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Article

Evaluation of biomarkers of heat stress by using automatic health monitoring system in dairy cows

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Simple Summary: The hypothesis of current study was to evaluate possible relationships between temperature-humidity index and biomarkers: reticulorumen ph and temperature and rumination time, milk traits, body weight, consumption of concentrate during summer period. Some biomarkers of heath during summer period in can be milk yield, milk lactose, somatic cell count, concentrate conception, rumination time, body weight, reticulorumen ph and milk fat – protein ratio. We can recommend to monitoring these parameters in the herd management program to identify possibility of heat stress.

Abstract: The objective of this study was to evaluate biomarkers of heat stress (HS) from automatic milking system (AMS), the relationships between measurements of temperature-humidity index (THI) and reticulorumen ph and temperature and some automatic milking systems parameters in dairy cows (rumination time (RT), milk traits, body weight (BW) and consumption of concentrate (CC)) during summer period. The experiment was carried out on a dairy farm at 54.9587408, 23.784146. Lithuanian Black and White dairy cows (n=365) were selected. The cows were milked with Lely Astronaut® A3 milking robots with free traffic. The rations were calculated to meet physiological requirements of the animals. Daily milk yield, rumination time, body weight, milk fat and protein ratio were collected from the Lely T4C management program for analysis. The pH and temperature of the contents of cow reticulorumen were measured using specific smaX-tec boluses manufactured for animal care. The daily humidity and air temperature in the farm zone were obtained from the adjacent weather station (2 km away). According this study during HS, the higher THI had positive correlation with ML, which show tendencies to increase risk of mastitis, and decrease CC, RT, BW, MY, reticulorumen ph and F/P. Some biomarkers of HS can be milk yield, milk lactose, somatic cell count, concentrate conception, rumination time, body weight, reticulorumen ph and milk fat – protein ratio. We can recommend to monitoring these parameters in the herd management program to identify possibility of heat stress.

Keywords: heat stress; cow; automatic milking system; temperature; milk

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1. Introduction

Rising ambient temperatures will increase the incidence of heat stress in livestock, particularly in dairy cows [1] Given the anticipated changes in climatic conditions globally, the number and intensity of heat stress periods will further increase [2]. Heat stress (HS) is becoming a serious problem due to the negative impacts it has on ruminant performance [3]. The negative effect of HS will become more severe in the future, as the consequence of global warming progresses and genetic selection for milk yield continues.

The hot environment is one of the major factors that could negatively influence milk yield of lactating cows, particularly in high producing animals [4]. Effects of thermal stress in dairy cows are

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manifold and has been reported on milk yield and composition [2], reproductive performance, welfare and health [5]. Physiological responses, such as respiration rate, core-body temperature, rectal temperature, skin temperature, sweating rate, feed and water intake, and production performance (milk yield, body weight gain, conception success rate), are affected to varying levels by heat stress [6]. Heat stress has adverse effects on milk production and reproduction in dairy cattle [1,7]. Heat-stressed animals change their metabolism in response to ambient temperature change [3]. As noted by Hansen [7] the problem of heat stress is a growing one because increases in milk yield result in greater metabolic heat production, which is due to anticipated changes in the global climate. Climatic condition is expected to mainly affect the welfare and productive performance of livestock animals [8]. Impacts on performance and functional traits are mainly caused indirectly by alterations in drinking and feeding behaviour [1]. Heat stress has a negative impact on animal health and higher rate of clinical mastitis or higher milk somatic cell counts in dairy cows exposed to hot environment [9]

Temperature-humidity index (THI) is significant because it involves the collective impacts of air temperature and humidity related to different levels of thermal stress [10]. The impacts of ambient environment on cow performance had been evaluated by temperature humidity index (THI), which was noted as a distinctive index and represented the combined impacts of ambient temperature and humidity [11] It has been adapted to describe thermal conditions that drive heat stress in dairy cattle [12]. Despite not being formulated using cow data, THI are related to body temperatures of cattle exposed to heat stress [13]. Recently, Bohmanova et al. [14] showed that various THI were predictive of milk yield in cows in the southeastern part of the United States and there are newer studies from Europe. According to West et al., [1] environmental modifications should target the effects of high temperatures on cow body temperature and should modify the environment at critical times during the day when cows are stressed, including morning hours when ambient temperatures are typically cooler and cows are not assumed to be stressed. The THI and several variations of it have been used extensively to estimate the degree of heat stress in dairy and beef cattle [15].

