Original Article

Efficiency of heat reduction in barns during rainy season and barn characteristics: their relationship to reproductive performance of dairy cows in a tropical environment

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Abstract

The objectives of this study were to evaluate the impact of efficiency of heat reduction on barns during the rainy season and barn characteristics on the reproductive performance of dairy cows in a tropical environment. Data of cows from 125 small-holder dairy farms in Chiang Mai, Thailand, during May to August 2014 was used. Reproductive performance indices, e.g. days to first insemination (DI) and days open (DO), were used as dependent variables. Microclimate factors included differences in temperature (TEMP_DIFF) and relative humidity (RH_DIFF) between the inside and outside of barns. From the total of 5,255 lactation data, 27 and 1,398 data was excluded due to missing data at the end of the follow-up for DI and DO models respectively. Results showed that both DI and DO were significantly related to calving season, roof materials and some farm management factors, but only DO were related to TEMP_DIFF. In conclusion, reproductive performance in the tropics is influenced by all factors related to climate. A decrease in pregnacy rate is more influenced by heat stress especially a decrease in temperature within barns than estrus expression.

Keywords: climate, microclimate, reproduction, dairy cow, tropical environment

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Introduction

Heat stress negatively affects animal welfare and productivity causing economic loss in the dairy industry globally (for review see Herbut *et al.*, 2018). The loss of most concern involves decreases in milk production and reproductive performance (West *et al.*, 2003; Herbut *et al.*, 2018). Impairment of reproductive performance in heat stressed cows includes negative effects of metabolic disorders, production of reproductive hormones, uterine environment (De Rensis and Scaramuzzi, 2003) and follicular development (Kanwichai *et al.*, 2019).

To monitor heat stress problems in dairy cows, the temperature-humidity index (THI), representing the combined effects of environmental temperature and humidity, was applied, where a THI value at >72 is considered as the value indicating a negative impact on reproductive performance (Armstrong, 1994; Morton *et al.*, 2007; Schüller *et al.*, 2014). Increases in THI cause more serious heat stress problems, especially for cows in tropical countries where the THI is always >72 or even >80 (Suriyasathaporn *et al.*, 2006; Moran and Chamberlain, 2017). Based on our literature review, most studies of heat stress effects on reproduction have been carried out in temperate countries where heat stress occurs during summer (Collier *et al.*, 2006). In a temperate climate, a wide range of temperatures occurs annually from -3 in the cold months to ≥10°C in the warmer months. In contrast, tropical climates are characterized by having an average monthly temperature of approximately 24±3°C; the term "winter" has no meaning in these areas and is often just called the "cool season". For example in Thailand, average temperatures are 23.4-27.0°C, 28.1-29.7°C, and 27.3-29.3°C for the cool, summer and rainy seasons, respectively. Thus climatic differences in temperate zones might not be relevant when seeking to understand reproductive impairment of cows in tropical climates.

Although heat stress is more serious in the tropics, reports of its effect on the reproductive performance of dairy cows are not consistent. Wide ranges of days open have been reported, e.g. 73±1 days in India (Kumar *et al.*, 2017), 151±15 days for cows in parity 1 and 2 in Taiwan (Liu *et al.*, 2018), and 253 days for crossbred dairy cows in Ethiopia (Lobago *et al.*, 2007). In the same tropical areas, heat stress conditions for cows may vary from one farm to another: cows are influenced not only by their climatic environment but also by generated microclimates within different locations and structures inside barns (Schüller and Heuwieser, 2016).

Numerous studies of microclimates have been conducted under controlled environments aiming at reducing the effects of heat accumulation on milk yield and fertility, for example by evaporative cooling achieved through a combination of sprinklers and fans (Her *et al.*, 1988; Flamenbaum *et al.*, 1995), but only a few have dealt with the contribution of barn design to the cow's response to heat stress especially for small holder dairy farms in tropical areas. In northern Thailand, the highest THI occurrs in the rainy season with the highest humidity and high temperatures causing the lowest reproductive performance in this area (Punyapornwithaya *et al.*, 2005). A barn that could decrease THI in the season would indicate its capacity to decrease THI in other seasons with less degree of heat stress, and thus help cows to improve their overall reproductive performance.

