Effects of seasonal change and parity on raw milk composition and related indices in Chinese Holstein cows in northern China

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ABSTRACT

This study was to investigate the effects of seasonal change and parity on milk composition and related indices, and to analyze the relationships among milk indices in Chinese Holstein cows from an intensive dairy farm in northern China. The 6,520 sets of complete Dairy Herd Improvement data were obtained and grouped by natural month and parity. The data included daily milk yield (DMY), milk solids percentage (MSP), milk fat percentage (MFP), milk protein percentage (MPP), milk lactose percentage (MLP), somatic cell count (SCC), somatic cell score (SCS), milk production loss (MPL), and fat-to-protein ratio (FPR). Data analysis showed that the above 9 indices were affected by both seasonal change and parity. However, the interaction between parity and seasonal change showed effects on MLP, SCS, MPL, and DMY, but no effects on MFP, MPP, MSP, and FPR. Duncan's multiple comparison on seasonal change showed that DMY (23.58 kg/d), MSP (12.35%), MPP (3.02%), and MFP (3.81%) were the lowest in June, but SCC ($288.7 \times 10^3/\text{mL}$) and MPL (0.69 kg/d) were the lowest in January; FPR (1.32) was the highest in February. Meanwhile, Duncan's multiple comparison on parities showed that MSP, MPP, and MLP were reduced rapidly in the fourth lactation, but SCC and MPL increased with increasing parities. The canonical correlation analysis for indices showed that SCS had high positive correlation with MPL (0.8360). Therefore, a few models were developed to quantify the effects of seasonal change and parity on raw milk composition using the Wood model. The changing patterns of milk composition and related indices in different months and parities could provide scientific evidence for improving feeding management and nutritional supplementation of Chinese Holstein cows.

Key words: milk composition, seasonal change, parity, model

INTRODUCTION

The proportion of major components in raw milk not only reflects the dairy quality and health status of dairy cows (Auldist et al., 1996), but also affects the nutritional value of milk and dairy products (Kefford et al., 1995). Therefore, it is necessary to investigate the factors that affect raw milk composition, which may improve the quality and yield of raw milk through scientific management of feeding and nutritional supplementation.

Factors that influence milk composition include 2 aspects: external factors (e.g., season, feeding system, and milking frequency) and internal factors (e.g., gene, parity, and stage of lactation; Fox and McSweeney, 1998; Cao et al., 2010). For external factors, higher percentages of milk fat and protein are found in the evening milking (Quist et al., 2008), but the season variation has more significant effect on the milk composition than the milking time. Among internal factors, genes have a major effect on milk composition (Khatib et al., 2007, 2008), but it is difficult to change milk production through gene manipulation. Some studies have focused on the effect of season variation and stage of lactation on milk composition (Ray et al., 1992; Auldist et al., 1998). As an internal factor, parity also has correlated with milk composition (Fukasawa et al., 2008; Cao et al., 2010). Therefore, season and parity might be considered as important factors for effectively analyzing the variation in raw milk composition.

Some studies have shown that seasonal variation affects milk composition through several aspects, such as ingestible diets, photoperiod, and temperature (Allore et al., 1997; Nudda et al., 2005; Heck et al., 2009). With diet change, milk composition is changed markedly (Elgersma et al., 2004). Furthermore, significant differences exist in the feeding management and climate in different countries (Lindmark-Månsson et al., 2003). On the other hand, few studies have analyzed the correlations among major components and other indices in raw milk, such as milk yield, SCC and milk losses, which are directly linked to the health status of cows. Furthermore, high SCC might reduce milk qual-

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Table 1. The formulation of TMR (based on DM)

Composition, %	Lactation stage		
	Early lactation	Mid lactation	Late lactation
Corn silage	23	28	34
Alfalfa hay	10	12	14
Chinese wild rye grass hay	12	15	19
Distillers dried grains (DDG)	5	5	3
Special concentrate supplement	50	40	30

ity and flavor (Ma et al., 2000), and high-SCC milk has poor coagulating properties, which could lead to the potential reduction in the yield of cheese (Auld et al., 1998). Therefore, the correlations between milk composition and seasonal changes in China are worth exploring.

