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Effect of seasonal changes on fertility parameters of Holstein dairy cows in subtropical climate of Taiwan

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Objective: The purpose of this retrospective study was to investigate the relationship between temperature–humidity index (THI), season, and conception rate (CR) of Holstein cows in central Taiwan.

Methods: The mean performance and number of observations were statistically evaluated for various parameters, including age at first service, number of days open, gestation length, CR, and calving interval for different parities.

Results: The results indicate that the mean age at first service was 493.2 days; the gestation length was similar across all cows of different parities, ranging from 275.1 to 280.7 days. The overall CR of all inseminations was significantly lower in multiparous cows (47.26%±0.22%) than in heifers (57.14%±0.11%) (p<0.05). At THI>72 and during the hot season (from June to November), CRs for multiparous cows were significantly reduced compared to that for heifers, while the ratio remained unchanged among heifers for all seasons.

Conclusion: To achieve a high CR, lactating cows should be bred in winter and spring (from December to May) from the start of the seasonal breeding program, whereas the heifer should be allowed to breed in summer and fall under the subtropical climate in Taiwan.

Keywords: Calving Interval; Dairy Cow; Fertility; Temperature–humidity Index; Heat Stress

INTRODUCTION

The economic return from individual cows is represented by the integration of several performance traits besides milk production. Specific traits such as the age at first service, age at first calving (AFC), number of days open, gestation length, conception rate (CR; pregnancy per artificial insemination (AI) service), calving interval for different parities, and length of herd life are important factors contributing to economic return and are partially determined by management policies. Lactating dairy cows prefer ambient temperatures (ATs) between −5°C and 25°C, which is known as the thermoneutral zone. Above this temperature, cows can no longer adequately cool themselves and experience heat stress [1]. Milk production leads to metabolic heat production because of the metabolism of large amounts of nutrients; thus, cows with high milk productivity are more vulnerable to heat stress compared to those with lower yields [2]. Heat stress, which is caused by a combination of seasonal influences such as air temperature, relative humidity (RH), solar radiation, and wind speed, depresses milk production and reproduction performance in lactating cows [3]. Heat stress is the main environmental factor responsible for the lower fertility of cattle, especially during the summer season in many warm areas of the world [4]. This worldwide problem inflicts considerable economic losses and affects about 60% of the world's cattle population [5]. For the US dairy industry, annual economic losses caused by heat stress alone have been estimated to be about

\$900 million [6]. The climate of central Taiwan is defined as subtropical with concentrated rainfall during the hot season. Summers in Taiwan are extremely hot and humid, with temperatures and RH often exceeding 30°C (above 85°F) and 80% from June to November. Even though the environmental conditions suggest the possibility of the presence of heat stress, there has been no assessment of the influence of heat stress on the fertility of dairy cows in central Taiwan. The knowledge of these factors will make it easy to establish corrective measures to attain better fertility. In addition, AFC plays a critical role in achieving high profitability in dairy enterprises. Rearing replacement heifers needs large financial expenditure with no returns until the animals enter the milking herd [7]. An earlier AFC can save rearing costs due to decreased feed, labor fee, and rearing space. A previous study showed that reducing AFC from 25 months to 24 or 21 months could reduce rearing costs by 4.3% or 18%, respectively [7]. To maximize lactation performance and to reduce rearing costs, the mean AFC in Holstein heifers is recommended to be ≤24 months of age with a body weight of <560 kg [8]. A majority of dairy producers in the United Kingdom aim to start breeding Holstein Friesian heifers from around 14 to 15 months of age, and thus, the target AFC is generally 24 months on their farms [7]. No similar investigation has been conducted in central Taiwan. Therefore, the objective of the present study was to evaluate the effects of temperature, RH, and temperature-humidity index (THI) on the reproductive performance (AFC, number of days open, gestation length, CR, and calving interval) in Holstein cows of different parities. These results will help establish a management guide for dairy cattle in a subtropical region similar to central Taiwan.

