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*Article*

# Effect of Calving Season and Related Climatic Factors on Productive Performance in Dairy Cows

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**Simple Summary:** In the last decade we are witnessing serious climate changes related to global warming. This affects all living beings including dairy cows. As the climate warms, dairy cows in the temperate climate zone experience heat stress, which affects the quantity and quality of milk they produce. In the present scientific work, the influence of the calving season on the yield and composition of cow's milk was studied. As a result of the study, it was found that cows calved in summer, under conditions of heat stress, had the lowest milk yield for lactation, the lowest milk yield at peak of lactation and the lowest fat content compared to cows calved in other seasons of the year. Regarding the protein content of the milk, a tendency was found for cows calved in the summer to have the highest values of this trait.

**Abstract:** The aim of the research was to study the influence of the calving season in the conditions of the upcoming climate changes on the productive traits of dairy cows in Bulgaria. The study was conducted in a cattle farm with a capacity of 500 dairy cows, loose-housed in open free stall barn (shed type). In the research 286 lactations of 199 Holstein cows from the studied farm were included. The cows with the highest average milk yield for lactation calved in the spring - 8522.2 kg, and the cows with the lowest milk yield calved in the summer - 8082.7 kg. Cows calved in the spring had the highest maximum daily milk yield (lactation peak) - 38 kg, and the lowest in the summer - 35.7 kg. Regarding the composition indicators of milk, fat and protein content, no significant effect of the calving season was found, but a tendency for the lowest values for the percentage of fat in milk was reported for cows calved in the summer - 3.68%, and the most - high for those calved in the spring - 3.71%. Regarding the percentage of protein in the milk, the lowest values were for cows that calved in autumn - 3.19%, and the highest for cows that calved in summer - 3.27%.

**Keywords:** calving season; THI; lactation curve; fat content; protein content

## 1. Introduction

Environmental conditions are a major stressor that affects animals and can lead to serious changes in their physiological indicators [1]. Until recently, it was assumed that high ambient temperatures negatively affected animals mainly in tropical regions. Nowadays, we are witnessing appreciable global warming, resulting in a rise in ambient temperature also in more northern latitudes with temperate climate [2–4].

For Europe, temperature increases are expected in all seasons [5]. Regional climate models expect strong warming in large parts of North-Eastern Europe, which is particularly pronounced in winter. The strongest summer warming is expected to be observed in southern and southwestern Europe. Along with rising temperature, changes in humidity (precipitation) and wind regime are expected.

There is evidence that animals in European countries will face heat stress, with climate change leading to hotter summers and an increased risk of extreme weather events [6,7]. As a result of climate change, heat stress (HS) in dairy cows is becoming an increasingly widely discussed topic. In addition, in some regions of the world the problem of heat stress in dairy cows is becoming more pronounced and at a much faster rate than others [8]. According to some authors Hempel et al. [9], in Europe over 30% of the days of the year will have critical conditions in terms of heat stress for cows and may cause up to a 2.8% drop in milk productivity compared to current conditions.

Research conducted by the Department of Meteorology of the National Institute of Meteorology and Hydrology at the Bulgarian Academy of Sciences [10] predicts an increase in the annual air temperature in Bulgaria by 2050 and 2080, respectively, from 1.6°C to 3.1°C and from 2.9°C to 4.1°C. In general, the increase in temperature is expected to be greater during the summer season (July to September).

Climate change is a problem facing farmers in many regions of the world, especially in the livestock industry. Although dairy cattle are able to cope with a wide range of climatic conditions, the sustainability of their productivity is potentially challenged by climate change, especially for larger breeds such as Holsteins [11]. The negative impact of HS on the amount of milk produced by cows is also reported by a number of authors [12,13], but in addition there are studies that report a deterioration of milk quality as content of protein [14] and fat [15]. In Bulgaria, Penev et al. [16] studied the relationship between HS and daily milk yield in dairy cows. André et al. [17] pointed out that the effect of HS depends on the conditions in the particular farm. Therefore, it is recommended to quantify the effect of HS using milk production data collected in the specific farm situation.

