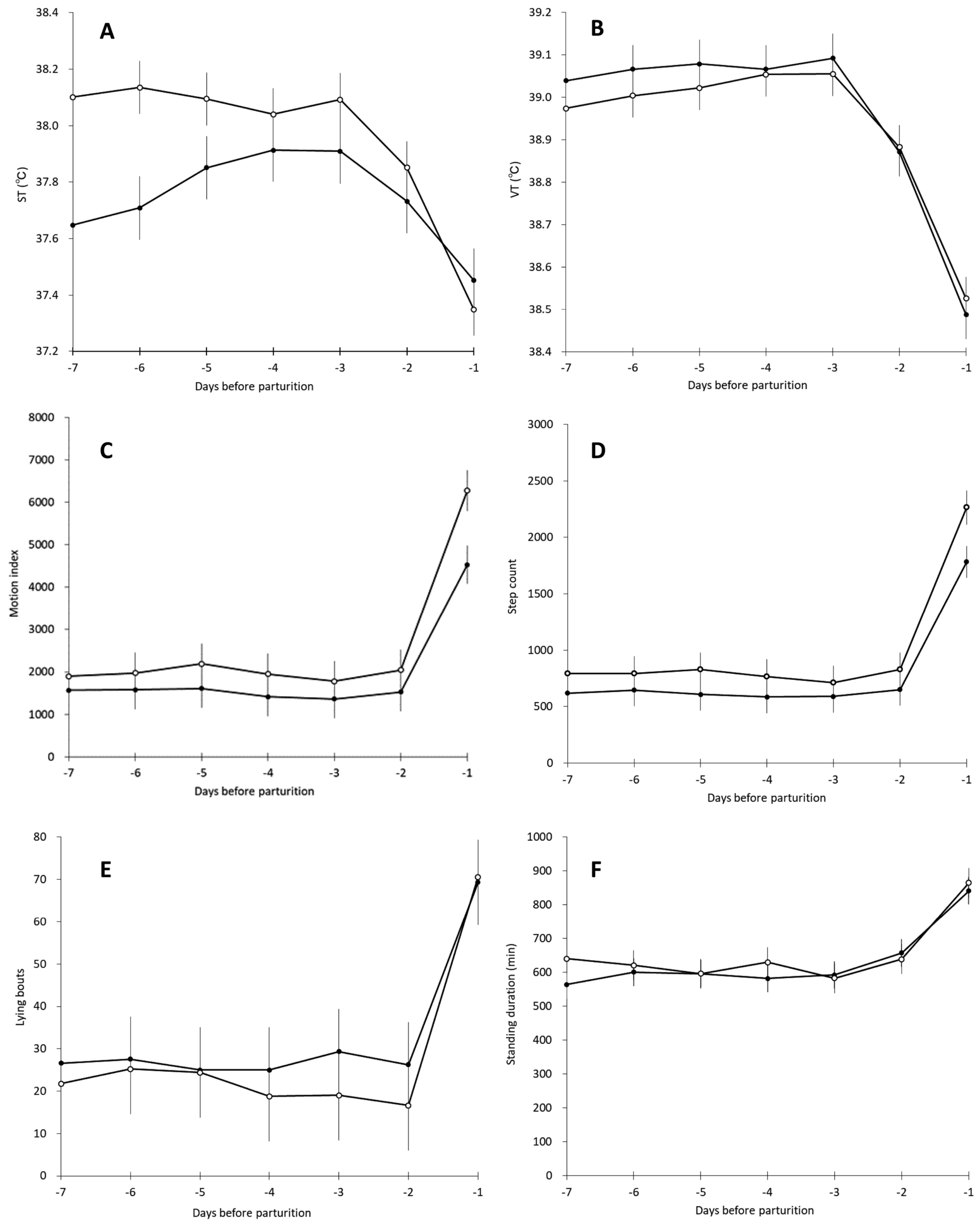

Fig. 3. Change in daily values of body temperature and behavioral indices until parturition in warm (n = 13, open circles) and cool (n = 9, closed circles) seasons. (A) Ventral tail base surface temperature (ST), (B) vaginal temperature (VT) and the behavioral indices (C) motion index, (D) step count, (E) lying bouts and (F) standing duration are shown. Values are expressed as least square means (± SEM).