The objectives of this study were to investigate effects of seasonal change and parity on the raw milk composition and analyze the correlations among different milk indices. A few models were also developed to evaluate the effects of seasonal variation on milk composition. The change in patterns of milk composition and related indices in different months and parities could provide scientific evidence for improving feeding management and nutritional supplementation for Chinese Holstein cows.

MATERIALS AND METHODS

Diets and Milking of Lactating Cows

In this study, Chinese Holstein cows enrolled in the DHI program were from an intensive dairy farm in the north of China (Tianjin city) in 2009. The diet was mixed and fed as TMR. The formulation of TMR is shown in Table 1. The same formulation of special concentrate supplement contained corn (52%), wheat bran (8%), wheat middling and red dog (10%), soybean meal (13%), cottonseed meal (5%), rapeseed meal (5%), bone meal (1%), feed-grade salt (1%), and premix (5%). The ratio of concentrate to forage was 5:5, 4:6, and 3:7, respectively, for early lactation, mid lactation, and late lactation based on DM content. Chinese Holstein cows were housed in cubicle sheds; given unrestricted access to drinking water; milked 3 times daily at 0600, 1200, and 2000 h by milking machine; and fed more than 45 kg of TMR for at least 20 h per day.

Collection and Preprocessing of DHI Data

The DHI data of lactating cows were collected and grouped by parity and natural month. Records with missing values were removed from DHI original data

(Table 2). Cows with parity greater than 4 (562 sets of DHI data) were deleted from 9,727 sets of DHI original data, because the number of cows (parity >4) declined obviously, which could not represent the effect of seasonal change and parity on the milk composition. Data with SCC greater than 2 million cells per milliliter were also excluded (Ministry of Health P. R. China, 2010). Finally, the 6,520 sets of complete DHI data were obtained, which included 8 indices: daily milk yield (**DMY**), milk solids percentage (**MSP**), milk fat percentage (**MFP**), milk protein percentage (**MPP**), milk lactose percentage (**MLP**), fat-to-protein ratio (**FPR**), SCC, SCS, and milk production loss (**MPL**). The number of cows in different parities and natural months was different; for example, the largest number of samples was 279 (parity 1 in March) and the least number of samples was 45 (parity 4 in July). The parity distribution of cows in the study was as follows: parity 1 ($n = 280$), 43.0%; parity 2 ($n = 160$), 24.7%; parity 3 ($n = 140$), 21.5%; and parity 4 ($n = 70$), 10.8%.

Data Analysis

Considering the imbalance of observation data, PROC GLM of SAS (version 8.01; SAS Institute, 1999) was used. Effects of parity and natural month on the above-mentioned 9 indices were analyzed using ANOVA. A level of $P < 0.05$ was set as the criterion for statistical significance. All data were then analyzed using PROC CANCORR (SAS Institute, 1999) for the canonical correlation analysis among 9 indices. To quantify the relationships between major indices and natural months, nonlinear regression models were constructed using PROC NLIN (SAS Institute, 1999) and the Wood model (Wood, 1967).

RESULTS AND DISCUSSION

Effects of Seasonal Change on the 9 Indices

The results of ANOVA in Table 3 showed that both seasonal change and lactation parity significantly affected 9 indices ($P < 0.05$). However, the interaction

Table 2. Description of collected DHI data

Item ¹	Mean	SD	Minimum	Maximum
Parity	1.99	1.03	1.00	4.00
DMY, kg/d	27.51	9.60	0.10	57.30
MFP, %	3.91	0.41	1.34	5.10
MPP, %	3.10	0.28	1.46	4.52
MLP, %	4.87	0.23	2.30	5.43
MSP, %	12.58	0.62	6.67	14.57
FPR	1.27	0.15	0.58	2.03
SCC, $\times 10^4$ /mL	31.40	32.27	4.00	199.40
SCS	4.06	1.29	1.68	7.32
MPL, kg/d	0.84	1.12	0.00	7.40

¹DMY = daily milk yield; MFP = milk fat percentage; MPP = milk protein percentage; MLP = milk lactose percentage; MSP = milk solids percentage; FPR = fat-to-protein ratio; MPL = milk production loss.

between parity and natural month showed significant effect on DMY, MLP, SCS, and MPL ($P < 0.05$), but other indices were not affected.