The automatic monitoring of core body temperature in dairy cattle could be useful for the identification of illness, heat stress, and general physiological stress. Body temperature is associated with rumen content temperature. Rumen temperature may be monitored by reticular temperature sensors [16] as well as rumination time (RT) may be assessed and recorded by RT sensors [17]. The Smart Bolus (TenXSysInc., Eagle, ID) system used a reticulorumen bolus to automatically record and transmit dairy cow temperatures [18]. The ambient temperature, milk production and breed influence the reticulorumen temperature [19].

The majority of earlier researches were interested in milk, protein and fat production alterations during HS. According to Nasr et al. [10], slight and scarce data exist in the literature regarding the relationships between THI, milk, fat%, protein % and SCC. Nars et al., [10] found that high THI is associated with higher SCC with a reduction in milk yield and quality, therefore, potentially reducing both welfare and economic return under subtropical Egyptian conditions. Indicators of heat stress can be directly measured on the animals (behavioural, physiological, productive and reproductive indicators) as well as those environmental parameters that can be considered as risk factors [20]. Early forecasting of heat stress risk makes it possible to limit its negative impact on cow welfare [20].

The objective of this study was to evaluate biomarkers of HS from AMS, the relationships between temperature-humidity index (THI) and reticulorumen ph and temperature and some automatic milking systems parameters in dairy cows (rumination time (RT), milk traits, body weight (BW) as well as consumption of concentrate (CC) during summer period in central Europe region.

2. Materials and Methods

2.1. Location, animals and experimental design.

The study was conducted during summer period from 2018-07-01 to 2018-08-31. The experiment was carried out on a dairy farm in 54.9587408, 23.784146. Lithuanian Black and White breed dairy cows (n=365) were selected based on the following criteria; having had a 2nd or more lactations (on average 2.9±0.13 lactation), from 1 to 200 (on average 98±6.15) days in milk (DIM), and being clinically healthy. The cows were kept in a loose housing system and were fed total mixed ration (TMR) throughout the year at the same time, balanced according to their physiological needs. Cow feeding took place every day at 06:00 and 18:00.

The cows were milked with Lely Astronaut® A3 milking robots with free traffic. To motivate the cows to visit the robot, 2 kg/d of concentrates were fed to them by the milking robot. The cows were kept in a loose housing system and fed total mixed ration (TMR) throughout the year at the same time, balanced according to their physiological needs. Cows were fed a TMR consisting of 25% corn silage, 15% grass silage, 4% grass hay, 50% grain concentrate mash and 6% of mineral mixture. Diets were formulated according to meet or exceed the requirements of a 550 kg Holstein cow producing 35 kg/d. Composition of ration – dry matter (DM (%) 48.8; neutral detergent fiber (% of DM) 28.2; acid detergent fiber (% of DM) 19.8; nonfiber carbohydrates (% of DM) 38.7; crude protein (% of DM) 15.8; neto energy for lactation (Mcal/kg).

2.2. Measurements.

Daily milk yield, rumination time, body weight, milk fat and protein ratio were collected from the Lely T4C management program for analysis.

