

## Effect of non-genetic factors on milk yield in a Romanian Spotted dual purpose breed

R.I. Neamt<sup>1\*</sup>, L.T. Csiszter<sup>1,2</sup>, G. Saplacan<sup>1</sup>, C.V. Mihali<sup>1</sup>, A.E. Mizeranschi<sup>1,3</sup>, S.I. Saplacan<sup>4</sup>, A.S. Anton<sup>1</sup>, D.E. Ilie<sup>1</sup>

<sup>1</sup>Research Department, Research and Development Station for Bovine, 310059, Arad, Calea Bodrogului No. 32, Romania; ORCID: 0000-0002-5812-8870; 0000-0001-5651-3903; 0000-0002-0464-3199

<sup>2</sup>Department of Animal Production Engineering, University of Life Sciences "King Michael I of Romania" from Timisoara, 300645, Timisoara, Calea Aradului No. 119, Romania; ORCID: 0000-0003-1896-1515

<sup>3</sup>Institute for Advanced Environmental Research, West University of Timișoara, 300223, Bd. V. Pârvan nr. 4, Timișoara, Romania; ORCID:0000-0002-1168-6285

<sup>4</sup>Aurel Vlaicu University of Arad, Faculty of Economic Science, 310130, Arad, Bulevardul Revolutiei No. 77, Romania; ORCID: 0000-0002-6724-1322

(Submitted 29 February 2024; Accepted 1 May 2024; Published 29 May 2024)

Copyright resides with the authors in terms of the Creative Commons Attribution 4.0 South African Licence.

See: <http://creativecommons.org/licenses/by/4.0/za>

Condition of use: The user may copy, distribute, transmit and adapt the work, but must recognise the authors and the South African Journal of Animal Science.

### Abstract

The objective of the current study was to establish the possible impact of several non-genetic factors on milk yield. Data from 1120 lactations were collected between 2019 and 2022 regarding cows' milk yield (MY) and 305-d MY, under the influence of year, calving season, lactation order, type of calving, and calving ease. MY proved to be influenced by the season of calving, being higher in summer ( $5745.5 \pm 156.24$  kg) compared to spring ( $5448.9 \pm 108.1$  kg) or winter ( $5327.35 \pm 200.6$  kg). A marked influence of the rank of lactation and type of calving were found. The maximum MY was found in the 4<sup>th</sup> lactation ( $6211.8 \pm 210.18$  kg), whereas the lowest MY was recorded in the 6<sup>th</sup> lactation ( $5535.8 \pm 116.6$  kg). No substantial effects for year and calving ease were recorded ( $6003.71 \pm 174.23$  vs.  $5712.14 \pm 163.9$  for eutocic vs. dystocic births). Season, lactation order, and twinning substantially influenced 305-d MY. In conclusion, MY and 305-d MY proved to be highly dependent on environmental and management factors. By ensuring good management practices *pre-* and *post-partum*, the farmers can maintain the milk production at a high level.

**Keywords:** dairy production, dual purpose cattle, influential factors, milk yield

\*Corresponding author's e-mail: [neamtr@yahoo.com](mailto:neamtr@yahoo.com)

### Introduction

The Romanian Spotted breed is the result of *Simmental* and local *Sură de Stepă* crossbreeding. The process began in 1862 and the new breed was approved in 1959. The new breed was immediately subjected to a successive breeding improvement process aimed to enhance the morphological, productive, and reproductive traits. Nowadays, the Romanian Spotted is the most prevalent breed in the country, accounting for 40% of the herds. Worldwide, the total numbers are estimated between 40 and 60 million *Simmental* cattle, with more than half in Europe (Huyghe *et al.*, 2014).

The breed's outstanding adaptability compared to specialized breeds, high degree of flexibility according to rearing systems, high resistance to environmental conditions, and dual nature of production (milk and meat), explain the higher prevalence in livestock. The breed adapts easily to the most varied conditions, from rural small-holder to large extensive ranching operations. Due to the

irregular inbreeding programs, random crosses, and the influence of numerous non-genetic factors, a substantial degree of trait variability was recorded, exacerbated by variability in the rearing systems. The cows' performances were exposed to a wide range of influential factors due to the variability in rearing areas and specific conditions.

Intensive inbreeding in milk producing cows has been recorded in the last five to seven decades (Oltencu & Broom, 2010). The results related to milk yield were found to be negatively correlated with other parameters, such as reproduction or well-being. The influence of the genetic structure on milk production is well known and can be counteracted with the specific means of livestock breeding. Milk yield is also influenced by random non-genetic factors, which are difficult to identify and control. The impact of random, non-genetic factors on the milk yield has received more attention recently, due to the variety of cow breeding systems, the diversity of environmental conditions, the intensification of production processes, and the increasingly noticeable changes in climate (Alpan & Akdoy, 2015; Kamal *et al.*, 2020).