MATERIALS AND METHODS

Animal housing and management

This study was conducted at the National Chung Hsing University (NCHU) Experimental Animal Farm in Taiwan (latitude and longitude is 24° 04' N and 120° 38' W, respectively). The herd consisted of Holstein heifers and cows, and the herd size was maintained at less than 110 animals. Animal health care was under the veterinary supervision. The animals were housed in groups in free-stall barns with slatted floors bedded with sand and equipped with overhead fans and a sprinkler system. Access to a shaded outdoor-exercise yard was also available for all lactating cows. Throughout the experimental period (from 2001 to 2013), cows were milked and fed twice daily with a diet of total mixed ration consisting of corn silage, sweet oats, chopped alfalfa, and Bermuda grass hay, as well as concentrate, minerals, and vitamins for lactating cows (Table 1). Fresh water was provided ad libitum. The mean milk yield for the herd was 8,700 kg for 305 days of lactation. The studied lactating cows (weighing 613 kg in average) were ranged from 2 year to 11

Table 1. Composition of concentrate mixtures fed to cows

Ingredients	Lactating cows (kg)	Dry cows (kg)
Corn	470.00	708.00
Soybean meal	150.00	185.00
Full-fat soy flour	80.00	0.00
Fish meal	50.00	0.00
Bran	120.00	50.00
Calcium carbonate	12.00	11.00
Dicalcium phosphate	4.00	10.00
Molasses	40.00	20.00
Salt	8.00	10.00
Saponification of fat powder	50.00	0.00
Sodium bicarbonate	13.00	0.00
Vitamins ¹⁾	1.00	2.00
Minerals ²⁾	2.00	4.00
Total	1,000.00	1,000.00

 $^{^{1)}}$ Vitamin premix formula: vitamin A, 16,000,000 IU; vitamin D₃, 6,000,000 IU; and vitamin E, 100,000 mg. The vitamin content (vitamins A, D, and E) was no lower than 1.5 times of the National Research Council (NRC [18]) recommended content.

year in age and 1 to 8 in parities. The reproductive assessments included CR, which was the ratio of the number of pregnancies per AI, and parity, which measured the number of births within the total fertility of cows over their reproductive life.

Pregnancy diagnosis

Pregnancy was confirmed using transrectal real-time ultrasonography 30 days after AI by the herd veterinarians. Pregnancy was verified by the presence of uterine fluid and an embryo with its heartbeat. The dates on which fertile insemination was performed as well as the number of inseminations per conception were recorded. Cows that were observed in estrus more than 10 days after AI were assumed as nonpregnant and reinseminated.

Meteorological data

Three different heat load indices are related to CR: mean THI, maximum THI, and minimum THI [5]. In this retrospective study, we aimed to investigate the relationship between THI and CR of Holstein cows in central Taiwan. In total, 13-year climatic records (from 2001 to 2013) were obtained from the official meteorological stations of the Taiwan Agricultural Research Institute, Wufeng, Taichung. The monthly and daily maximum temperatures and mean AT and RH were recorded throughout the year; the AT and RH data were used to calculate THI using the following equation: $THI = (1.8 \times AT + 32) - [(0.55 - 0.0055 \times RH) \times (1.8 \times AT - 26)]$ [5]. The official meteorological station is approximately 10-kilometer away from the NCHU Experimental Animal Farm. Average temperatures

²⁾ Mineral premix formula (per kg): ferrous sulfate, 60.00 g; manganese sulfate, 50.00 g; copper sulfate, 12.50 g; sodium selenite, 0.40 g; zinc sulfate, 50.00 g; and cobalt carbonate, 0.15 g. The mineral content (Co, Cu, Fe, Mn, S, Se, Na, and Mg) was no lower than 1.2 times of the NRC [18] recommended content.

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and humidity are similar for the two locations. However, the effects of the artificial cooling system in the barn were not considered in the study.