The pH and temperature of the contents of cows (n=20) reticulorumens were measured using specific smaX-tec boluses manufactured for animal care. SmaXtecanimal care technology®enables the continuous real-time display of data such as ruminal pH and temperature. According to the directions of the manufacturer, the boluses were inserted into the reticulo-rumens of the cows investigated with the help of a specific tool. The data was measured with the help of specific antennas (smaXtec animal care tech-nology®). For monitoring the reticuloruminal pH, an indwelling and wireless data transmitting system (smaXtec animal care GmbH, Graz, Austria) was used. The system was controlled by a microprocessor. The data (pH temperature) was collected by means of an analogue to digital converter (A/D con-verter) and stored in an external memory chip. Due to its dimensions (length:12cm, width:3.5cm, weight: 210 g), this indwelling system can be orally administered to an adult cow and is shock-proof and resistant to rumen fluid. Calibration of the pH-probes was performed at the beginning of the experiment using pH 4 and pH 7 buffer solutions. The data were read every 10 minutes daily. The study was conducted from 2018-07-01 to 2018-08-31. All data were obtained by smaXtec messenger®computer software.

The average of daily humidity and air temperature in the farm zone were obtained from the adjacent weather station (2 km away). Measurements were made every hour. The collected data were used to compute the daily temperature-humidity index using the formula described by Kendall and Webster (2009). THI=(1.8* AT +32) – ((0.55 – 0.0055* RH) – (1.8* AT – 26)), where AT = Ambient temperature (°C), RH = Relative humidity (%).

2.3. Data analysis and statistics

The records (n=33480) of the tested cows (n=365) in the summer season (92 days) were analyzed using IBM SPSS statistics Version 20.0 for Windows. The evaluated indicators were normally distributed (were tested for normality using Kolmogorov-Smirnov test; P>0.05). The test-day data of SCC were transformed to somatic cell score (SCS) in order to achieve a normal distribution for this indicator (SCS = log2(SCC/100) +3).

The descriptive statistics (mean and standard error), one-way ANOVA test and correlation coefficient between evaluated traits of cows (MY –milk yield, BW, CC, RT, ML – milk lactose, SCS and F/P -milk fat and protein ratio) and metrological parameter (THI) were estimated. The relationship between the categorical variables (classes of THI, RT, milk SCC and F/P) were evaluated using $\chi 2$ test. The results were considered reliable under P level <0.05.

The data of cows were grouped according to level of the risk to ruminal acidosis by rumination time (min/d.): very high risk of ruminal acidosis (RT<432) and high risk of ruminal acidosis (432≤RT≥473) [21]. We grouped the data of SCC into two classes: healthy cows – milk SCC<200 thousand/ml and risk of subclinical mastitis – SCC≥200 thousand/ml.

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The THI was used to determine the influence of heat stress on dairy cows and it is divided into 2 categories: THI <72 (comfort zone) and THI ≥72 (higher risk for thermal stress) [22].

3. Results

3.1. Relationship between THI with milk traits, body weight, consumption of concentrates and rumination time in dairy cows.

During the summer period we have set 7 days (7.5%) for THI≥72. The average value of THI was 64.58±0.272, average of AT - 18.92±0.191 °C, and RH - 70.62±0.620%.

The milk SCC was negatively correlated with THI (r=-0.351, P<0.001). The ML was positively related with THI (r=0.120-0.124, P<0.01). The THI was negatively related with CC (r=-0.065, P<0.01) and milk F/P (r=-0.009; P<0.01).

There was a significant difference in daily MY (it reduced by 4.17% from low THI to high THI). Milk lactose % and F/P were not affected by THI. The BW of cows was slightly affected by THI with the lowest BW (-5.26 kg) found at the highest THI level (Table 1). The heat stress decreased to average of the CC (53 g, P<0.001) (Table 1).

Table 1. The evaluated characteristics in dairy cows at different levels of temperature-humidity index during the research period.

THI (daily average)		RT	BW	MY	F/P	SCC	ML	CC
<72	M	447.53^{a}	677.37^{a}	36.94^{a}	1.13^{a}	165.95^{a}	4.59^{a}	3096.29^{a}
(n=30960; 92.5%)	SEM	3.24	1.22	0.25	0.01	10.37	0.001	17.45
≥72 (n=2520; 7.5%)	M	433.57^{b}	672.11^{b}	35.4^{b}	1.13^{a}	152.14^{b}	4.59^{a}	3043.00^{b}
	SEM	14.91	5.65	1.97	0.03	26.21	0.001	18.12

^{a,b} Values within a column with diferent superscripts differ significantly at *P*<0.05. RT – rumination time; BW – body weight; MY – milk yield; F/P- milk fat protein ratio; SCC – somatic cell count; ML – milk lactose; CC - consumption of concentrate (CC).