Previous studies conducted in respect of milk yield variability have generated contradictory results, with documented 305-d milk yields ranging from 2000 kg to 5700 kg (Gorgulu, 2011; Czerniawska *et al.*, 2012). Contradictory tendencies were related to influential factors on milk production. Year, season of calving, rank of lactation, type of calving, and calving ease were identified as factors with a marked impact on the milk yield (Petrović *et al.*, 2015), whereas other results conclude otherwise (Özkan & Güneş, 2011). The morpho-productive type of the breeds included in studies and the well-known higher sensitivity of the specialized breeds could both be considered as explanations for the inconsistent results found in the literature. The breeds adaptation to the environment, as well as the rearing system, could provide a source of contradictory outcomes, given the well-established and strong association between these parameters.

Knowing the impact of non-genetic factors and the possibilities of mitigating or even avoiding their negative effects on milk production in dual-purpose cows facilitates the adaptation of rearing systems with a positive impact on the efficiency of farms. The aim of the current research was to investigate the dependence of 305-d MY on year, calving season, range of lactation, type of birth, and calving difficulty in Romanian Spotted cows reared under a temperate, European climate.

## Material and Methods

All procedures associated to the current research were approved by the Scientific and Ethics Committee of the Research and Development Station for Bovine Arad belonging to the Academy for Agricultural and Forestry Sciences. The research activities were performed in accordance with the European Union's Directive for animal experimentation (Directive 2010/63/EU).

The study was conducted at the Research and Development Station for Bovine Arad, Romania, located in the western plain of the country. The research station's farm is considered as a mixed crop–livestock farm. The climate of this area is continental–moderate, with weak Mediterranean influences. An average temperature of 21 °C in summer and -1 °C in winter is recorded. The multiannual average amount of precipitation is 582 mm. The highest amounts of precipitation are recorded in June (88.6 mm); in general the warm season records 58% of the total amount as a direct consequence of the dominance of the winds from the west. A secondary maximum is recorded in the autumn months (24% of the average annual quantity). The area is subjected all year round to the advent of humid air from the west and its ascent in contact with the Apuseni mountains, hence the explanation of the high frequency of days with 120 mm precipitation.

The animals were reared in a semi-intensive system. Cows included in the current study were managed under a loose system all year round and were between lactations one and six. Cows were kept on straw bedding, with a space allowance of 9 m<sup>2</sup> and free access to forage, water, and paddocks. They received a daily feed ration consisting of 40 kg of green fodder (lucerne), 15 kg maize silage, 8 kg of lucerne hay, and 6 kg of concentrates starting from spring until late autumn and a ration consisting of 40 kg of maize silage, 8 kg of lucerne hay, and 6 kg of concentrates during winter. Cows were fed three times a day. They were housed in groups of 70 head/paddock. Cows were milked twice per day (starting at 05:00 and 17:00) in a herringbone milking parlour (2 × 14 units).

All cows were included in the Official Performance and Recording Scheme. In order to collect data regarding milk yield, the parlour was equipped with Afimilk 3.076 A-DU software. All cows were fitted with Afitag pedometers for production, reproductive trait, and specific disease detection. The current study comprised 280 cows that survived the whole four-year trial period. Multiple data sets were collected between January 2019–December 2022 from 1120 lactations associated with the 280 cows,

resulting in 6720 records (280 cows × 4 years × 6 traits) containing cows' milk yield (MY) and 305-d MY under the influence of year, calving season, lactation order, type of calving, and calving ease. Considering the successive lactations recorded, the data regarding the milk yield were corrected for mature equivalents with indices related to 305-d MY, calving season, and cow parity.

Multiple data were collected and analysed for each cow individually and annually related to the milk yield in order to establish its dynamics according to the non-genetic influential factors included in the current study. Data related to the cows' identification and milk quantity were recorded in the milking parlour using the AfiTag pedometer system and were correlated to the Official Performance and Recording Scheme. Data regarding reproductive activity were recorded from the field by the farm technicians.

Cows having too short lactations (less than 205 d) or prolonged lactations (more than 400 d) were eliminated from the study, whereas lactations of 205–400 d were adjusted to 305-d MY using correction indices (Çilek & Tekin, 2006). Cows that had abortions or those with very severe dystocia (4 and 5 on a scale of 1 to 5) and whose calves succumbed in the first 72 hours *postpartum* were excluded from the study based on an obvious negative influence of dystocia on the dams' well-being and implicitly on milk production. Primiparous cows and those with parity greater than 6 were also excluded from the study. The average difficulty in birth was 2.59 on a scale of 1 to 5 for dystocia score. Average body condition score (BCS) was 3.48 for -21 days and 3.33 for +21 days after calving. All data were recorded in successive years.