Therefore, the objectives of this study were to determine, for dairy cows in a tropical environment, the effects on reproductive performance of 1) calving season as a climate factor; 2) the capacities of barns to optimize their interior microclimate, indicated by the degree of barn temperature and humidity reduction during the rainy season; and 3) barn structure characteristics. Indicators were the days to first insemination, the intervals of calving to first insemination and the days open and intervals of calving to conception. In addition, farm management factors were evaluated as control factors relating to reproductive performance.

Materials and Methods

A retrospective cohort study was performed using 125 smallholder dairy farms in Chiang Mai Province, Thailand (18°47'25.37"N, 98°59'4.85"E). All farms had approximately 10-25 milking cows, mainly crossbred Holstein-Friesian. In most farms, the cows were fed roughage from agricultural by-products, such as corn trunks, corn husks and rice straw, supplemented with commercially formulated concentrates according to their milk production. These farms had been in the routine herd health management program of the Dairy Satellite Hospital, Faculty of Veterinary Medicine, Chiang Mai University, where the reproductive data of the cows was routinely updated. To evaluate the effects of current barn structure, cow reproductive data beyond the last dates of barn reconstruction was collected.

Independent variables: Independent variables comprised 4 factor types including 1) calving season, 2) the degree of barn temperature and relative humidity (RH) reduction, 3) barn characteristics and 4) farm management factors. Calving seasons were determined by calving dates - summer (March to May), rainy (June to October), and cool (November to February). To define the degree of barn temperature and RH reduction, referring to microclimate within farms, temperatures and RH on the inside and outside of farms were measured by temperature-humidity loggers at 11:00-14:00 during May to August 2014. The outside data was measured at the closed open field surrounding the barn and the inside data was measured at the center on both width and length axes of the barn. The degrees of barn temperature and RH reduction were defined by differences of temperature (TEMP_DIFF) and relative humidities (RH_DIFF) between the inside and outside of the barns.

Data on barn characteristics was collected including barn width (m), barn length (m), barn height (m), types of roof (1- and 2-level gable roof), roof materials (tile; corrugated galvanized iron), and barn orientation (north-south; east-west). Both the width and length of barns were measured based on the areas covered by the barn roofs and the barn heights were taken as the height of the distal ends of the roofs to the floors. Data on farm management included the type of housing (tiestall; freestall), farm size defined by the numbers of milking cows (<10, 10-25, and >25 milking cows), numbers of available freelance artificial insemination technicians (1-3; >3 AI technician/farm), and average cow body condition scores (Farm_BCS) to estimate farm nutritional management efficiency. During the farm visit, all cows within a specified farm were scored for their BCS (Edmonson *et al.*, 1989), and then the data was averaged.

Outcome variables: Outcome variables or reproductive performance variables in this study were the days to first service (DI) and the days open (DO). Based on averages of event-time or survival analysis for analysis of both outcome variables (Suriyasathaporn *et al.*, 1998), the outcome variables were defined as follows: referring to survival time, by the intervals from the starting point or calving date to the date of either of the specified successful events, namely first insemination for DI or having conception for DO; for so-called failed or censored cases, the end of follow-up as the last date of known status without an event. To minimize the inexplicable factors on both outcome variables, the maximal values of survival times of both DI and DO were the values of 75th percentile of overall data of either DI or DO. Therefore, cows having the first insemination or conception beyond their 75th percentiles value were defined as censored cases and having their survival times equal to those calculated values, respectively.

Statistical analysis: Data on climate and microclimate within the barn and their relationship were described by THI value using the formula THI=(1.8T+32)-([0.55-0.0055RH]×[1.8T-26]) (Schüller *et al.*, 2014) and scatter plots. Using the cutting values from the 75th percentile of DI and DO, univariable analyses for factors that were related to having first insemination and having conception within both cutting values days were performed. For continuous variables, Student's T tests were used to differentiate the means of those variables between cows with and without first insemination or conception within the specified days, respectively. For category variables, Fisher's Exact test was used to determine the association of independent variables of either having the first insemination or conception within the specified days, respectively.

The final Cox proportional hazard models (SAS Institute Inc., Cary, NC, USA) were used to determine independent variables that were associated with days to first insemination and days open. The aim of the analysis was to assess the hazard ratio (HR) of a particular value in comparison with a reference value (HR=1). Therefore, if the first insemination risk or conception risk at a particular value was >1, it meant that the first insemination rate or pregnancy rate of the particular value was higher than that of the reference value, or had shorter DI and DO days, respectively. The free-entering method using the maximum-likelihood test was used to assess the effects of each independent variable on either DI or DO. Only a variable with $P < 0.05$ could enter and remain in the final model. As for the categorical variables, all the values of the variable were entered at the same time.