Although, the study showed that the mean of SCC in the risk class of THI was lower (Table 2), on the other hand, there was 5% higher number of cows with an increased risk of mastitis (SCC≥200 thousand/ml) at a higher THI level (Table 2).

Table 2. Relations between THI with risk level to mastitis and ruminal acidosis in dairy cows.

THI	SCC (thousand/ml)		F/P		RT	
	<200	≥200	>1.15	1.15-1.20	<432	432-473
<72	90.7%	9.3%	85.7%	14.3%	2.3%	97.7%
≥72	85.7%	14.3%	84.9%	15.1%	42.9%	57.1%
χ2 test	P=0.049		P=0.867		P=0.000	

RT – rumination time; BW – body weight; MY – milk yield; F/P- milk fat protein ratio; SCC – somatic cell count; ML – milk lactose; CC - consumption of concentrate (CC).

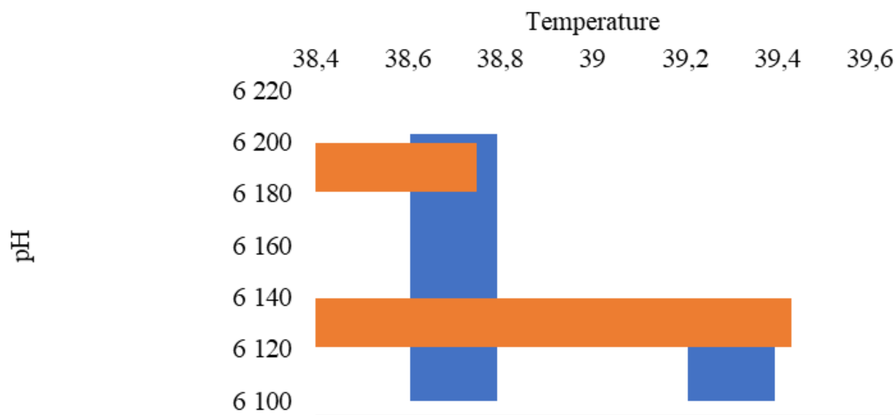
It was also found that THI (≥72) had significantly more (40.6%) cows which were prone to a very high risk of rumen acidosis depending on the time of rumination (when RT <432).

3.2. Relation of THI with reticulorumen pH and temperature.

From our investigation, the average reticulum pH of cows was 6.17±0.003, and reticulum temperature – 39.12 ±0.003°C.

There was a negative correlation between reticulum pH and THI (r=-0.381 P<0.001) and between reticulum temperature and THI (r=-0.439, P<0.001).

The increase in THI is related to decrease in reticulum pH (from 6.25 to 6.09) (P<0.05). The lower (1.72%, P<0.05) reticulum temperature was estimated at the higher level of THI≥72 (Fig. 1).



	THI<72	THI≥72
Reticululum pH	6 203	6 136
Reticulum temperature °C	39,43	38,75

Figure 1. The reticulum pH and temperature characteristics in dairy cows at different levels of THI.