In order to clean data for human recording errors and outliers, the Grubbs' test (Grubbs, 1969) was performed in a univariate data set characterised by a normal distribution:

$$G=(\bar{y}-y_{\min})/s \qquad G=(\bar{y}-y_{\max})/s \qquad (1)$$

where  $\bar{y}$  = sample mean;  $s$  = standard deviation;  $y_{\min}$  = minimum value;  $y_{\max}$  = maximum value.

In the current data set, more than one outlier was recorded and the Tietjen–Moore test (Tietjen & Moore 1972) being applied in order to reject them:

$$Lk=\frac{\sum_{i=1}^{n-k}(y_i-y_k)^2}{\sum_{i=1}^n(y_i-\bar{y})^2} \qquad (2)$$

Where  $k$  = exactly  $k$  outliers in the data set;  $n$  = number of data points sorted from smallest to largest;  $y_i$  = the  $i$ th largest data value;  $\bar{y}$  = mean of the full sample;  $\bar{y}_k$  = sample mean with largest  $k$  point deleted.

The non-genetic influential factors were included in the following categories: the year of calving was grouped in four categories, 1<sup>st</sup>-2019, 2<sup>nd</sup>-2020, 3<sup>th</sup>-2021, 4<sup>th</sup>-2022; the season of calving was 1<sup>st</sup>-spring (from March to May), 2<sup>nd</sup>-summer (from June to August), 3<sup>rd</sup>-fall (from September to November), 4<sup>th</sup>-winter (from December to February); rank of lactation was grouped into five classes: L2, L3, L4, L5, and L6; type of calving was represented by 1<sup>st</sup>-single calving and 2<sup>nd</sup>-twin calving; calving ease was represented by 1<sup>st</sup>-eutocia and 2<sup>nd</sup>-dystocia.

Comparison between milk yields related to influential factors was assessed using one –way ANOVA. Differences were tested using Tukey test.

$$Y_a = \mu + Q_a + e_a \qquad (3)$$

$$Y_a = \mu + S_b + e_b \qquad (4)$$

$$Y_a = \mu + L_c + e_c \qquad (5)$$

$$Y_a = \mu + T_d + e_d \qquad (6)$$

$$Y_a = \mu + C_f + e_e \qquad (7)$$

where  $\mu$  = the mean milk yield,  $Q_a$  = the effect of calving year,  $S_b$  = the effect of the calving season,  $L_c$  = the rank of lactation,  $T_d$  = the effect of type of calving,  $C_f$  = the effect of calving ease, and  $e_{a-e}$  = the random error.

A univariate test of significance for comparison based on Duncan's multiple-range tests was performed in order to establish the multiple comparisons within the subgroups of influential factors (Duncan, 1955).

$$R_p = Q_p(S^2_p/n)^{1/2} \qquad (8)$$

where  $Q_p$  depends on the number of means being compared at one time, denoted  $p$ , and degrees of freedom  $k(n-1)$ ,  $n$  being the number of individuals.

The analysed data were expressed as least square means and standard error of the mean. All the statistical processes were carried out using the software package, Statistica (Hill & Lewicki, 2007). Decisions about the acceptance or rejection of statistical hypotheses have been made at the 0.05 level of significance. Phenotypic correlations between milk yield and categories of non-genetic influential factors were estimated using the analysis of variance (Grosu & Oltenacu 2005). To determine how various influential factors affect the milk yield, this would be considered as the dependent variable and the influential factors included in the study were the independent variables of interest.

## Results and Discussion

Table 1 summarizes the findings of the current study. The mean MY and 305-d MY was 6108.72  $\pm$  201.3 and 5906.6  $\pm$  216.63 kg, respectively. The herd's average lactation number was 3.92. A total of 123 twin births (10.98% of the total) were recorded. Dystocia occurred in 17.94% births (201 calvings).