Results

Data on climate and microclimate within barns is shown in Fig. 1. The THI values ranged from 80.6 (temperature=30°C, RH=64%) to 88.8 (temperature=38.2°C, RH=48%). Most farms (85%) had THI values from 83 to 87. Comparisons of temperatures and RH between the inside and the outside of barns are shown in Fig. 2. All temperatures measured outside were higher than the temperatures measured inside, ranging from 0.1 to 7.9°C. For RH, the outside values ranged from 32% to 69%, and the inside values from 25% to 67%. Most barns (93.6%) had inside RH higher than outside RH with ranges of RH_DIFF from -6 to 22%.

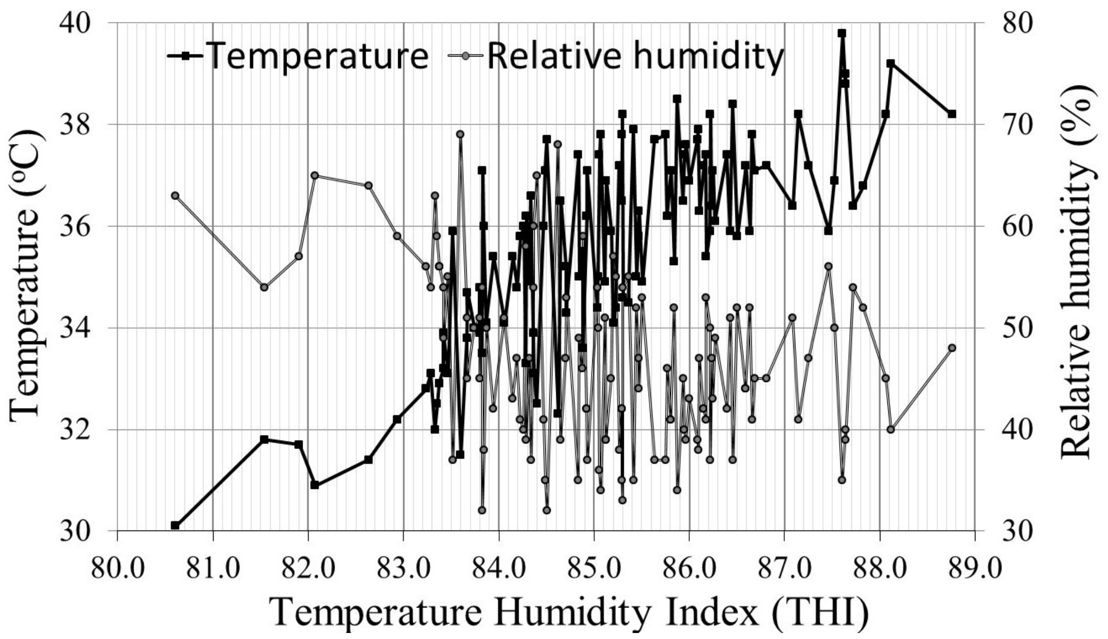

Figure 1 Scatter plots of temperature-humidity index (THI) against temperature and relative humidity inside the cow barns (n=125) measured during May 2014 in the time period from 11.00 to 14.00 in the Chiang Mai area