All this gives us the basis for conducting a study on the influence of the calving season and the related climatic factors on the indicators of milk productivity in dairy cows loose-housed in semi-open free stall barn, under the climatic conditions of Central South Bulgaria.

## 2.. Materials and Methods

The research was conducted in a cattle farm, which was located in Parvomai municipality, Plovdiv region with GPS coordinates - 41.951257, 25.085302. The farm had a capacity of 500 dairy cows (including lactating, dry cows, heifers and calves). Cows were reared under conditions of open shed type free stall dairy barn production system. The farm had four production buildings and a milking parlor.

The study included 286 lactations of 199 Holstein cows from the studied farm. Only lactation records with at least 8 controls for a lactation available were used - a total of 2744 monthly controls for the period 2017-2022. Milk performance traits - milk yield, percentage of fat and protein in milk were for a standard lactation with duration from 240 to 305 days. Lactations from first to third inclusive were covered, later lactations have wide variation in duration, productivity and uneven performance by calving season and others, therefore excluded from the study.

The cows included in the study were fed year-round with a total mixed ration with a constant composition, according to the physiological condition of the animals and the level of productivity and with constant access to water.

The data on the main climatic factors for the area of the farm - temperature and air humidity - were taken from the nearest Meteorological Station, the city of Plovdiv and were for the period 2017 - 2022. From the data on the temperature and humidity of the air, the values of the Temperature-Humidity Index (THI) were also calculated according to the formula according to Thom [18]:

$$0,8 \times T_0 + \left( \frac{B_0}{100} \right) \times (T_0 + 14.4) + 46.4$$

Where:  $T_0$  is the air temperature in °C,  $B_0$  is the percentage of air humidity.

To report the effect of calving season, all calvings were distributed depending on calving date by season as follows: from 01.12 to 28 (29).02 - winter, from 01.03 to 31.05 - spring, from 01.06 to 31.08 - summer and from 01.09 to 30.11 - autumn.

Lactation stage: 1 – to the 30th day, 2 – from the 31st to the 60th, 3 – from the 61st to the 90th, 4 – from the 91st to the 120th, 5 – from the 121st to the 150th, 6 – from 151 to 180th, 7 – from 181 to 210th, 8 – from 211 to 240th, 9 – from 241 to 270th and 10 – from 271 to 305th day.

The MS Excel package was used for basic statistical processing of the data, and the corresponding STATISTICA modules of StatSoft (Copyright 1990-1995 Microsoft Corp.) were used to obtain means, errors, and analysis of variance.

The following model was used to evaluate the influence of the factors on the productive traits for 305 days lactation:

$$Y_{ijk} = \mu + L_i + S_j + e_{ijk}$$

Where:

$Y_{ijk}$  is the dependent variable (milk yield, average fat % and average protein %);  $\mu$  is the average for the model;  $L_i$  is the lactation number effect,  $S_j$  is the calving season effect, and  $e_{ijk}$  is the random residual effect.

The following model was used to assess the influence of the controlled factors on the productive signs for a Test day:

$$Y_{ijkl} = \mu + L_i + S_j + Pl_k + e_{ijkl}$$

Where:

$Y_{ijkl}$  is the dependent variable (milk yield, fat % and protein %);  $\mu$  is the average for the model;  $L_i$  is the lactation number effect,  $S_j$  is the calving season effect,  $Pl_k$  is the lactation stage effect and  $e_{ijkl}$  is the random residual effect.

By analysis of variance (ANOVA) for the model was obtained by classes of the fixed factors the least squares mean (LSM).