**Table 1.** Least square means ( $\pm$  SEM) significance and comparison related to the influence of studied factors on MY and 305-d MY

| INFLUENTIAL FACTORS                | N           | MY (kg)                               | 305-d MY (kg)                         |
|------------------------------------|-------------|---------------------------------------|---------------------------------------|
| <b>COHORT</b>                      | <b>1120</b> | <b>6108.72 <math>\pm</math> 201.3</b> | <b>5906.6 <math>\pm</math> 216.63</b> |
| <b>YEAR</b>                        |             |                                       |                                       |
| <b>1<sup>st</sup> (2019)</b>       | 280         | 5472.5 $\pm$ 192.3 <sup>a</sup>       | 5512.36 $\pm$ 184.17 <sup>a</sup>     |
| <b>2<sup>nd</sup> (2020)</b>       | 280         | 5527.54 $\pm$ 181.8 <sup>a</sup>      | 5626.9 $\pm$ 156.9 <sup>a</sup>       |
| <b>3<sup>rd</sup> (2021)</b>       | 280         | 5613.54 $\pm$ 243.3 <sup>a</sup>      | 5644.11 $\pm$ 181.14 <sup>a</sup>     |
| <b>4<sup>th</sup> (2022)</b>       | 280         | 5675.4 $\pm$ 116.55 <sup>a</sup>      | 5685.43 $\pm$ 139.12 <sup>a</sup>     |
| <b>SEASON OF CALVING</b>           |             |                                       |                                       |
| <b>Spring (March–May)</b>          | 301         | 5448.9 $\pm$ 108.1 <sup>a</sup>       | 5202.19 $\pm$ 151.12 <sup>a</sup>     |
| <b>Summer (June–August)</b>        | 294         | 5745.5 $\pm$ 156.24 <sup>b</sup>      | 5648.2 $\pm$ 166.3 <sup>b</sup>       |
| <b>Autumn (September–November)</b> | 220         | 5704.9 $\pm$ 181.31 <sup>b</sup>      | 5607.12 $\pm$ 142.8 <sup>b</sup>      |
| <b>Winter (December–February)</b>  | 305         | 5327.35 $\pm$ 200.6 <sup>a</sup>      | 5274.3 $\pm$ 169.91 <sup>a</sup>      |
| <b>RANK OF LACTATION</b>           |             |                                       |                                       |
| <b>2<sup>nd</sup></b>              | 184         | 5838.4 $\pm$ 183.27 <sup>a</sup>      | 5631.9 $\pm$ 144.6 <sup>a</sup>       |
| <b>3<sup>rd</sup></b>              | 212         | 5872.24 $\pm$ 177.61 <sup>a</sup>     | 5712.31 $\pm$ 181.22 <sup>a</sup>     |
| <b>4<sup>th</sup></b>              | 287         | 6211.8 $\pm$ 210.18 <sup>b</sup>      | 5913.3 $\pm$ 177.26 <sup>b</sup>      |
| <b>5<sup>th</sup></b>              | 198         | 6027.21 $\pm$ 203.44 <sup>b</sup>     | 5909.25 $\pm$ 136.8 <sup>b</sup>      |
| <b>6<sup>th</sup></b>              | 239         | 5535.8 $\pm$ 116.6 <sup>c</sup>       | 5487.46 $\pm$ 191.24 <sup>c</sup>     |
| <b>TYPE OF CALVING</b>             |             |                                       |                                       |
| <b>Single</b>                      | 997         | 5873.26 $\pm$ 187.93 <sup>a</sup>     | 5779.66 $\pm$ 181.12 <sup>a</sup>     |
| <b>Twin</b>                        | 71          | 5422.9 $\pm$ 199.7 <sup>b</sup>       | 5399.2 $\pm$ 111.17 <sup>b</sup>      |
| <b>CALVING EASE</b>                |             |                                       |                                       |
| <b>Eutocia</b>                     | 919         | 6003.71 $\pm$ 174.23 <sup>a</sup>     | 5974.3 $\pm$ 201.17 <sup>a</sup>      |
| <b>Dystocia</b>                    | 201         | 5712.14 $\pm$ 163.9 <sup>a</sup>      | 5706.34 $\pm$ 193.17 <sup>a</sup>     |

<sup>a,b,c</sup> Column means with different superscripts differ significantly at  $P < 0.05$

The discrepancies related to MY and 305-d MY based on studied years were found to be non-significant from a statistical point of view. An upward tendency was recorded for 305-d MY over time, with a total gain of 173.07 kg of milk corresponding to an annual average of 57.69 kg.

The differences in milk production can be attributed to either farm breeding programs or environmental conditions and applied management (forage ratio, forage quality, rearing conditions, hygiene, health and welfare condition). Bolacali (2018) highlighted the discrepancies in milk production related to the effects of herd breeding strategies (Bolacali & Öztürk, 2018). The effects of breeding programs were avoided in the current investigation because the same cows were considered ( $n = 280/\text{year}$ ). Thus, the observed non-significant differences were undoubtedly caused by non-genetic causes and the implemented management practices.