The study also found that seasonal change affected milk composition and related indices. The effect of seasonal change on SCC was significant ($P < 0.05$), but its effects on other 8 indices were highly significant ($P < 0.01$; Table 3). Duncan's multiple comparisons of seasonal change indicated that DMY (23.58 kg/d), MFP (3.81%), MPP (3.02%), and MSP (12.35%) were the lowest in June (Table 4). Although the main milk components in some studies were different from each other, similar results have been found (Allore et al., 1997; Auldist et al., 1998; Heck et al., 2009). Those authors explained that the change of main milk components in different seasons is caused by different ingestible diets. For example, pasture was a major component of the diet fed to cows in the summer, whereas in the winter, pasture supplemented with silage was used. The nutritional fluctuations of diet during different seasons are related to changing availability and quality of pasture. However, the TMR is mainly constituted of silage, mixed hay (alfalfa hay and Chinese wild rye grass hay), and special concentrate supplement for the whole year round in China. Total mixed ration with different proportions of the same feedstuffs could meet the nutrient requirements of different lactation stages of cows, so

the change in milk composition in different seasons was not due to different ingestible diets. Previous studies showed that increasing air temperature, temperature-humidity index, and rectal temperature above critical thresholds result in a decrease in DMI, milk yield, and efficiency of milk production (West, 2003), and significantly affect the milk composition (Bernabucci and Calamari, 1998). Climate change can affect the neuroendocrine system in animals and then influence hormonal equilibrium, energy balance, water balance, and body temperature, and eventually affect growth, reproduction, milk production, and the immune system in animals (Cappa, 1998). However, further investigations are needed to explain the change in milk composition in different seasons.

The SCC was the highest (385.2×10^3 /mL) in August, but decreased to the lowest level (288.7×10^3 /mL) in January, which is consistent with previous studies (Quist et al., 2008). The reason is attributed to heat stress (Hammami et al., 2013). A sprinkler and fan cooling system installed at a feed bunk could effectively reduce heat stress in dairy cows (West, 2003), which was given as a recommendation of how to reduce SCC in raw milk, to achieve an improvement in milk sanitation.

It is interesting that the highest value of SCS was in November and not in August, which is different from

Table 3. The ANOVA of 9 indices in different months and parities

Item	Month	Parity	Month $\times$ parity
Daily milk production (DMY)	***	***	***
Milk fat percentage (MFP)	***	**	†
Milk protein percentage (MPP)	***	**	†
Milk lactose percentage (MLP)	***	***	*
Milk solid percentage (MSP)	***	*	†
Fat-to-protein ratio (FPR)	***	*	†
Somatic cell count (SCC)	*	***	†
Somatic cell score (SCS)	***	***	*
Milk production loss (MPL)	***	***	*

*** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$; † $P > 0.05$.

Table 4. Duncan's multiple comparisons of 9 indices in 12 mo¹

Month	DMY, kg/d	MFP, %	MPP, %	MLP, %	MSP, %	FPR	SCC, $\times 10^4$ /mL	SCS	MPL, kg/d
1	28.46 ^{cd}	4.01 ^b	3.11 ^c	4.99 ^a	12.82 ^b	1.29 ^b	28.87 ^c	3.82 ^f	0.69 ^f
2	27.88 ^{cde}	4.11 ^a	3.14 ^{bc}	4.98 ^a	12.94 ^a	1.32 ^a	29.79 ^c	3.91 ^{ef}	0.75 ^{de}
3	26.73 ^{ef}	3.94 ^c	3.05 ^{de}	4.84 ^d	12.54 ^c	1.30 ^{ab}	30.06 ^c	3.87 ^{ef}	0.72 ^{de}
4	25.45 ^g	3.94 ^c	3.05 ^{de}	4.85 ^d	12.55 ^c	1.30 ^{ab}	31.33 ^{bc}	3.90 ^{ef}	0.75 ^{de}
5	26.02 ^{fg}	3.82 ^{ef}	3.06 ^d	4.87 ^d	12.44 ^d	1.26 ^{cd}	30.45 ^c	3.85 ^{ef}	0.70 ^e
6	23.59 ^h	3.81 ^f	3.02 ^e	4.80 ^e	12.35 ^e	1.27 ^c	29.62 ^c	3.88 ^{ef}	0.74 ^{de}
7	26.24 ^{fg}	3.83 ^{ef}	3.03 ^{de}	4.89 ^c	12.45 ^d	1.27 ^c	32.88 ^{bc}	4.00 ^{de}	0.86 ^{dc}
8	27.49 ^{de}	3.86 ^{def}	3.04 ^{de}	4.94 ^b	12.54 ^c	1.27 ^c	38.53 ^a	4.21 ^c	0.99 ^{abc}
9	27.20 ^{ef}	3.89 ^d	3.20 ^a	4.79 ^e	12.59 ^c	1.22 ^f	33.94 ^{bc}	4.16 ^{cd}	0.95 ^{bc}
10	28.76 ^c	3.87 ^{de}	3.14 ^{bc}	4.90 ^c	12.61 ^c	1.24 ^{de}	30.82 ^{bc}	4.10 ^{cd}	0.87 ^{cd}
11	30.09 ^b	3.88 ^{de}	3.12 ^c	4.75 ^f	12.45 ^d	1.24 ^{de}	35.59 ^{ab}	4.61 ^a	1.12 ^a
12	31.99 ^a	3.87 ^{de}	3.17 ^b	4.81 ^e	12.56 ^c	1.23 ^{ef}	32.74 ^{bc}	4.46 ^b	1.06 ^{ab}

^{a-h}Values within a column with different superscripts are significantly different ($P < 0.05$).

¹DMY = daily milk yield; MFP = milk fat percentage; MPP = milk protein percentage; MLP = milk lactose percentage; MSP = milk solids percentage; FPR = fat-to-protein ratio; MPL = milk production loss.

the pattern of SCC. The original DHI data showed that the proportion of SCC less than 200,000 cells/mL in August was higher than that in November (42.41 vs. 17.80%), but the proportion of SCC between 200,000 and 500,000 cells/mL in August was lower than that in November (36.30 vs. 64.22%). The increasing proportion of SCC between 200,000 and 500,000 cells/mL in November might be a statistical reason for the peak of SCS in November (Olde Riekerink et al., 2007). The high level of SCS from November to December might be related to the increased DMY, because a higher milk yield may increase the risk of infection, acting as a stress factor, thereby increasing the SCS (de los Campos et al., 2006).

The FPR acts as a suitable indicator of energy status (Buttchereit et al., 2010). Here, we observed the highest FPR (1.32) in February, indicating that cows mostly utilize their body fat in February. As for other milk indices, similar seasonal patterns have been found in several studies (Allore et al., 1997; Fox and McSweeney, 1998; Heck et al., 2009). These indices are related, more or less, to hot temperature in most regions. Therefore, one of the best ways to improve milk quality is to make the environmental temperature appropriate to create comfortable conditions for dairy cows.

Effects of Parity on the 9 Indices

Duncan's multiple comparison of parities (Table 5) showed that both DMY and MFP in the first parity were lower than other parities ($P < 0.05$). The main reason for this is that heifers need more amino acids and fat for their body growth (Oltner et al., 1985).

Some studies reported that the parity of Holstein cows influences the concentrations of milk corticosteroid (Fukasawa et al., 2008) and MUN (Cao et al., 2010). Other studies described the relationship between parity and milk composition in sheep (Zeng and Escobar, 1995; Sevi et al., 2000; Carnicella et al., 2008) and reported that the effect of parity on DMY was not significant, but the milk protein, casein, and milk fat contents in the third parity were significantly higher than those in the first and second parities (Sevi et al., 2000), which are different from our findings.