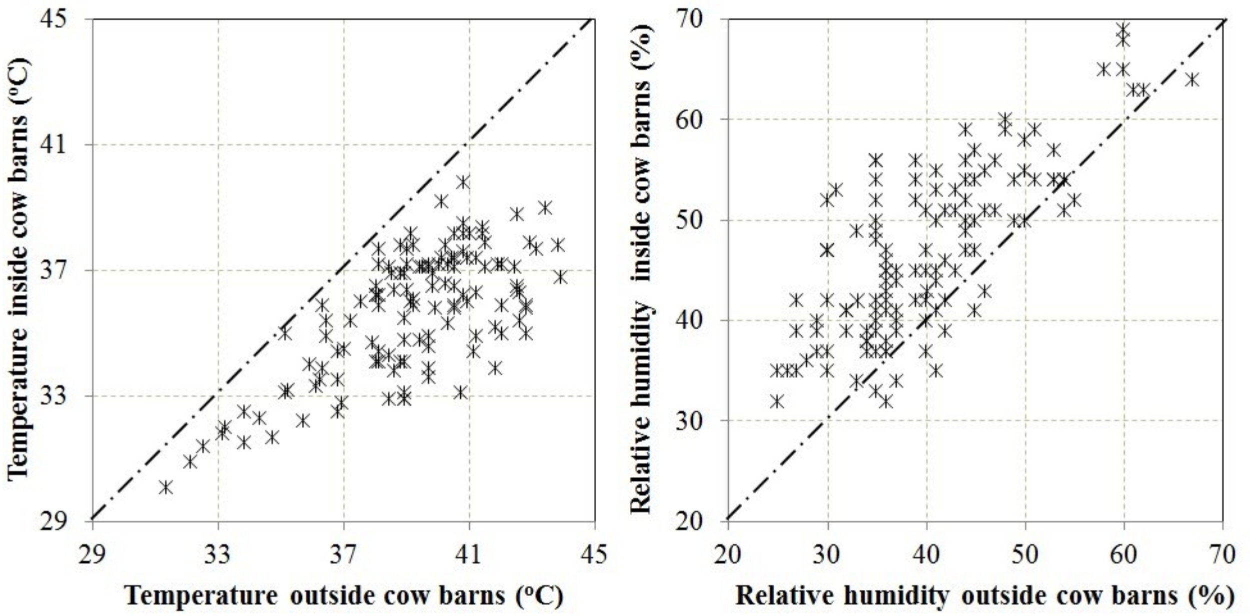

Figure 2 Relationships of environmental temperature (Left), and relative humidity (Right) between the inside and the outside of cow barns. The cross-lines indicate the points at which the inside value was equal to the outside value (n = 125)

From the total of 5,255 lactation data, 27 and 1,398 data were excluded due to missing end of follow-up data for DI and DO, respectively. The overall means of days to first insemination and days open were 97 d and 142 d, respectively. The 75th percentile of DI and DO were 130 d and 207 d, respectively. Consequently, the survival times of cows having days to first insemination >130 d or having days open >207 d were equal to 130 d and 207 d, respectively, and their lactations were then defined as censored cases.

Results on univariable analyses for the continuous variables on reproductive performance indices are shown in Table 1. The mean of barn width for cows inseminated within 130 days was higher than that for cows without insemination (P = 0.05). Barn height, TEMP_DIFF and RH_DIFF were significantly related to conception within 207 days postpartum. Results on univariable analyses for the categorical variables are shown in Table 2. Calving season, roof materials, types of housing, AI technician/farm and Farm_BCS were significantly related to having first insemination within 130 d. Factors including calving season, type of roof, type of housing, and AI technician/farm were related to conception within 207 days.

Factors relating to Days to First Insemination (DI): For DI, calving season, two barn characteristics factors (roof materials and barn orientation), and three farm management factors (type of housing, farm size and Farm_BCS) were in the final model (Table 3). Cows calving in the cool season were first inseminated postpartum later than cows calving in the summer (HR=0.826, $P < 0.01$). Cows in barns with tiled roofs (HR=1.197) had shorter DI than those in barns with corrugated galvanized steel roofing (P = 0.01), while barns oriented north-south (HR=0.92) had longer DI than barns oriented east-west (P = 0.02). Cows housed in freestalls had shorter DI than cows in tiestall housing (HR=1.3, $P < 0.01$). Cows in the biggest farm (>25 milking cows) had shorter days to first insemination. Cows in farms with Farm_BCS >2.75 had significantly longer DI than Farm_BCS 2.5-2.75 (HR=0.915, P = 0.05), but not for BCS_Farm <2.5.

Factors relating to Days Open (DO): The final model for DO included eight independent variables, as shown in Table 4. Cows calving in the cool season had significantly higher DO than those calving in summer (HR=0.828), but significantly lower than those calving in the rainy season (HR=1.103). The degree of barn temperature reduction or Temp_Diff were negatively related to DO ($P < 0.01$). Each °C of temperature reduction within the barn increased the risk to conception by about 4% (HR=1.04, $P < 0.01$). The barn characteristics in the final model for DO were barn length, type of roof and roof materials. Cows housed in longer barns, 2-level gable roofs and tiled roofs had significantly shorter DO. Cows kept in freestall housing had about 2 times (HR=2.234) greater conception risk compared to cows kept in tiestall housing ($P < 0.01$). In contrast to the result of the DI model, farms with Farm_BCS >2.75 had longer DO than Farm_BCS 2.5 to 2.75 (HR=1.183, $P < 0.01$) and Farm_BCS <2.5 (HR=1.139, P = 0.03). Farms using fewer inseminators (HR=1.805) had shorter DO than farms using more inseminators.