### 3. Results

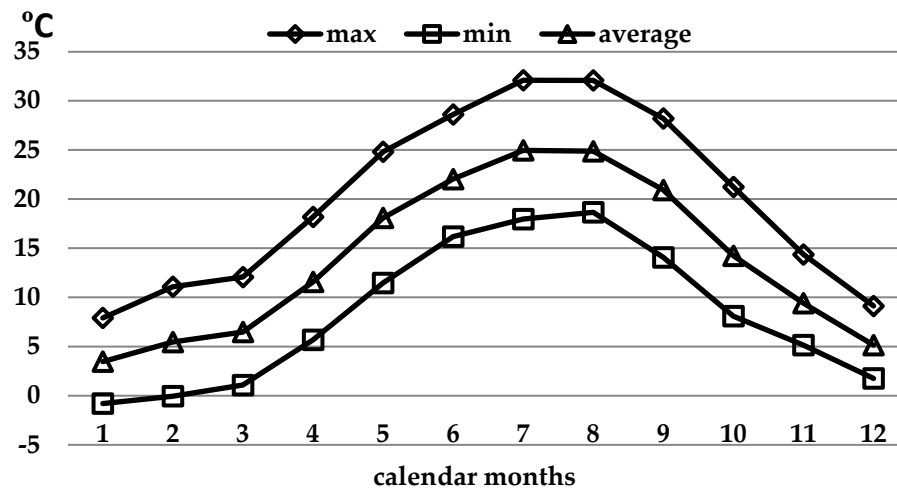
The average milk yield for the 305-day lactation of the cows included in the study was 8277.6 kg, with an average percentage of fat 3.69% and protein 3.22% (Table 1). With the highest milk yield were the cows on third lactation - 8688.4 kg, followed by those on second - 8415.7 kg and with the lowest on first - 8068.8 kg. This ratio in the milk yield is normal and biologically determined. No such pattern was observed in the average percentage of fat and protein in milk for 305-day lactation. Although slightly, both average fat and protein percentages were highest in second-lactation cows, 3.71% and 3.26%, respectively. The cows on first and third lactation had slightly lower values. The differences between the average values for the two productive traits for the cows with different number of lactations were very small.

**Table 1.** Average values and variation of the main productive traits for 305-day lactation by lactation number.

Lactation number	Number	Productive traits for 305-day lactation					
		Milk yield, kg		Average fat, %		Average protein, %	
	n	$\bar{x} \pm Se$	SD	$\bar{x} \pm Se$	SD	$\bar{x} \pm Se$	SD
First	150	8068.78±90.20	1104.75	3.68±0.01	0.163	3.21±0.01	0.096
Second	90	8415.67±147.46	1398.91	3.71±0.02	0.182	3.26±0.05	0.473
Third	46	8688.41±268.52	1819.52	3.70±0.03	0.166	3.20±0.02	0.103
Total	286	8277.57±79.95	1352.08	3.69±0.01	0.170	3.22±0.02	0.277

In Figure 1. the variation of the average monthly minimum, maximum and average daily temperatures for the farm area for the 6 years of the study (2017-2022) is presented. The average minimum temperatures by month were relatively high, as the lowest were recorded in the winter season - around and slightly below 0 °C. The lowest minimum temperatures of -13.3°C were reported

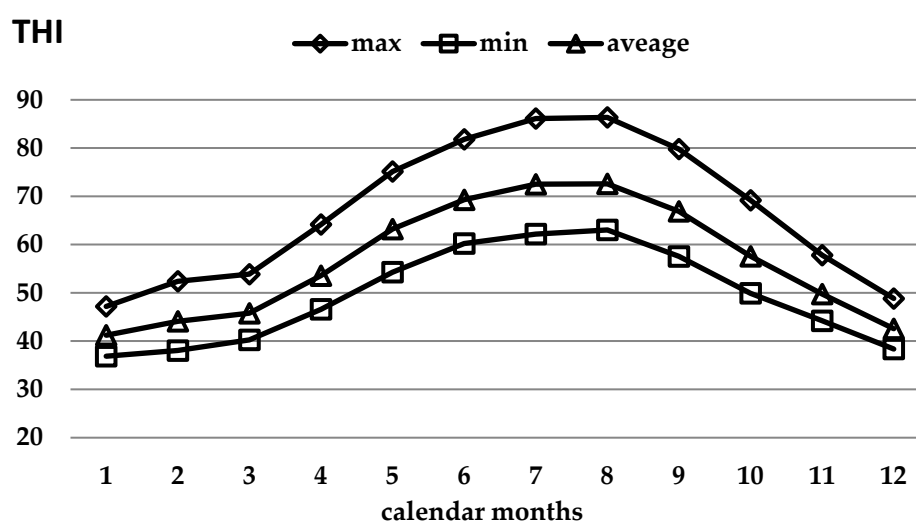
in February in 2017 and 2021, indicating that the farm area had a mild climate without extreme low temperatures. Average daily temperatures were also high, with minimum daily temperatures of around 5°C recorded during the winter months. Relatively high daily temperatures - over 20°C were measured for a rather long period - from June to September. At the maximum temperatures, quite high values were reported. Maximum temperatures above 25°C were reported from May to September. The highest air temperature was recorded in July 2021 – 40.6°C.