In the present study, MPP, MLP, and MSP were the lowest in the fourth lactation ($P < 0.05$). The SCC, SCS, and MPL were significantly increased from the second parity ($P < 0.05$), indicating that with increasing parity, the milk quality and hygiene status deteriorated, whereas the milk loss increased. This may be part of the reason why cows in China have to be culled after

Table 5. Duncan's multiple comparisons of 9 indices in different parities from first to fourth¹

Parity	DMY, kg/d	MFP, %	MPP, %	MLP, %	MSP, %	FPR	SCC, $\times 10^4$ /mL	SCS	MPL, kg/d
1	25.94 ^b	3.88 ^b	3.09 ^{ab}	4.91 ^a	12.58 ^a	1.26 ^b	29.73 ^c	3.94 ^c	0.74 ^c
2	28.45 ^a	3.93 ^a	3.11 ^a	4.85 ^b	12.59 ^a	1.28 ^a	32.24 ^{bc}	4.08 ^b	0.86 ^b
3	28.81 ^a	3.92 ^a	3.09 ^{ab}	4.85 ^b	12.56 ^a	1.27 ^{ab}	34.46 ^{ab}	4.18 ^a	0.97 ^a
4	29.05 ^a	3.91 ^{ab}	3.07 ^b	4.82 ^c	12.49 ^b	1.27 ^{ab}	35.36 ^a	4.23 ^a	1.00 ^a

^{a-c}Values within a column with different superscripts are significantly different ($P < 0.05$).

¹DMY = daily milk yield; MFP = milk fat percentage; MPP = milk protein percentage; MLP = milk lactose percentage; MSP = milk solids percentage; FPR = fat-to-protein ratio; MPL = milk production loss.

the third parity. The cows in the third parity accounted for 87.5% of those in the second parity, whereas the number of cows in the fourth parity rapidly declined to half of cows in the third parity. Therefore, considering the potential economic returns, it is important for dairy enterprises to improve health and production performance, rather than milk yield in the dairy cows at high parity.

Correlation Among the 9 Indices

The canonical correlation analysis of the 9 indices (Table 6; $P < 0.05$) demonstrated that DMY was negatively correlated with MPP ($r = -0.4077$), but no significant relationship was observed between DMY and the other 7 indices. The decrease in protein concentration may be due to the increased efficiency of milk production (Wu and Huber, 1994). The MSP is composed mainly of MFP, MPP, and MLP; here, we found that MSP was significantly correlated with MFP ($r = 0.8094$), MPP ($r = 0.6449$), and MLP ($r = 0.4856$). Sutton (1989) mentioned that nutrients in the diet obviously affected milk fat, but not milk protein and lactose, which is consistent with our result that MSP was more easily influenced by MFP. In addition, the FPR had a positive relationship with MFP ($r = 0.6749$) and a negative relationship with MPP ($r = -0.5337$), which was conceptually reasonable. Moreover, SCC was positively correlated with SCS ($r = 0.8306$) and MPL ($r = 0.7861$), whereas a high positive correlation was observed between SCS and MPL ($r = 0.8360$). Several articles from the literature also demonstrated that the decrease in milk production is increased with increasing SCC (Miller et al., 2004; Halasa et al., 2009; Hand et al., 2012).

Models for Seasonal Variation of Major Milk Components

The values of MSP, MPP, MFP, or MLP from January to December were grouped into 4 parity groups; the analysis results showed the changes in major milk

Figure 1. Seasonal variation in milk solids percentage from parity 1 to 4. Color version available in the online PDF.

components in different parities (Figures 1–4). Based on the arithmetic average of each index in different parities from January to December, parameters of the Wood model (Wood, 1967) were estimated and showed good performances in convergence. The following equations were obtained and used to fit the seasonal trends of major milk components in mixed-cow herds:

$$\text{MSP} = 12.862 \cdot t^{-0.0317} \cdot e^{0.00459t}; \quad [1]$$

$$\text{MPP} = 3.094 \cdot t^{-0.0464} \cdot e^{0.0117t}; \quad [2]$$

$$\text{MFP} = 4.2116 \cdot t^{-0.0344} \cdot e^{0.00276t}; \quad [3]$$

$$\text{MLP} = 4.9824 \cdot t^{-0.0196} \cdot e^{0.00106t}; \quad [4]$$

where t represents each natural month.