Table 1 Comparisons of barn structures and environments of cows with and without first artificial insemination (AI) within 130 d postpartum (pp), and cows with and without successful conception within 207 d postpartum (pp)

	AI within 130 d pp					Conception within 207 d pp				
	Yes (n=3234)		No (n=1165)		*P*-value	Yes (n=2506)		No (n=1913)		*P*-value
Variable	**Mean**	**SEM**	**Mean**	**SEM**		**Mean**	**SEM**	**Mean**	**SEM**	
TEMP_DIFF (°C)	3.58	0.03	3.60	0.05	0.17	3.66	0.04	3.49	0.04	< 0.01
RH_DIFF (%)	7.10	0.11	7.09	0.17	0.96	7.36	0.12	6.75	0.14	< 0.01
Barn width (m)	12.88	0.10	12.51	0.15	0.05	12.87	0.11	12.66	0.12	0.19
Barn length (m)	23.33	0.17	23.29	0.27	0.89	23.45	0.20	23.15	0.22	0.33
Barn height (m)	2.41	0.01	2.40	0.01	0.32	2.42	0.01	2.38	0.01	< 0.01

TEMP_DIFF and RH_DIFF: differences in temperatures and relative humidities between the inside and outside of barns, respectively.

Table 2 Comparisons of cow barn characteristics and management factors of cows with and without first artificial insemination (AI) within 130 d postpartum (pp), and cows with and without successful conception within 207 d postpartum (pp)

	Total (n)	AI within 130 d pp			Conception within 207 d pp		
Variable		**n**	**%**	***P*-value**	**n**	**%**	***P*-value**
Calving season				< 0.01			< 0.01
Summer	951	636	66.9		503	52.9	
Rainy	2038	1549	76.0		1258	61.7	
Cool	1430	1066	74.6		745	52.1	
Barn characteristics							
Type of roof				0.19			< 0.01
1-level gable roof	3724	2756	74.0		2075	55.7	
2-level gable roof	675	478	70.8		424	62.8	
Roof materials				< 0.01			0.47
Tiled	3893	2910	74.8		2221	57.1	
Corrugated galvanized iron	506	324	64.0		278	54.9	
Barn orientation				0.13			0.43
North-south	1851	1340	72.4		1037	56.0	
East-west	2568	1911	74.4		1469	57.2	
Farm management							
Type of housing				< 0.01			< 0.01
Tiestall	3270	2355	72.0		1770	54.1	
Freestall	1149	896	78.0		736	64.1	
Farm size (number of milking cows)				< 0.01			0.12
<10	619	414	66.9		336	54.3	
10-25	2645	1917	72.5		1488	56.3	
>25	1155	920	79.7		682	59.1	
Farm_BCS				0.05			0.91
BCS <2.5	1427	1031	72.3		810	56.8	
BCS 2.5-2.75	1821	1327	72.9		1038	57.0	
BCS >2.75	1171	893	76.3		658	56.2	
AI technician/farm				< 0.01			< 0.01
1-3 Inseminators	3236	2332	72.1		1767	54.6	
>3 Inseminators	1183	919	77.7		739	62.5	

Farm_BCS: averaged farm body condition score

Table 3 Results from Cox proportional hazard model for significant factors on calving season, barn characteristics and farm management in relationship to days first insemination (DI) of cows in a tropical environment (n=5,228 lactations)

Variable	Levels	β	SE (β)	HR	*P*-value
Calving season	Summer	-0.19	0.05	0.83	< 0.01
	Rainy	0.01	0.04	1.01	0.87
	Cool[1]	-	-	1.00	-
Barn characteristics					
Roof materials	Tiled	0.18	0.07	1.20	0.01
	Corrugated galvanized steel[1]	-	-	1.00	-
Barn orientation	North-south	-0.08	0.04	0.92	0.02
	East-west[1]	-	-	1.00	
Farm management factors					
Type of housing	Freestall	0.28	0.04	1.32	< 0.01
	Tiestall[1]	-	-	1.00	-
Farm size	<10 milking cows	-0.40	0.06	0.67	< 0.01
	10-25 milking cows	-0.23	0.04	0.79	< 0.01
	>25 milking cows[1]	-	-	1.00	-
Farm_BCS	BCS <2.5	-0.07	0.05	0.93	0.16
	BCS 2.5-2.75	-0.23	0.05	0.92	0.05
	BCS >2.75[1]	-	-	1.00	-