**Figure 1.** Average monthly maximum, minimum and average daily temperatures for the farm area for the 6 years by months.

Since the risk of temperature stress is determined not only by air temperature, but also by humidity and other climatic indicators Figure 2 shows the variation of THI values, which reflects the combined effect of the temperature and humidity.

Considered in this aspect, the daily average THI values by calendar months for the period 2017-2022 were over 60 for the period from May to September. For the same period, the maximum daily values of THI were above 72 or risky for heat stress occurrence.



**Figure 2.** Average monthly maximum, minimum and average daily THI values for the farm area for the 6 years by months.

From the analysis of variance (Table 2), it was found that the milk yield for 305-day lactation was significantly affected by lactations number ( $P < 0.001$ ) and calving season ( $P < 0.05$ ). Neither of the

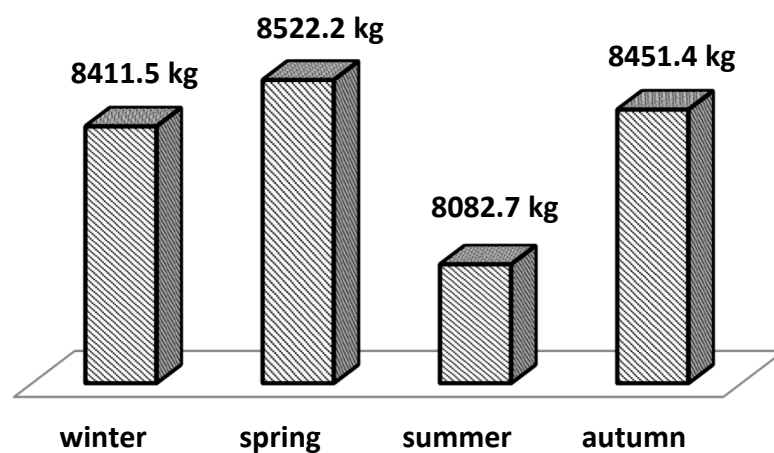
two controlled factors had a significant effect on the average milk fat and protein percentage. As was reported from the average values (Table 1), the differences by lactations number were very small for both traits.

**Table 2.** Analysis of variance for the influence of the controlled factors on the productive traits for 305-day lactation.

Sources of variation	Degrees of freedom (n – 1)	For a 305-day lactation								
		Milk yield, kg			Average fat, %			Average protein, %		
		MS	F	P	MS	F	P	MS	F	P
Total for the model	5	6345148	3.75	**	0.016	0.56-		0.108	1.50-	
Lactation number	2	8.01	4.73	**	0.033	1.15-		0.171	2.37-	
Calving season	3	4.72	2.79	*	0.006	0.20-		0.067	0.92-	
Error	280	1.69			0.029			0.072		

\*- significance at  $P < 0.05$ ; \*\*- significance at  $P < 0.01$ ; \*\*\*- significance at  $P < 0.001$ ; – no significant effect.