Environmental patterns, temperature–humidity index, and heat stress (THI and HS), more or less variable in time, are able to substantially affect MY (Nardone *et al.*, 2010). This is accomplished directly and indirectly. Directly, THI and HS may affect MY by lowering the availability of forages and decreasing their quality. These environmental variables can impair the cows' appetite, health, comfort, and welfare. Marked differences in terms of milk production, in both the sequence of years or within the same year, were obviously observed under heat stress conditions. Prolonged heat stress affects both milk production and lactation length and, implicitly, 305-d MY (Rodriguez-Venegas *et al.*, 2023). Our study period, characterized by fairly constant annual temperatures of 10.2–10.8 °C and a rainfall pattern of 553–597 mm, typical of a continental–moderate climate, allowed for rather favourable conditions, and the quantity and quality of available forages was correlated with the needs of the cows. In the sequence of study years, the lack of change in the structure of the rations and the constant quality and quantity of the forage represented crucial variables in maintaining productive stability in the herd. Rearing, hygiene, and health conditions were able to meet the cows' demands, creating conditions for the outsourcing of genetic potential in terms of MY. Naceur (2012) recorded substantial differences related to the study year. These differences were attributed to changes in herd size, age of the cows, and management practices combined with the environmental conditions (Naceur *et al.*, 2012).

The season of calving was found to be a factor in both MY and 305-d MY ( $P < 0.01$ ). The dynamics of milk production varied in relation to environmental and management patterns and were influenced at two levels: the structure of the fodder ration, the THI and HS, through the impact that this stress has on the establishment of gestation and, implicitly, on the length of lactation. A ration structure based on silage fodder associated with winter and early spring did not allow for a high MY as compared to a ration structure based on green, lactogenic fodder, which is associated with summer and autumn ( $P < 0.01$ ), a correlation that was found in other studies (Abdul *et al.*, 2016; Emal *et al.*, 2021). For 305-d MY, similar differences were recorded, but especially the fact that the hot summer months led to the protraction of lactation and a delay in gestation.

The daylength influenced behavioural specificity, with cows spending additional time consuming feed, which increased MY and 305-d MY. Thus, previous studies conducted by different researchers such as Miller (1999) or Porter (2002) highlighted that an increase in natural light by 3 h could increase the MY (Miller *et al.*, 1999; Porter & Luhman, 2002). Dahl (2000) reported an increase of 2–2.5 kg/d in MY in the months with an extended amount of natural light (Dahl *et al.*, 2000). The current study validates the previous findings. In the current study, there were no changes in MY ( $P > 0.05$ ) between summer and autumn or between winter and spring ( $P > 0.05$ ). Lactation length was influenced by seasonal patterns (temperature, amount of natural light) and 305-d MY was similar ( $P > 0.05$ ), except for 305-d MY in winter, which was decreased ( $P < 0.01$ ) due to forage structure and the correlations between management and environmental traits. Most of seasonal factors could be adjusted and enhanced, whereas others could be partially or completely eliminated (environmental issues).

In the current study, the primiparous cows were eliminated because their udder did not reach complete morphological and functional development. Regarding lactation order, two complementary analyses including MY and lactation length were performed. Lactation order exerts a marked influence on the 305-d MY. A productive Gaussian trend was recorded when analysing the dynamics of MY related to lactation order; results were confirmed by previous studies (Ahmad *et al.*, 2011; Ayalew *et al.*, 2015).

Despite the non-statistical differences recorded, MY exhibited an increased tendency between the second and third lactations. The peak of MY was recorded in the fourth rank, increased compared to the second, or the fifth and sixth ranks ( $P < 0.01$ ). MY's downward slope began in the fifth rank and got more noticeable in the sixth ( $P < 0.05$ ). The productive differences between ranks 2 and 3 were reduced (0.58%) and increased in time, resulting in a total increase of 6.38% in lactation 4 when compared to lactation 2. Furthermore, the upward curve of milk production was extended till the fourth lactation, which is normal for the Simmental dual-purpose breed, with a slower growth process, compared to dairy breeds. Mayakrishnan (2017) found that for the Holstein breed, there was a 1.9% increase in MY between ranks 2 and 3, with the highest MY attributed to the third rank, followed by an abrupt 16% decline after that (Mayakrishnan *et al.*, 2017). This dynamic may vary considerably as a result of many environmental influences. Mellado (2011) reports a constant increase in MY of more than 25% between the second and sixth rank, with the highest MY attributed to the fifth rank (Mellado *et al.*, 2011). Despite the upward tendency of MY being universally agreed upon and observed in all of the investigated breeds, its intensity varies greatly depending on environmental variables (Zukiewicz *et al.*, 2012). Habibi (2021) observed an uninterrupted increase between the 1<sup>st</sup> and the 3<sup>rd</sup> rank, although, under the effect of thermal heat stress, the highest MY is attribute to the 3<sup>rd</sup> rank, followed by an early