A significant relationship was observed between milk composition and month. The results showed that MSP decreased quickly in the first quarter and then entered the stable period (Figure 1). The seasonal change of MPP was similar to MSP, but obvious valley values were observed in the curve of MPP (Figure 2). A fluctuant reduction in MFP was found in Figure 3, and MLP in 4 parities showed a downward but relatively stable

Table 6. The inner correlation of canonical correlation analysis of 9 indices ($P < 0.05$)¹

Correlation	MFP	MPP	MLP	MSP	SCC	SCS	MPL	FPR
DMY	-0.1134	-0.4077	0.1504	-0.2013	-0.0505	-0.0719	0.2133	0.2033
MFP		0.2525	0.1157	0.8094	0.0478	0.0622	0.0070	0.6749
MPP			0.0803	0.6449	0.0744	0.1501	-0.0143	-0.5337
MLP				0.4856	-0.1473	-0.1975	-0.1048	0.0250
MSP					0.0096	0.0343	-0.0409	0.2094
SCC						0.8306	0.7861	-0.0183
SCS							0.8360	-0.0643
MPL								0.0121

¹DMY = daily milk yield; MFP = milk fat percentage; MPP = milk protein percentage; MLP = milk lactose percentage; MSP = milk solids percentage; FPR = fat-to-protein ratio; MPL = milk production loss.

Figure 2. Seasonal variation in milk protein percentage from parity 1 to 4. Color version available in the online PDF.

Figure 4. Seasonal variation in milk lactose percentage from parity 1 to 4. Color version available in the online PDF.

trend (Figure 4). We tried to build linear regression models of MSP, MPP, MFP, and MLP, but the results were not good enough because of the seasonal fluctuation in milk composition. In this case, nonlinear models had been chosen to fit these fluctuations. This study found that the Wood model (Wood, 1967) was more suitable to simulate the seasonal dynamic variation of milk compositions compared with the Dijkstra model (Dijkstra et al., 1997) and Gompertz model (Gompertz, 1825). An important finding in our study was that the Wood model could not only be used to set up lactation curves of dairy cattle (Xiong et al., 2011), but was also good at simulating seasonal changes in milk composition. Nevertheless, it is necessary to evaluate these models in more studies.

According to the changing patterns of major milk components shown in the models, dairy farmers could improve feeding management and nutritional supplementation of Chinese Holstein cows to minimize differences in major milk components in different seasons. Specifically, to improve the low level of major milk

components in the summer shown in the models, improving feeding management, including the addition of fans and sprinklers, and shade and barns, which enhance passive ventilation, could increase body heat loss and improve the DMI of cows, thereby increasing the concentrations of major milk components in raw milk (West, 2003). On the other hand, modifying nutritional manipulation, including increasing the concentration of net energy and MP in dairy rations and the supplementation of dairy rations with protected vegetable or fish oils could directly improve the concentration of MPP and MFP in raw milk (Emery, 1978; Spoerndly, 1989; Chilliard et al., 2001).

CONCLUSIONS

The DHI data of Chinese Holstein cows were mined and analyzed using GLM ANOVA and PROC CANCORR of SAS (SAS Institute, 1999). The results showed that season and parity significantly affected milk yield, milk composition, and other related indices. Season, especially the high-temperature season, significantly affected the production, components, and sanitary state of milk. Correlation analysis demonstrated that MSP was significantly correlated with MFP, MPP, and MLP, and the correlations among SCC, SCS, and MPL were also significant. The Wood equation was appropriate for a quantitative description of seasonal variation in milk composition in individual cows or herds. The changing patterns of milk composition and related indices in different months and parities provide scientific evidence to reasonably improve the feeding management and nutritional supplementation of Chinese Holstein cows.

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Figure 3. Seasonal variation in milk fat percentage from parity 1 to 4. Color version available in the online PDF.

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