Statistical analyses

The date, age, parity, breed of semen-donor bull, season, date of estrus, names of AI technicians, and the time and date of AI were recorded. Raw data were collated using Microsoft Excel (Microsoft Corp., Redmond, WA, USA) and were subsequently transferred to SPSS analytical software (version 11.5; SPSS, Inc., Chicago, IL, USA) for multiple logistic regressions, whereas continuous variables (calving to first service interval, calving to conception interval, THI, CR, and different parities) were evaluated using the analysis of variance (PROC general linear model from SAS). The significance of differences for continuous variables was tested using Tukey's test (least square means statement). Results were considered to be significant at p<0.05.

RESULTS

The effects of climate changes on fertility

The dataset contained 1,321 breeding records and associated pregnancy diagnoses from 607 dairy cows. The overall CR obtained was 48.5%. Minimum, maximum, and mean ATs during the study period were 12.2°C, 32.8°C, and 24.13°C, respectively, and mean RH during the study period was 80.08% (Table 2). Minimum, maximum, and mean THIs during the study period were 61.30, 80.39, and 72.19, respectively. THI showed seasonal fluctuations with lower monthly means (from 66.44 to 70.90) during cold climate conditions (from December to May) and higher monthly means (from 74.93 to 79.75) during hot climate conditions (from June to November) as shown in Table 3. Temporal trends of the evaluated THI are shown in Figure 1. THI was significantly affected by the season, with a monthly variation ranging between 60 and 80, although it did not vary significantly over the years. Table 3 shows that CRs were significantly affected by the seasons, particularly in dairy cows. Hot seasons (from June to November) significantly reduced CRs for multiparous cows (p<0.05), while CRs in heifers remained unchanged in all seasons. As shown in Table 4, at THI of >72, CR for multiparous cows was significantly

Table 2. Monthly temperatures (mean, max, and min, °C), mean relative humidity (RH %), and mean temperature—humidity index (THI %) during the study period (from 2001 to 2013) in central Taiwan

Months	Ter	nperature (Mean RH	Mean THI		
MOULTIS	Mean	Max	Min	%	%	
Jan.	17.50	22.79	12.21	78.24	61.30	
Feb.	18.97	23.83	14.10	78.99	63.77	
Mar.	20.53	25.06	16.00	77.82	63.56	
Apr.	24.02	28.33	19.70	80.67	72.38	
May	26.60	30.75	22.45	81.44	76.75	
Jun.	28.07	31.96	24.18	82.37	78.96	
Jul.	28.92	32.45	25.38	80.20	79.91	
Aug.	28.89	32.81	24.97	82.08	80.39	
Sep.	28.16	32.20	24.11	83.23	79.22	
Oct.	25.81	30.50	21.11	79.20	74.77	
Nov.	22.62	27.15	18.08	79.27	70.80	
Dec.	19.46	24.80	14.11	77.50	64.45	

reduced compared to that for heifers (p<0.05). CRs for multiparous cows were 43.84% and 50.68% at THIs of >72 and \leq 72, respectively, over the 13-year study. For a THI<72, CR was high throughout the year, with no significant differences between the months or seasons.

Effect of different parities on conception rate, days open and gestation length

The reproductive efficiency of the herd is important to the economic success of the dairy operation. Table 5 shows that the number of required AI services differed across cows of different parities, where cows in parities 6 and 7 have higher CRs than those in parities 2, 3, 4, 5, and 8 (p<0.05). The overall CR for all inseminations was significantly lower in multiparous cows (47.26%±0.22%) than that in heifers (57.14%±0.11%) (p<0.05). Table 6 shows the time elapsed for getting pregnant (days open) of Holstein cows with different parities. A larger number of days open indicate a decline in fertility of cows. At the NCHU Experimental Animal Farm. Significant differences in the number of days open were found across between parity 7 to 8 cows and parity 1 to 2, parity 3 to 4 and parity 5 to 6, parity 6 to 7 cows. Table 7 shows that the length of the gestation periods varied from 252 days to 304 days across all cows of different parities. Most of the calves were born between the

Table 3. Temperature, mean temperature—humidity index (THI), and seasons associated with conception rates (CR) of heifers and multiparous cows