productive drop (Habibi *et al.*, 2021). In parallel, the 305-d MY was analysed, and somewhat identical dynamics as MY were recorded. This parameter's defining feature was the classification of production based on lactation order, with computed differences substantially lower. Thus, the lactation length proved to be longer in the second and third ranks, with milk production being similar ( $P > 0.05$ ). The productive decline recorded in the fourth rank was much smoother, with the length of lactation approaching the normal length of 305 d. A lactation length of 305 d resulted in lower milk production in the sixth rank versus the fourth and the fifth ranks ( $P < 0.01$ ), although similar to the second and the third ranks ( $P > 0.05$ ). We may conclude that lactation number influences MY. Results obtained by Bolacali (2018) reported no statistical differences regarding 305-d MY (Bolacali & Öztürk 2018).

The research regarding 305-d MY according to the lactation number does not have a common denominator, presumably due to the multiple potential influencing factors (breed, productive level, environment, age, or efficiency of reproductive activity). Several studies found marked differences according to lactation number (Andrýsek *et al.*, 2014); others recorded no significant results in this respect (Pantelić *et al.*, 2013). In the current study, the 305-d MY increased slightly between the second and the fourth ranks, but differences were not significant (+1.43%). Subsequently, a slightly decline in 305-d MY was registered until the fifth rank (-0.06%,  $P > 0.05$ ), which was amplified in the sixth (-7.2%,  $P < 0.05$ ). Previous studies conducted by Cilek (2005) and Pantelic (2010) found a continuous increase in 305-d MY up to the age of 6 y (Çilek & Tekin, 2005; Pantelić *et al.*, 2010). This tendency is not found by Bolacali (2018), who recorded a productive increase in the first two ranks, followed by a prolonged downward trend up to the seventh rank (Bolacali & Öztürk, 2018). The herd structure may shed light on the upward trend in the 305-d MY in the sequence of productive cycles. In our study, the most productive cows were associated with the 2<sup>nd</sup>, 3<sup>rd</sup>, and 4<sup>th</sup> ranks (683 out of 1120 cows, representing ~61%).

The average incidence of twinning in the current study was 6.3% ( $n = 71$ ). This incidence proved to be similar to those recorded by other studies, which found values of 1–8% (Fernando *et al.*, 2008; Victor & Paul, 2021). This incidence could be based on a better metabolic balance of this dual purpose breed (Romanian Spotted Simmental type). In general, the studies regarding the occurrence of twinning have been aimed at breeds specialized in milk or meat production. However, the current incidence of twinning seems to be substantially lower compared to the 30% found in multiparous cows (Garcia & López, 2018). A difference was recorded for both MY (+8%,  $P < 0.05$ ) and 305-d MY (+6.6%,  $P < 0.05$ ) related to type of calving. Twinning proved to decrease the MY for both total and 305-d lactation, confirming previous studies (Nielen *et al.*, 1989; Bicalho *et al.*, 2008; Hossein-Zadeh, 2010). The issue of twinning remains unclear, both in terms of the consequences on farm efficiency and the relationship with MY. The correlation or association between twinning and MY has not been thoroughly explained; the results are still contradictory. The influence of twinning on MY is reflected in the dynamics of lactation. Thus, MY decreases substantially in the first 100 d of lactation with twin births compared to singular births, reducing the entire production associated with the current lactation of dams. This indicates the presence of metabolic issues found in twinning during the early period of lactation compared to singular birth. In this respect, Mostafa (2009) found a marked decrease in milk yield of 487 kg per lactation for cows with twin births. Özden (2010) recorded a reduced lactation length of 2–10 d in cows producing twin calves, with a negative impact on MY compared to single births (Özden 2010). Twinning constitutes a better possibility to increase efficiency in beef, based on more weaned calves (+70%) and a >48% increase in total weaning weight (Echternkamp & Gregory, 2002). In this respect, the lower incidence of only 1–2% of twinning documented in beef breeds needs to be increased in order to produce more weaned calves from a single dam (Jolanta & Zbigniew, 2002).