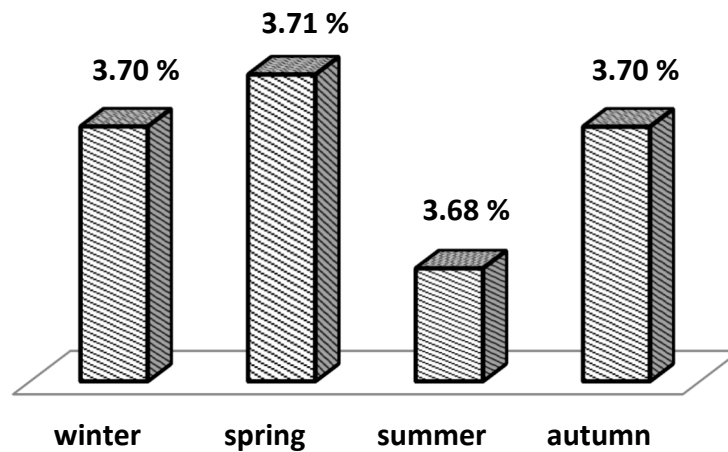
The dependence of 305-day lactation milk yield on the lactations number was reported and commented at the average values of this trait and the trend was also confirmed by the obtained LS mean values from the model. Figure 3 presents the LS mean values for milk yield depending on the calving season. The highest milk yield was reported for cows that calved in spring - 8522.2 kg, followed by those that calved in autumn and winter - 8451.4 kg and 8411.5 kg. With the lowest milk yield for standard lactation were the cows calved in the summer, respectively 8082.7 kg. The standard lactation milk yield dependency on calving seasons is shown in Figure 3.



**Figure 3.** LS mean milk yield values for 305-day lactation depending on calving season.

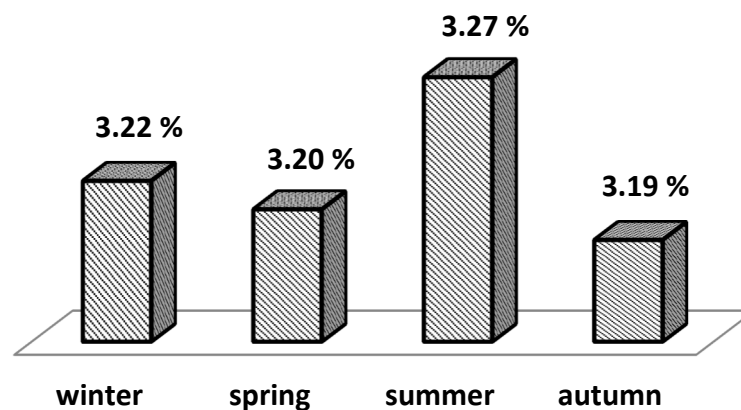
Figure 4 presents LS means from the model for the effect of calving season on fat % values for 305-day lactation.





**Figure 4.** LS mean fat % values for 305-day lactation depending on calving season.

On Figure 5, the effect of calving season on the mean milk protein percentage for 305-day lactation is presented. With the highest value it was in lactations of cows that calved in summer – 3.27%.



**Figure 5.** LS mean protein % values for 305-day lactation depending on calving season.

Table 3 presents the average values for the productive traits for a Test day by lactation number. Although with small differences, a certain tendency for higher values in later lactations for the traits milk yield and % fat in milk for a Test day was reported. In % protein for a Test day, no difference was reported depending on lactation number.

**Table 3.** Average values and variation of productive traits for a Test day by lactation number.

Lactation number	Number n	Productive traits for a Test day					
		Milk yield, kg		% fat		% protein	
		x ± Se	SD	x ± Se	SD	x ± Se	SD
First	1479	28.69±0.22	8.37	3.68±0.01	0.31	3.21±0.01	0.20
Second	890	29.25±0.33	9.85	3.70±0.01	0.27	3.21±0.01	0.19
Third	375	29.83±9.55	10.65	3.73±0.02	0.35	3.21±0.01	0.21
Total	2744	29.01±0.18	9.21	3.69±0.01	0.30	3.21±0.00	0.19

From the analysis of variance for the influence of the controlled factors on the three productive traits for a Test day, it was found that milk yield was significantly influenced by lactation number