4. Discussion

According to Nasr et al. [10], thermal stress may influence animal welfare as well as milk yield and composition. THI is calculated from ambient temperature and relative humidity and can help to evaluate the impacts of environment on cow performance and health [1,15]. High THI (higher than 72), might irritate the neuroendocrine system, consequentially influence energy and water balance, hormonal equilibrium and body temperature, which ultimately disturb the growth, reproduction, milk production and the immune system [23]. These investigators reported that a daily afternoon maximum THI of 87 and a minimum morning THI of 77 should be considered as the upper and lower daily THI values for maximum risk of death of dairy cows to heat stress [24]. According to Collier et al. [23]. The indirect effects include, impacts of the environment on food and water availability, pest and pathogen populations, and resistance of the immune system to immunologic challenge. Supplemental rumen-active fat has advantages over starch-based concentrate to increase energy density of diets for lactating cows during the warm summer months. Addition of fungal cultures to dairy diets has been shown to reduce respiration rate and body temperature during thermal stress in several studies. Rumen acidosis is often increased under heat-stress conditions associated with reduced forage intake and increased high energy feed intake [23].

4.1. Heat stress and SCC

Bertocchi et al. [9] have reported the normal SCC in healthy cow milk to be 100.000–314.000 cells/mL. In the current study, the SCC was 158.000 – 191.000 cells/mL. Somatic cells are an important constituent in the milk used as a marker for healthy mammary gland and milk quality [25]. In our study, we found that the mean of SCC in the risk class of THI was lower, on the other hand, there was 5% higher number of cows with an increased risk of mastitis at a higher THI level. According to Nasret al. [10], high THI (higher than 72) was associated with higher SCC (SCC significantly increased 36%) and production losses in Holsteins, and SCC was within the normal level for healthy mammary gland and it was positively associated with the THI level. SCC was higher in milk from Holstein dairy cows suffering from heat stress than dairy cows under the comfort condition [26]. Bertocchi et al. [9] stated that, there was a significant increase in SCC of lactating Holsteins in cold weather, which decreased in hot weather. It is possible that increased SCC of milking Holstein cows through hot weather may be attributed to several types of stress [27]. In summer, the growth and count of bacteria is amplified in the animal bedding stuff owing to suitable temperature and humidity which facilitate the extra exposure of teat ends to these bacteria. Elevated SCC was coupled with a reduction in the synthetic performance of the mammary gland [28].

4.2. Heat stress and MY

Climate is the biggest single factor affecting animal production and welfare (HS adversely influences dairy cattle welfare and productivity [23,29]. Reduced milk productivity of cows is considered as the most negative effect of heat stress as its economic results are usually visible after a few days [30]. Nevertheless, it was found that MY reduced by 4.17% from low THI (<72) to high THI (≥72). Furthermore, Nasr et al. [10] found that the daily milk production was higher in low THI when compared with the higher THI. In dairy cattle, heat stress decreases milk yield [31] which has been traditionally attributed to the heat-induced reduction in DMI [1]. However, the utilization of pair-fed thermal neutral controls demonstrated that reduced DMI only partially (about 50%) explains the decrease in productivity, suggesting that hyperthermia itself directly affects milk production [31]. The high-yielding dairy cows started to reduce milk yield at a THI of approximately 68 [32]. In the current study, dairy cows were exposed to conditions well above this threshold (average THI = 82.4), causing hyperthermia, tachypnea, and marked DMI reductions [33].

According to Nasr et al. [10], cows at low THI were better in milk yield and composition. When a cow becomes heat stressed, an immediate coping mechanism is needed to reduce DMI, causing a decrease in the availability of nutrients used for milk synthesis [1]. Simultaneously, there is an increase in basal metabolism caused by activation of the thermoregulatory system. Mild to severe heat stress can increase metabolic maintenance requirements by 7 to 25% [34]. further exacerbating both the existing metabolic stress and the decrease in milk production. As a result, it is often assumed that milk production can be interpreted to be a direct welfare indicator in that it offers producers the ability to monitor the animal's individual response to a challenging event (e.g., increasing ambient temperature or changes in nutrition) [35]. However, others have challenged the use of milk production as an acceptable welfare indicator [36]. Despite the documented challenges with using milk production as an indicator of welfare in lactating dairy cows, very recent evidence suggests that changes in milk composition may be more useful to assess cows in immediate heat stress [37]. Du Preezet al. [38] under Southern African conditions found that productivity of cows was affected by heat stress when THI values were higher than 72 (which corresponds to 22 °C at 100 % humidity, 25 °C at 50 % humidity, or 28 °C at 20 % humidity).