β: regression coefficient (parameter estimate), SE(β): standard error of regression coefficient, HR: hazard ratio
[1] Reference value (HR=1)

Table 4 Results from a Cox proportional hazard model for significant factors on calving season, degrees of barn temperature and humidity reduction, barn characteristics and farm management in relationship to days open (DO) of cows in a tropical environment (n=3,857 lactations)

Variable	Level	β	SE (β)	HR	*P*-value
Calving season	Summer	-0.19	0.06	0.83	< 0.01
	Rainy	0.10	0.05	1.10	0.04
	Cool[1]	-	-	1	-
Degree of barn temperature and humidity reduction					
TEMP_DIFF		0.04	0.01	1.04	< 0.01
Barn characteristics					
Barn length		0.005	0.002	1.01	0.05
Type of roof	1-level gable roof	-0.19	0.07	0.83	0.01
	2-levels gable roof[1]	-		1.00	
Roof materials	Tiled	0.20	0.07	1.22	< 0.01
	Corrugated galvanized steel[1]	-	-	1.00	-
Farm management					
Type of housing	Freestall	0.80	0.16	2.23	< 0.01
	Tiestall[1]	-	-	1.00	-
Farm_BCS	BCS <2.5	0.13	0.06	1.14	0.03
	BCS 2.5-2.75	0.17	0.06	1.18	< 0.01
	BCS >2.75[1]	-	-	1.00	-
AI technician/farm	1-3	0.48	0.23	1.81	< 0.01
	>3[1]	-	-	1.00	-

β: regression coefficient (parameter estimate), SE(β): standard error of regression coefficient, HR: hazard ratio
[1] Reference value (HR=1)

Discussion

Due to its inland nature and latitude, the northern part of Thailand experiences a long period of warm weather. During the hottest time of the year (March to May), temperatures usually reach 40°C (104°F). Starting in May, the southwest monsoon brings a stream of warm moist air causing abundant rain over the country. This climate causes different degrees of heat stress throughout the year. This study was conducted using 125 farms within an area of 20 km diameter and therefore, the general climatic effects on all farms would be similar. In this area, the THI values were clearly increased to above 72 after February (end of the cool season), presaging the heat stress environment exhibited during the summer and rainy seasons (Kanwichai *et al.*, 2019). The within farm THI measured in the rainy season in this study were also higher than 80 (Fig. 1).

Calving season: Calving seasons were associated with both days to first insemination and days open (Tables 3-4). In general, reproductive activity starts at 45 days after calving, and therefore activity will occur during the following season. Seasons are directly related to the climatic effects of this tropical area. In an earlier study in Thailand, dairy cows had the highest number of days open when calving in summer and the lowest number of days open when calving in the rainy season (Boonkum *et al.*, 2011). Cows calving in the rainy and cool seasons performed their best in estrus expression as shown by the shortest days to first insemination. In contrast, cows calving only in the rainy season had the best reproductive performance on estrus expression, fertilization and pregnancy, as the days open in this season were the shortest. This might indicate that heat stress had a more sensitive effect on either fertilization or pregnancy processes. Exposure of cows to elevated temperatures during oocyte maturation and ovulation (Putney *et al.*, 1989) caused impairment of estrus expression and the consequent insemination. In addition, for conception, heat stress before insemination has been associated with decreased fertility in cattle (AI-Katananiet *et al.*, 1999), impairment of follicular development (Roth *et al.*, 2001; Torres-Júnior *et al.*, 2008; Kanwichai *et al.*, 2019) and decreased embryonic viability and development (Jordan, 2003; Hansen, 2013) due to alteration in the uterine environment as well as growth and secretory activity of the conceptus (Geisert *et al.*, 1988).