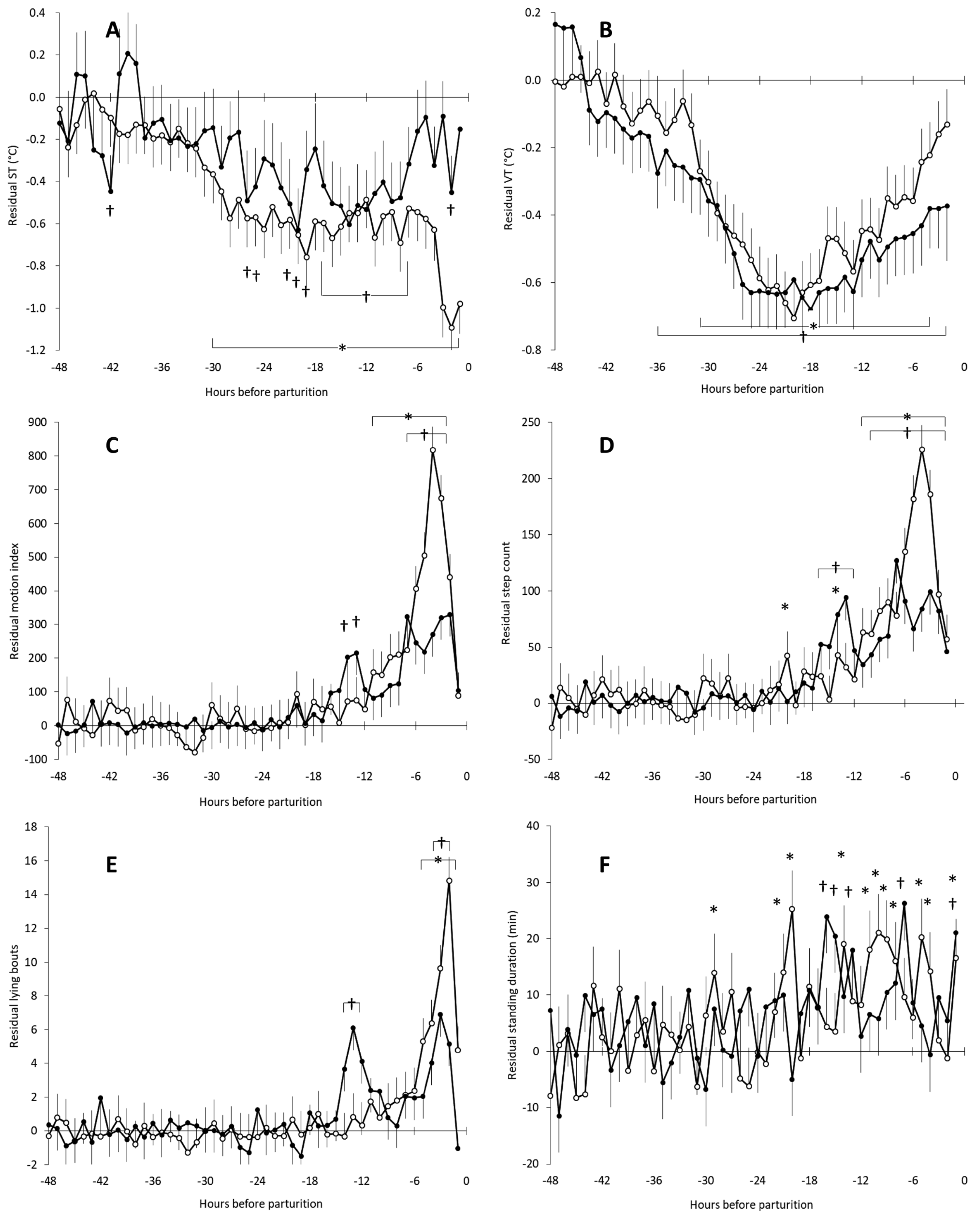

Fig. 4. Residual hourly values of body temperature and behavioral indices in the last 48 h until parturition in warm (n = 13, open circles) and cool (n = 9, closed circles) seasons. (A) Ventral tail base surface temperature (ST), (B) vaginal temperature (VT) and the behavioral indices (C) motion index, (D) step count, (E) lying bouts and (F) standing duration are shown. Values are expressed as least square means (± SEM). *,† Significant difference compared with zero in warm and cool seasons, respectively ($P < 0.05$).

addition, the present analyses included data on calving induction, assisted deliveries and still birth deliveries without separating them from the normal deliveries, and these human interventions, especially in the case of delivery assistance, can affect the cow posture in the last few hours prior to parturition. Moreover, behavioral indices have been associated with a variety of maternal factors such as dystocia [4] and parity [7], and therefore further research will be needed to understand the effect of these factors on the behavioral indices other than standing duration.