Increasing the ambient temperature was coupled with production losses in livestock farms which are a portion of the continuing and expected environmental alteration [8]. High yielding lactating cows are mostly sensitive to thermal stress especially in hot environment [39] which is attributed to the significant increase in metabolic heat output of high milk production [1]. Direct effect of HS on milk production, independent of intake [31].

During summer period, THI and AT showed similar correlation coefficients with cow's productivity, BW, RT and CC. Our study showed that, when THI reached 72, milk production of cows as well as feed intake decreased. We found that under central Europe climatic conditions, milk yield drops by 0.22±0.001(R^2=0.821, P<0.001) kg per cow per day for each point increase in the value of THI above 72to the level of THI=78. The heat stress influenced decreasing of CC in dairy cows (1.72%, P<0.001). Beside changes in milk yield, heat stress could also decrease rumination time of cows (3.12%) (Table 1) and increase frequencies of mastitis (5%) (Table 2).

To improve and alleviate the heat disparity, a reduction in feed intake, metabolism, body weight and milk yield had occurred [40]. Heat stress especially in summer was associated with fodder shortage which might be the main reason for reduction in milk production [41].

4.3. Heat stress and milk composition

Milk composition is also discordantly altered during hyperthermia, which indicates that HS regulates component synthesis in addition to its overall effect on milk yield [31]. Milk F/P correlated negatively with THI - negatively (r=-0.009-0.020; P<0.01). Nasr et al. [10] revealed that percentages and yields of fat and protein were lower in the high THI compared to the low THI. The fat% in cool weather was higher than in hot weather [9] but was lower than that reported by other researchers [42]. The reduction in saliva HCO3− content and the decreased amount of saliva entering the rumen make the heat-stressed cow much more susceptible to subclinical and acute rumen acidosis [43].

The milk lactose (ML) was positively correlated with AT and THI (r=0.120-0.124, P<0.01), but was negatively correlated with RH (r=−0.208, P<0.01). Nasr et al. [10] revealed that lactose % did not differ between low and high THI which was in agreement with the findings reported by other researchers [44].

4.4. Heat stress and RT, BW and CC

There was a negative relationship between RT, BW of cows and metrological parameters (P<0.01). The CC was positively related with RH (r=0.117, P<0.01) and negatively – with AT and THI (r=-0.065-0.084, P<0.01). Moallem et al. [45] indicated that the primary negative effect of THI is a depression of RT, which subsequently led to a reduction in DMI, followed by a decline in milk yield. A statistically significant effect of THI on RT was identified, with RT decreasing as THI increased [46]. During heat stress, rumination is reduced in cows [45], and a lower blood flow to rumen epithelium is observed. Acatincai et al. [47] observed that with an increase in air temperature above 27 to 28°C, the rumination process was severely affected in Romanian Black and White multiparous cows. Moallem et al. [45] indicated that the primary negative effect of high temperature-humidity index (THI) is a depression of rumination time (RT), which subsequently led to a reduction in DMI, followed by a decline in milk yield. Those authors suggested that rumination time plays a pivotal role in the negative effect of heat stress on DMI and, consequently, on milk production. However, Bernabucci et al. [48] recently showed that reduced nutrient intake (an indirect effect of heat) accounts for only about 35% of the heat stress–induced decrease in milk synthesis. Moallem *et al.* [45] indicated that the primary negative effect of high temperature-humidity index (THI) is a depression of rumination time (RT), which subsequently led to a reduction in DMI, followed by a decline in milk yield. Those authors suggested that rumination time plays a pivotal role in the negative effect of heat stress on DMI and, consequently, on milk production.