Season	Moon tomporature (°C)	Mean THI	Numbe	r of animals (N)	CR (%)		
	Mean temperature (°C)	Weall Ini	Heifers	Multiparous cows	Heifers	Multiparous cows	
Spring (from Mar. to May)	23.72	70.90	30	95	60.73ª	51.25°	
Summer (from Jun. to Aug.)	28.63	79.75	36	98	56.25°	42.25 ^b	
Fall (from Sep. to Nov.)	25.53	74.93	25	118	54.26°	43.13 ^b	
Winter (from Dec. to Feb.)	18.98	66.44	14	126	57.41 ^a	50.66°	

CR: conception rate (pregnancy per artificial insemination service).

 $^{^{}a,b}$ Means with different superscripts in the same row differ at p < 0.05.



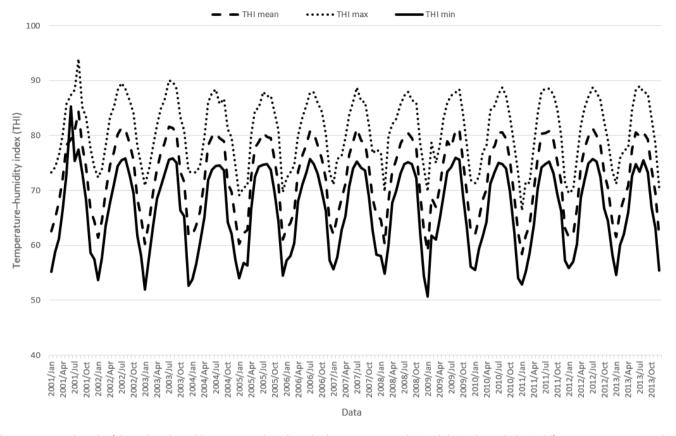


Figure 1. Temporal trends of the evaluated monthly temperature—humidity index (THI mean, max, and min %) during the studied period (from 2001 Jan to 2013 Oct) in central Taiwan.

275th and 280th days of gestation, inclusively. No significant difference regarding the gestation lengths was found across all cows of different parities.

DISCUSSION

In Quebec, the AFC occurs, on an average, at 27 months, whereas the target is between 23 months and 24.5 months to

Table 4. Mean temperature—humidity index (THI) thresholds associated with conception rates (CR) of heifers and multiparous cows

	Number	of animals (N)	CR (%)				
THI threshold	Heifers	Multiparous cows	Heifers	Multiparous cows			
Mean THI < 60	8	57	61.26	52.21			
Mean THI 60-65	20	84	58.41	51.15			
Mean THI 66-70	31	79	56.15	49.13			
Mean THI 71-75	21	82	57.72	46.82			
Mean THI 76-80	13	75	54.42	42.26			
Mean THI > 80	12	60	52.61	40.41			
Mean THI ≤72	65	225	59.14	50.68			
Mean THI > 72	40	212	55.12 ^b	43.84ª			

CR: conception rate (pregnancy per artificial insemination service).

maximize herd profitability [9]. The data show that the mean age at the first service and AFC in Holstein heifers at the NCHU Experimental Animal Farm are 493.2 days and 813.5 days (27.1 months), which are similar to that in the United Kingdom and Quebec, respectively [7,9] (Table 7). The normal gestation period for cows is between 275 days and 280 days [10]. At the NCHU Experimental Animal Farm, the mean gestation lengths based on gestation data were 275.1 days and 277.9 days for Holstein heifers and cows, respectively. Furthermore, the gestation lengths were similar across all cows of different parities. Three cows at this farm went into early labor (parity 1, on the 252nd day, parity 2 on the 262th day, and parity 3 on the 262th day), which may have been due to heat stress or illness, judging by the observation records. CR is commonly used as a critical indicator to evaluate the reproductive efficiency of a dairy farm. Although high CRs were observed on cows in parity 6 and 7 with unknown reason, probably due to too few sample size of cows in the records, our results suggest that in general multiparous cows have lower CRs than heifers (Table 5). Multiparous cows have been reported to have lower reproductive abilities, in which their embryos may die in the early stages of pregnancy, and the implantation of embryos become more difficult after multiple pregnancies [11,12]. In addition, lower values of days open were found on cows in parity 6 and 7,

 $^{^{}a,b}$ Means with different superscripts in the same row differ at p < 0.05.