In the present study we used a leg-attached behavior sensor and only focused on locomotion activity indices around parturition, such as motion (motion index, step count, lying bouts) and posture (standing duration). On the other hand, other activity indices such as duration of feeding, ruminating and water drinking also change in the days and hours before parturition [7, 25] and would provide another clue for forecasting the parturition time. Therefore, we can infer that the results would be different if a collar-mounted behavior sensor that quantifies such neck motions were used. Moreover, the combined use of multiple devices to measure both behavioral and body temperature changes would provide a more accurate prediction of calving time [7].

Effects of ambient temperature

Of body temperature indices, ST were positively correlated with ambient temperature ($P < 0.05$; $r = 0.35$), while VT were not over the last 7 days before parturition (Table 3). In addition, when the tested animals were divided into two season-groups, the daily values of ST over the 7 days prepartum tended to be lower in the cool season than the warm season, while VT did not differ significantly between the seasons (Fig. 3), and the residual hourly values of ST were significantly lower in the cool season over the last 48 h before parturition, while those of VT were not (Fig. 4A, B). Furthermore, even the unique pattern of prepartum change in ST (i.e., second decrease in last several hours before parturition; Fig. 2A) didn't seen in cool season. On the other hand, of the behavior indices, motion index and step count correlated with ambient temperature ($P < 0.05$; r =, 0.20, and 0.20, respectively) while lying bouts and standing duration were not. When comparing two season-groups, only step count tended to be different between the warm and cool seasons in both the daily and hourly data analysis (Fig. 3 and 4C, D, E, F). The interactions between behavior indices and seasons were significant for each of motion index, step count, and lying bouts, and showed a nonsignificant trend toward significance for standing duration (Fig. 4C, D, E, F). These results imply that ST is more sensitive to ambient temperature than the other body temperature and behavioral indices.

Core body temperature is assumed to reveal the internal temperature of the body, and thus to be very close to the main organs, including the heart, brain, and viscera [33], and robust to effect of environmental conditions. In their experiments in non-lactating Japanese Black cows, Sakatani *et al*. [45] reported that the average VTs did not differ significantly between the cool and hot season in either the non-estrus or estrus periods. In contrast, ST is classified as a peripheral surface temperature, and thus would be intermediate between physiological phenomenon and environmental conditions [33], and would deviate from the core temperature in situations where the influence of the external environment becomes large. Made and Kreikemeier [46] compared the peripheral and core body temperature of the ear (ear surface and tympanic temperature, respectively) in heifers and reported that the average temperature of the ear surface in winter was markedly lower than that in summer, while the tympanic temperature did not differ between winter and summer. Walls and Jacobson [43] reported that environmental temperature had considerable influence on the skin temperature of the tail, in accordance with ST in the present study. Although the use of residual values of ST could mitigate the effect of season in the previous reports [17, 18], these values were not sufficient to mitigate the seasonal effect in the present study. Thermal stress has previously been reported to affect the behavior of cows [47–49], and maternal behaviors are no exception; Duncan and Meyer *et al*. [38] compared behavioral indices between their spring and fall calving experiments and observed an effect of seasons. In our present experiments, however, the effect of ambient temperature on behavior indices was limited to changes in the degree and timing of behavioral changes, implying that behavior indices are less sensitive to ambient temperature than ST.

Nevertheless, in both seasons the residual values of ST began to decrease approximately one day before parturition (Fig. 4A), and therefore the day of parturition could be predicted from ST, at least within the range of ambient temperature experienced in the present study. There were differences in the timing and degree of decrease in ST among seasons (Fig. 4A), and if a prepartum change in VT was true, the timing by which ST revealed the onset of calving in the cool season was less reliable than that in the warm season, because decreases in the residual values of ST and VT were consistent in the warm season but not the cool season. Although the present study did not investigate the accuracy and precision of calving prediction based on ST, some efforts will be needed for accurate calving prediction based on ST to set an appropriate threshold for change in ST depending on ambient temperature [18] or to control the effect of ambient temperature change in the maternity pen by herd management (e.g., installation of a heater in the maternity pen).

Conclusion

Maternal change in ST of beef cows was compared with those in VT and behavioral indices. A decrease in ST occurred approximately 30 h before parturition, which was similar to the time point of the decrease in VT and earlier than the increase in behavioral indices. In addition, a unique fluctuation of ST observed in the last few hours before parturition indicates that ST could provide a sign for parturition not only in the long-term like VT, but also in the short-term like behavioral indices. Although ST was more sensitive to ambient temperature than VT and the behavior indices, the day of parturition could be predicted from ST in both the warm and cool seasons.

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