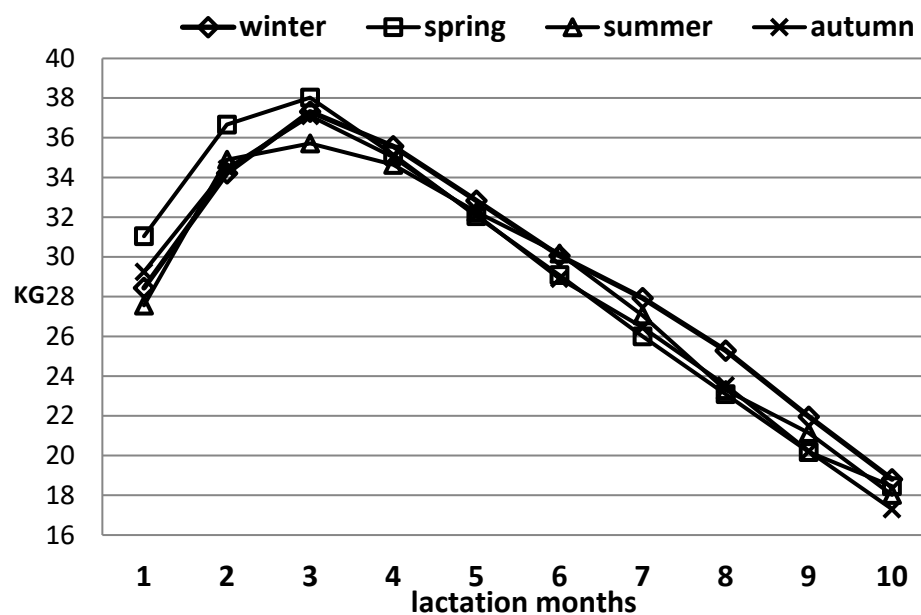
( $P < 0.05$ ), calving season ( $P < 0.05$ ) and lactation period ( $P < 0.001$ ), Table 4. On the fat percentage for a Test day, only the lactation number had a statistically significant effect ( $P < 0.01$ ). The reported effect can also be seen from the average values for lactation number in Table 3. A significant effect on protein percentage for Test day was found only by the lactation period ( $P < 0.05$ ).

**Table 4.** Analysis of variance for influence of controlled factors on milk yield for a Test day.

Sources of variation	Degrees of freedom (n - 1)	Test day Milk yield			Test day fat %			Test day protein %		
		MS	F	P	MS	F	P	MS	F	P
Total for the model	14	7083,76	144,96***		0,16	1,69-		0,06	1,87-	
Lactation number	2	196	4,01 *		0,46	5,0 **		0,01	0,4-	
Calving season	3	148	3,03 *		0,09	0,09-		0,08	0,69-	
Lactation period	9	10926	223,59***		1,12	1,3-		0,09	2,1 *	
Error	2729	40			0,09			0,03		

\*- significance at  $P < 0.05$ ; \*\*- significance at  $P < 0.01$ ; \*\*\*- significance at  $P < 0.001$ ; - no significant effect.

Figure 6 shows the lactation curves depending on the calving season. The highest milk yield for a Test day (peak of lactation) was reached by lactations that started in the spring - 38.0 kg, the same had the highest milk yield at the beginning of lactation until the 30th day - 31.0 kg. After the peak, these lactations showed a significant decline in Test day milk yield.



**Figure 6.** Lactation curves depending on calving season.

#### 4. Discussion

Taking into account that the study included only lactations from the first to third (predominantly the first), the reported productivity (Table 1) was relatively high for the breed in our country. According to the data of the Executive Agency of Selection and Reproduction in Animal Breeding, the average milk yield of the controlled cows of the Black-and-white breed (including Holsteins) in our country is 5300-5600 kg, with 3.6-3.8% fat and 3.2-3.3% protein in the milk [19].

The higher productivity of the farm was due to the constant work and selection aimed at increasing the productivity of the farmed cows, as well as satisfying them with adequate rations, consistent with their physiological needs.



According to National Institute of Meteorology and Hydrology [10] data, 2020 was the second warmest year since 1930 for our country. The average annual temperature for the country was 12.4°C and was 1.9°C higher than the climatic norm for the period 1961-1990. The average annual maximum temperature for the country was 18.7°C, which was 2.9°C above the climatic norm. The highest maximum temperature for 2020 was 40.8°C and was measured on July 31 in the town of Lyubimets, region Haskovo, which is in the same region as the studied farm.