Table 5. Statistics of artificial insemination in Holstein cows with different parities

Variables Parity P (n) N	ity 1	Par	ity 2	Par	ity 3	Par	ity 4	Par	ity 5	Pari	ty 6	Par	ity 7	Par	ity 8	
	P (n)	N (n)	P (n)	N (n)	P (n)	N (n)	P (n)	N (n)	P (n)	N (n)	P (n)	N (n)	P (n)	N (n)	P (n)	N (n)
2001	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
2002	1	0	0	0	1	1	0	0	0	0	0	0	0	0	0	0
2003	0	0	1	2	1	0	1	1	0	0	0	0	0	0	0	0
2004	2	2	0	0	1	2	1	4	1	0	0	0	0	0	0	0
2005	5	1	3	2	0	0	1	1	1	1	1	0	0	0	0	0
2006	5	5	5	4	1	2	0	0	0	2	1	1	0	0	0	0
2007	6	8	4	4	6	4	0	0	1	0	2	1	1	0	0	0
2008	11	5	5	9	3	5	3	9	0	0	1	0	0	1	0	2
2009	18	11	8	18	4	15	4	17	3	4	0	0	2	2	1	1
2010	12	5	9	9	6	9	5	5	2	9	2	0	0	0	1	2
2011	15	10	8	8	12	16	4	11	4	7	1	2	3	0	0	3
2012	14	11	8	8	7	7	9	9	2	5	1	2	1	2	1	0
2013	15	7	6	8	3	5	6	11	4	4	0	0	0	2	1	3
Number of cows with pregnancy	1	05	!	58	۷	! 5	3	34	1	8		9		7	•	4
Number of services per conception		±0.11ª	2.31 ±	± 0.22 ^{a,b}	2.53	± 0.24 ^b	2.38 ±	0.13 ^{a,b}	2.39 ±	0.42 ^{a,b}	1.56 ±	± 0.29 ^a	1.57 =	±0.30 ^a	2.75	± 0.63 ^b
CR (%)	57.	14 ^{a,b}	43	.29ª	39	.53ª	40	.02ª	41	.84ª	64.	.10 ^b	64	.10 ^b	36	.36ª

P, pregnancy; N, non-pregnancy; n, number of cows; CR, conception rate (pregnancy per artificial insemination service).

which supported to the results of CRs in the same parities (Table 6). A higher value of days open and the increased culling rate typically decreased the profitability of dairy herds. The studied farm had 100 pregnant heifers; 30 of them were sold and 70 were studied. The culling rate of cows was 27.14% for parity 1 to 2, 15.69% for parity 2 to 3, 34.88% for parity 3 to 4, 50% for parity 4 to 5, 42.86% for parity 5 to 6, 25% for parity 6 to 7, and 50% for parity 7 to 8. Among these cows, parity 4 to 5 and 7 to 8 had the highest culling rates. After the fourth delivery, cows were culled due to reproductive disorders (37.66%) occurred during lactation or pregnancy or due to

illnesses such as mastitis (18.18%), or other health problems including lameness, milk fever, abomasum dislocation, bovine ephemeral fever, tuberculosis, or death (44.16%). Our results suggest that the reproduction-related parameters should be weighted in the culling criteria for multiparous cows above 4 parties in Taiwan.