Higher milk production is associated with a lower level of progesterone (Lopez *et al.*, 2005; Sawa, *et al.*, 2015). The decrease in progesterone levels is attributed to the acceleration of hepatic metabolism based on an increased blood flow caused by a high feed intake (Sangsritavong *et al.*, 2002). This fact generates a negative response on the hypothalamic axis, materialized by increasing the concentration of FSH and LH and implicitly by stimulating multiple ovulation among the codominant follicles that end up multiplying (Stevenson *et al.*, 2007). Previous studies have recorded an accessional tendency of twinning incidence of 10–25%, in time (Fricke, 2001; Lopez *et al.*, 2017). A continuous increase in twinning has been recorded over the years. Kinsel *et al.* (1998) calculated a growth rate of ~0.1% per year in the interval 1983–1993. An increased incidence of twinning was recorded in multiparous compared with primiparous cows (Silva del Río *et al.*, 2007). Financially, the economic losses of the farms due to twinning vary within relatively wide limits, being between \$59 and \$225 depending on numerous factors (Eddy *et al.*, 1991; Gaspárdy *et al.*, 2018; Mur-Novales *et al.*, 2018; Fodor *et al.*, 2019). Due to the economic losses caused in dairy farms, the increase in the incidence of twinning represents a concerns for dairy farmers with respect to reducing or at least limiting it (Caraviello *et al.*,

2006). However, the possibilities of selection and control of twinning remain low due to the minimal heritability (0.01–0.06) and a low repeatability (0.04–0.28) (Moioli, *et al.*, 2017; Beth & Brian, 2018).

Hormonal strategies used to carry out reproductive activity can represent an efficient method of decreasing the incidence of twinning. By increasing the level of progesterone in hormonal treatments, the incidence of twinning is reduced and fertility is increased, which are desirable effects in highly productive dairy cows (Carvalho *et al.*, 2019).

The cows included in the present study recorded increased average productions compared to the average of the breed (>6000 vs. 4000 kg milk); this may have led to an increased incidence of twinning. However, by analysing the dynamics of milk production, a tendency towards a linear increase was recorded over the study years. According to the lactation order, MY followed an upward trend until the 4th lactation, after which it started to decrease, the dynamics being physiologically normal. The increase in MY over time could be attributed to reaching the morpho-physiological maturity of the cows and was also recorded in other previous studies conducted by Pantelić *et al.* (2010) and Petrović *et al.* (2015). Increased milk production may increase the twinning incidence, therefore shortening the lactation length and has a detrimental influence on 305-d MY in dams with twin deliveries.

Even if twinning appears to be beneficial at first sight, it involves plenty of issues that target multiple levels. Some issues associated with twinning include infertility, higher incidence of morbidity and mortality in calves, lower reproductive performance in cows caused by dystocia and retained placenta, and a decrease in gestation and lactation length, which are issues that should be avoided in dairy herds (Hosseini-Zadeh *et al.*, 2008; Mostafa, 2009; Özden, 2010).

Dystocia exerts a marked effect on animal welfare due to the reduced reproductive indices, health conditions, and production. The impact of the severity of dystocia on productive parameters occurs in both the current lactation and in subsequent productive cycles. The impacts of difficult calving are highly persistent and they have the potential to decrease production in subsequent lactations as well (Eaglen *et al.*, 2011). The effects of dystocia are felt in terms of milk quality. Previous studies have recorded decreased amounts of fat, protein (Rajala & Gröhn, 1998), and lactose in cows with dystocia (Miglior *et al.*, 2006). The reduced productivity is related to injuries and inflammation that occurs due to the dystocia (Pohl *et al.*, 2015), as well as the reduction in feed and water intake after calving (Proudfoot *et al.*, 2009). In this respect, a *pre-partum* treatment with non-steroidal anti-inflammatory drugs could reduce the effects of the inflammation caused by the dystocic calving and could improve the MY in the subsequent lactation (Shock *et al.*, 2018; Gladden *et al.*, 2021). Behavioural changes, such as reduced appetite and prolonged time allocated to standing and self-grooming, detriment food and water intake (Barrier *et al.*, 2012).

The incidence of dystocia in the current study was 17.94% (n = 201), with 8.12% ease assistance (score 1–2), 6.42% moderate assistance (score 3), and 3.4% with intense assistance (score 4–5), the average score being 2.59. This incidence proved to be similar to the value recorded by Mee (2008) of 1.5–22% worldwide. Even for births that required ease or moderate assistance (scores 1–3), the results obtained in the current study were higher than those from other studies of 8.2–16% (Meyer *et al.*, 2001; Wall *et al.*, 2010; Atashi *et al.*, 2012). The cows with dystocia delivered slightly less milk compared to ones with eutocia but no statistical differences were recorded in the current study related to MY (291.57 kg milk) and 305-d MY (267.96 kg milk). These findings were similar to those obtained by Bicalho *et al.* (2008) and slightly lower than those recorded by Berry *et al.* (2007) of -300 to -700 kg milk or Eaglen *et al.* (2011) of approximately -250 kg milk. In general, the literature provides conflicting results regarding the influence of dystocia on MY, possibly due to the influence of a large number of influential factors and low correlations between them. Moore *et al.* (1991) calculated low or medium correlation coefficients of -0.11 and -0.3 for these traits.