Degree of barn temperature and humidity reduction: With the same climate in all the farms in this study, the differences in microclimate within the barns are shown in Fig. 2. The degree of barn temperature and humidity reduction in both variables, TEMP_DIFF and RH_DIFF, can be used to standardize the microclimate within barns for the whole year. Cows in barns with higher values TEMP_DIFF and RH_DIFF would be in a better microclimate, and therefore would have a lower degree of heat stress. It is surprising that the microclimate within barns affected only the days open (Table 1 and Table 4), not days to first insemination. This indicates that the severe heat stress might be related directly to fertilization, early embryo development and implantation process. The TEMP_DIFF, not RH_DIFF, was in the final model of days open. Exposing oocytes to 41°C during maturation increased the proportion of oocytes with fragmented DNA, the expression of apoptotic genes, ocurring in repeat breeder cows (Ferreira *et al.*, 2016). The reproductive tract, in particular the ovarian components (i.e., follicles, oocytes, CL), and preimplantation embryos are highly sensitive to elevated temperatures (for review see Wolfenson and Roth, 2019).

Barn characteristics: Microclimate in relation to heat stress, as a THI calculation, does not take into account the impact of wind speed or ventilated air movement (Kadzere *et al.,* 2002; West *et al*., 2003). Higher speeds of air movement result in the convection cooling of cows during heat waves (Davis and Mader, 2003). Barn characteristics are not only related to heat accumulation and heat dissipation causing changes of microclimate within barns, they are also related to times of sunlight and wind involving intervals of heat dissipation. The economic limitations of dairy farmers and the national policy for promoting small-holder dairy farms are the main influences on barn structure. Most dairy barns in Thailand including those studied were open barns without any heat dissipation tools. Shoshani and Hetzroni (2013) reported that barns oriented with a longitudinal axis perpendicular to the prevailing wind direction and hence barns with open ridge roofs could reduce the heat stress condition of cows. Solar radiation is one of the leading environmental factors that affect livestock (Herbut *et al*., 2018). East-west orientation can reduce the cows' exposure to solar radiation throughout the day compared with north-south orientation. In this respect, a previous study indicated that a tiled roof relieved heat stress significantly as, when measured, it reduced the respiration rates of dairy heifers and transmission of radiation was observed compared to galvanized steel roofs in a tropical environment (Conceição *et al*., 2008).

Farm management factors: Cows kept in the freestall housing system had both a higher first insemination risk and conception risk than cows kept in tiestall housing (Tables 3-4). As supported in previous studies, cows in freestall houses had shorter calving to the first insemination interval (Löf *et al*., 2007) and hence higher pregnancy rates than those in tiestalls (Löf *et al*., 2014). Only cows in big farms had shorter days to first insemination (Table 3). Larger herds had shorter days to first insemination due to higher numbers of heat detection (Löf *et al*., 2007). In this study, Farm_BCS, referring to the feed management efficiency of farms, indicated that farms with a higher value of Farm_BCS might have better feed management efficiency. The Farm_BCS were related to days to first insemination and days open in the different axes. Farms with better BCS (>2.75) had shorter days to first insemination (Table 3), but longer days open (Table 4). BCS is a score for estimating negative energy balance postpartum or NEB (Suriyasathaporn *et al*., 1998), and many studies have shown that BCS or NEB are related to increased days to first insemination and days open (Suriyasathaporn *et al*., 1998; Beam and Butler, 1999; López-Gatius *et al*., 2003; Hoedemaker *et al*., 2009).

It is not clear to us that the Farm_BCS were related to numbers of NEB cows within farms during reproductive activities. In addition, a combination of heat stress and NEB might have either synergistic or antagonistic effects on days open and their related physiology. Farms using fewer inseminators had fewer days open (Table 4) possibly due to farmers opting for good inseminators. According to Paul *et al*. (2011), the skills or experience of inseminators is considered to be an important risk factor for a successful first insemination pregnancy rate.

In conclusion, the reproductive performance of dairy cows in the tropics is influenced by heat stress caused by climate, microclimate within barns and barn characteristics, in which the barn characteristics are related to a modification of both climate and microclimate, for example speeds of air movement and sunrise times. The climate and barn characteristics influenced days to first insemination, referring to ovarian resumption postpartum, and days open, referring to fertilization and conception performance, with some differences in detail. However, the microclimate within barns affected only the days open, and this might indicate that severe heat stress causes further impairment of either fertilization or pregnancy precesses of cows in the topics.

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