Energy intake is considered the most production-limiting nutrition component during summer [1]. According Renaudeau et al. [5] heat-stressed dairy cows reduced the feed intake to avoid excessive metabolic heat production, while maintenance costs increased [49]. However, recently, it was demonstrated that reduced intake accounted for only 35% of the decline in milk yield under heat stress conditions, and it was suggested that a shift in postabsorptive metabolism and nutrient partitioning might account for the remaining reduction in milk yield [50]. Mean air temperature had the greatest influence on milk yield for Holstein cows fed high or low concentrate diets under hot conditions [51].

As rumination time decreases with increasing THI [17], ruminal passage rate as well as DMI is reduced. For instance, heat stress animals reduce DMI, activity, and metabolic rate in an attempt to decrease metabolic heat production [34].

Recent research [21] showed that decrease in RT is related to the prevalence of ruminal acidosis in dairy cows. We found that the risk of acidosis increased significantly (40.6%) when THI ≥72, because RT reached the lowest level (RT<432).

4.5. Heat stress and reticulorumen ph and temperature

High environmental temperature adversely affects rumen health [52] due to a variety of biological and management reasons [17]. We found that increase in THI is related to decrease in reticulum pH and the lowest reticulum temperature was estimated at the highest levels of AT and

THI and at the lowest level of RH. In agreement with Lefcourt et al. [53], the daily maximum RT was significantly greater than the daily minimum RT in all seasons. The reticulorumen temperatures decreased after water intake and returned to the baseline temperature within 0.3 to 3.5 hr [54]. Season also had a profound effect on the circadian body temperature rhythm, but there was little difference between the breeds of dairy cows [11]. This is important for assessment of the health condition by temperature. However, these changes may also reflect a biological change in the core body temperature associated with changing seasons. The seasonal body temperature baseline may be useful for interpretation of the differences in body temperature and used for cow management by season. At a minimum, these differences indicate the need to adjust the thresholds used for management purposes by season [19]. The summer months may be associated with heat stress. A heat-stressed cow is prone to rumen acidosis, and many of the lasting effects of warm weather (laminitis, low milk fats, etc.) can probably be traced back to a low rumen pH during the summer months [55]. According to Antanaitis et al. [56] the reticulorumen temperature in lactating cows was found to be influenced by circadian rhythm, and season (the lowest temperature observed in the springtime was 38.81±0.001, and the highest was in autumn, 39.17±0.001).

Sievers et al. [57] suggested that an intraruminal measuring system would be advantageous because it is independent of external disturbing factors, cannot be manipulated from the outside, and is less likely to be lost. Rumen temperatures have been established as effective measures of core body temperature [58]. Because of the activity of heat-producing rumen microorganisms, ruminal or reticular temperatures are generally approximately 0.5°C greater than the core body temperature [59, 58].

According to Gao et al. [60], ruminal pH was reduced (9.5 and 6% before and after feeding, respectively) during heat stress. Additionally, the intraruminal temperature may affect rumen metabolism [61]. These authors showed that an increase in intraruminal temperature implies a reduction in feed and water intake (water intake increased only when needed for the thermoregulation of the whole body), implying a decrease in volatile fatty acids production and a shift in their composition with a significant decrease in the acetate to propionate ratio [3].

5. Conclusions

According this study during HS, the higher THI had positive correlation with ML, which show tendencies to increase risk of mastitis, and decrease CC, RT, BW, MY, reticulorumen ph and F/P. Some biomarkers of HS during summer period in can be milk yield, milk lactose, somatic cell count, concentrate conception, rumination time, body weight, reticulorumen ph and milk fat – protein ratio. We can recommend to monitoring these parameters in the herd health management program to identify possibility of heat stress.

Author Contributions: Ramunas Antanaitis: supervision of the whole study; Vida Juozaitiene: software and algorithm development, design and setup of field experiments, data collection and analysis; Dovile Malasauskiene: setup of field experiment and data collection, selection and management of the experimental group of animals; Mindaugas Televicius: setup of field experiment and data collection, selection and management of the experimental group of animals. Arunas Rutkauskas setup of field experiment and data collection, selection and management of the experimental group of animals; The manuscript was written by Ramunas Antanaitis and revised by all co-authors.

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