THI is used for assessing thermal stress, and high-producing dairy cows are adversely affected when THI exceeds 72 [13,14]. In the present study, THI was significantly affected by the season, with a monthly variation ranging between 60 and 80. Taiwan has an island climate, with the sea breeze blow-

Table 6. Variations in the number of days open of Holstein cows with different parities

Variables	Parity 1 to 2	Parity 2 to 3	Parity 3 to 4	Parity 4 to 5	Parity 5 to 6	Parity 6 to 7	Parity 7 to 8
Number of cows	51	43	27	14	8	6	3
Days open (min, max)	151 ± 15 ^a (52, 604)	182 ± 17 ^{a,b} (39, 498)	156 ± 28^{a} (49, 665)	178 ± 40 ^{a,b} (53, 558)	157 ± 47^{a} (49, 414)	158 ± 32^{a} (59, 302)	219±31 ^b (165, 273)

 $^{^{}a,b}$ Means with different superscripts in the same row differ at p < 0.05.

Table 7. Gestation length of Holstein cows with different parities

Variables	Parity 1 ¹⁾	Parity 2	Parity 3	Parity 4	Parity 5	Parity 6	Parity 7	Parity 8
Number of cows	70	51	43	28	14	8	6	3
Gestation length								
Mean (d)	275.1 ± 6.5	277.8 ± 6.28	276.8 ± 5.4	279.1 ± 6.4	275.4 ± 6.2	277.4 ± 4.0	278.2 ± 5.0	280.7 ± 2.9
Min (d)	252	263	260	270	262	273	271	279
Max (d)	294	297	289	304	283	285	286	284

¹⁾ The mean age at the first service and age at first calving in Holstein heifers are 493.2 days and 813.5 days (27.1 months), respectively.

 $^{^{}a,b}$ Means with different superscripts in the same row differ at p < 0.05.

ing inland in the evening during summer; this carries away the heat built up in the animal housing, and thus reduces the extent of cows exposed to heat stress in the evening [15]. Therefore, while CR appeared to be lower in cows under heat stress than in those not under heat stress, it did not reach a significant difference. Although CR showed no significant difference between both THI>72 and THI≤72 ambient conditions in heifers or multiparous cows, there is a significant difference between heifers and multiparous cows when THI>72. Ferreira et al [11] suggested that cow oocytes are more sensitive to heat stress during the hot season. Heifers and cows showed a similar number of recovered and viable oocytes during winter, but during summer the cow oocyte numbers significantly decreased [12,16]. In the present study, lactating cows have lower CR during summer and fall than that during winter and spring. However, during spring, summer, fall, and winter, CRs were similar in heifers. Because heat stress during summer and fall resulted in a decreased CR of multiparous cows in the current study, lactating cows are suggested to be bred in winter and spring from the start of the seasonal breeding program, whereas the heifer can be bred in all seasons under the subtropical climate in Taiwan.

In Taiwan, the heat stress is a potential threat that induces impaired reproductive performance such as syndromes of puerperal metritis (PM) and retained placenta (RP) in dairy cows [17]. Additional reproductive strategies are needed to counteract the adverse effect of heat stress on fertility during summer and fall. To improve the reproductive efficiency of dairy cows with PM/RP, we previously reported an effective, two-step treatment strategy using combined medical manipulation for regulating uterine involution and ovarian function in such cows, which enhanced reproductive efficiency via controlling uterine infections well and, thereby, facilitating an early resumption of ovarian activity of dairy herds [17]. In addition, from the beginning of the summer till the end of fall, keeping lactating cows cool can provide a good return on investment as it makes cows more comfortable, thereby, making them more productive in subtropical climates. Because the price of raw milk is relatively higher in summer than that in winter, regardless of the effect of heat stress, many dairy farmers in Taiwan prefer to breed cows in the early summer season in order to obtain higher milk yields in hot seasons of the next year. The suggested breeding strategy in the study allows farmers to use heifers to maintain a higher production of raw milk in summer.

IMPLICATIONS

In conclusion, our data shows that at THI of >72 and during the hot season (from June to November), CRs were significantly higher in heifers than in multiparous cows, although no significant differences were observed in the number of days open or gestation length on a dairy farm in a subtropical region in Taiwan. The contributions of physiological condition and genetic background will need to be included for further characterization of the relationship between stress response and reproductive performance in subtropical regions in Taiwan.

CONFLICT OF INTEREST

We certify that there is no conflict of interest with any financial organization regarding the material discussed in the manuscript.

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