According to a number of authors, a THI above 72 has been accepted as the threshold for inducing heat stress in the tropics [20,21], while in temperate zones, milk yield in high-producing cows may be affected at THI value lower than 60 [22,23].

As mentioned, the cows were housed in open barns (shed type, without solid walls), which means that there was almost no isolation of the animals from the external climatic conditions such as temperature and humidity. The THI values for the farm area (Figure 2) showed that the cows were exposed to conditions at risk of heat stress to varying degrees for a long period of about 5 months of the year, almost around the clock.

In a study by Penev et al. [24] the season also had a significant effect on milk yield of cows ( $P < 0.001$ ), but not on milk composition parameters, which was also confirmed by the present study (Table 2). The standard lactation milk yield was 4 to 5% lower than the reported for the other calving seasons (Figure 3). Gantner et al. [25] found that THI values in spring and summer in the three regions (eastern, Mediterranean and central) of Croatia often exceed 72, during which cows tend to develop heat stress. During the autumn and winter, when THI values are typically lower than 72, cows rarely experience heat stress. In addition, milk yields differed significantly between periods with heat stress conditions and periods without heat stress ( $p < 0.01$ ), indicating that milk yield from dairy cows in regions with different climates can be significantly affected, when THI reaches heat stress levels. Liu et al. [26] found that milk yield of Holstein cows decreased by 10% to 40% in summer compared to that in winter [27], further highlighting the impact of heat stress on milk production. According to Liu et al. [26], it is not correct for all regions and farms to determine the risk of heat stress according to whether the THI exceeds 68 or 72, but specific problems should be analyzed more concretely.

As reported (Figure 3), no significant effect of calving season and lactation number on average milk fat percentage for standard lactation was found. Between the LS means for the three calving seasons, winter, spring and autumn (Figure 4), it can be said that there was no difference in the mean milk fat percentage (of 3.70 and 3.71%), but for the summer season a lower mean value was reported, although with a small difference - 3.68%. André et al. [17] found a milk loss due to heat stress of 31.4 kg/cow/year, which is 0.32% of the farm-averaged production of 9,855 kg/cow/year. This loss is low compared to losses in the United States [28], ranging from 68 (Wyoming) to 2072 (Louisiana) kg/cow/year. In the study by Bernabucci et al. [14], however, the lowest protein values in milk were found precisely during the summer season. The authors also demonstrate a decrease in  $\alpha$ S-CN and  $\beta$ -CN proteins at the expense of unidentified proteins, which leads to a deterioration of the coagulation properties of milk milked of cows in summer. Despite the observed trends in the difference in milk fat and protein values presented in Figures 4 and 5, respectively, the analysis of variance (Table 2) showed that calving season did not significantly affect these traits. Since the cows on the farm were under the same conditions of rearing and the same composition of the ration throughout the year, the main reason can be sought in the change of climatic and, as a consequence, microclimatic conditions by seasons. The optimal climatic conditions during the spring months allow reaching a relatively high peak at the beginning of lactation, but after the third-fourth month of lactation, the summer months came with high daily temperatures and THI values (Figures 1 and 2). This leads to a steeper decline in milk yield after the peak. The high milk yield at the peak of lactation, however, provides them with the highest reported milk yield for the 305-day lactation (Figure 3). These results confirm research by other authors [31,32] who also found the highest daily milk yield in spring calving cows.

Lactations started in autumn and winter have almost identical lactation curves. At the beginning of lactation (up to the 30<sup>th</sup> day) their milk yield was 28.4 and 29.3 kg, and at the peak 37.3 and 37.1 kg. A difference appears at the end of lactation, when lactations started in winter had a slightly higher

daily milk yield than those started in autumn. The similar lactation curves of the autumn and winter calvings were also consistent with the almost identical milk yields for 305 days of lactation (Figure 1).

With the worst characteristics was the lactation curve for lactations that started in the summer. Their daily milk yield at the beginning (until the 30<sup>th</sup> day) was the lowest compared to the others - 27.6 kg, as well as in the peak, which was not clearly expressed, 35.7 kg. It can be said that after the end of the summer period the curve was slightly stabilized. In these lactations, the lowest milk yield for a 305-day lactation was also reported, Figure 3. M'Hamdi et al. [31] also found an influence of heat stress on the lactation curves of dairy cows, depending on the Temperature-humidity index values.

According to Joksimović-Todorović et al. [32], constant selection for higher milk yield leads to increased sensitivity of cows to heat stress. This is the reason why high-producing dairy cows are more sensitive to heat stress than cows with lower genetic potential for milk production [33]. Also, dairy cows in the beginning of lactation have small chance of overcoming heat stress and thus it has the strongest effect on milk production in the first 60 days of lactation. The negative energy balance in dairy cows at the beginning of lactation is further increased by creating and radiating higher amounts of heat energy during the period when the animals consume less feed [32].

This was one of the reasons why the lactation curve in summer tended to decrease compared to spring, during which the lactation curve was maintained at high levels. Additionally, when cows calve in early spring and summer, high summer temperatures coincide with lactation peak when cows are more sensitive to heat stress [21]. High-producing cows are most sensitive to the influence of heat at the beginning of lactation and in cases where the body temperature is higher than 39°C, the milk yield decreases significantly [20]. At an external temperature of 35°C, the amount of milk decreases by 33%, and at a temperature of 40°C by 50% [34].

Sacido et al. [35] and Segnalini et al. [36] indicated that in heat-stressed dairy cows, milk production decreased by between 10 and 30%, with significant reductions in fat and protein. At a temperature above 30°C, the amount of milk decreases by up to 30%, while the fat content is reduced from 3.6% to 3.2%, and the protein content from 3.34% to 3%. Also, Kadzere et al. [21] point to the fact that at a temperature of 35°C the amount of milk decreases by 33% and at 40°C by 50%. The same authors indicated that the percentage of milk fat was reduced by 39.7% and the protein by 16.9%.

West et al. [37] found that milk yield of Holstein cows decreased by 0.88 kg per unit increase in average THI and daily milk yield decreased by 0.85 kg for each degree (°C) increase in average air temperature.

High daytime temperatures during the summer period and inadequate cooling of dairy cows have a negative effect, particularly pronounced in terms of milk yield and, to some extent, milk fat. M'Hamdi et al. [31] found an effect of heat stress on both fat content and protein content of milk. In our study, however, differences in milk protein values by seasons were not found. This may be due to the selection in the studied farm as well as the level of feeding which does not change throughout the year. Given that this was a sample of all animals on the farm, the total milk losses, expressed in quantity and composition quality, can be considerable for dairy cattle farms in Bulgaria.

## 5. Conclusions

For the research period from 2017 to 2022 in the region of central southern Bulgaria, significantly high values of the maximum and average daily temperatures and THI values, representing a risk of heat stress in dairy cows were reported for a rather long period of about 5 months (from the May to September). Based on the conducted research, it can be claimed that there was a significant difference in the productivity of cows calved during the different seasons of the year. The cows with the lowest milk yield calved in the summer, and the cows with the highest calved in the spring. The influence of the season on the amount of milk affects the milk yield of the lactation curve, which also reflects on the total amount of milk for the entire lactation. The calving season affects not only the quantity but also the quality of milk produced. In the present study, such an influence was found on the

percentage of milk fat by decreasing and increasing the percentage of protein, but we must note that these differences were not significant.

**Author Contributions:** Conceptualization, M.S. and D.D.; methodology, T.P. and D.D.; software, I.M.; validation, I.M., T.P. and D.D.; formal analysis, D.D.; investigation, T.P.; resources, M.S.; data curation, D.D.; writing—original draft preparation, M.S. and T.P.; writing—review and editing, D.D. and I.M.; visualization, I.M.; supervision, T.P.; project administration, T.P.; funding acquisition, T.P. All authors have read and agreed to the published version of the manuscript.

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**Data Availability Statement:** All data presented in this study are available on request from the corresponding author.

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