The lactation curve dynamics could serve to clarify why cows that experienced dystocia produced less milk. Cows with dystocia frequently produce less due to the productive drop that occurs during the first 100 d of lactation. A 2 kg milk/day loss was found for the first 100 days of lactation (Eaglen *et al.*, 2011). Appropriate management, both *pre-* and *post-partum*, can mitigate this decline.

The occurrence of dystocia, the degree of difficulty in calving, and the subsequent impact on MY could be impacted by animal-dependent factors such as season of calving, lactation order, dam body condition score. A low incidence was recorded in cows with more than three lactations that had already reached morphological maturity. There is a dystocia–placental retention–stillbirth combination which is related to a reduction in MY (Fourichon *et al.*, 1999).

To better comprehend the non-significant differences in MY between cows with eutocia and dystocia from the current study despite the relatively high incidence of dystocia, we considered the calving season, body condition score, lactation order, and dystocia score as influential factors. In the current study, the average dystocia score was 2.59, generated by 80.6% of low and medium dystocia

scores of 1–3. This fact may substantially reduce the harm caused to the dams, as well as the detrimental impacts on the MY during the first 100 days of lactation, with a positive effect on the 305-d MY. The analysis performed related to the BCS highlighted a balance in terms of the average score of 3.48 *pre-partum* and 3.33 *post-partum* ( $P > 0.05$ ). An optimal *pre-partum* BCS of 3.25–3.5 (Gheise *et al.*, 2017) with appropriate *post-partum* management can substantially reduce the decrease in MY by maintaining energy balance and fat mobilisation (Yujie *et al.*, 2019). In this respect, we can conclude that the balance in BCS in the cows in the current study was integral in maintaining MY and 305-d MY at comparable levels in eutocic and dystocic deliveries.

It is well known that the incidence of dystocia is increased in primiparous cows (Gaafar *et al.*, 2011; De Amicis *et al.*, 2017). The higher frequency of dystocia is mostly due to failure to reach maturity in terms of both morphological and functional traits in primiparous animals. Multiparous cows that have reached morphological maturity and have sufficient energy reserves can quickly rebalance metabolically, having the resources to maintain high milk production in an instance of dystocia, thus minimizing the difference in MY according to calving ease. In the current study, the most productive cows (64.64%) were over the 3<sup>rd</sup> lactation (lactations 4–6), and 35.36% were in the 2<sup>nd</sup> and 3<sup>rd</sup> lactations; primiparous cows were omitted. The majority of cows were assigned to higher lactation numbers, thus explaining why there were no statistical differences in MY and 305-d MY between cows that experienced eutocia and dystocia. In a recent study, Steven *et al.* (2023) recorded an impact of mixed season of calving and calving ease on MY. Those cows that experienced dystocia in autumn produced less milk than their counterparts. Analyses performed in this respect found differences ( $P < 0.01$ ) in the percentage of calves born in autumn (19.6%), compared to spring, summer, and winter (26.87%, 26.25%, and 27.2%, respectively). This relationship presents a plausible explanation for similarities in MY and 305-d MY according to calving ease.

## Conclusions

The results of the current study indicate that milk production is strongly influenced by the season, lactation order, and twin births. By avoiding extreme situations such as a lack of or poor quality fodder; extreme environmental conditions, particularly in terms of temperature and humidity; and struggling health and welfare, milk production can be maintained at levels that correspond to genetic potential through managerial practices that fulfil the cows' requirements.

## Acknowledgments

This work was supported by the Research and Development Station for Bovine Arad under a self-funded Grant, number 5173/2021.

## Author contributions

The idea for the article was conceived by R.I. Neamt. The experiments were designed by R.I. Neamt, Ghe. Saplacan, and L.T. Czisster. The experiments and data collection were performed by C.V. Mihali, A.S. Anton, and D.E. Ilie. The data were analysed by S.I. Saplacan and A.E. Mizeranschi. The article was written by R.I. Neamt, L.T. Czisster, and Ghe. Saplacan.

**Conflict of Interest Statement:** The authors declare that the current research was performed without any special interest (commercial or financial).

## References

- Abdul, B., Huque, K.S., Nathu, R.S., Mukter, H., Nazrul, I., 2016. Effect of season, genotype and lactation on milk yield and composition of local and crossbred dairy cows reared under different feed base region, Bangla. J. of Livestock Res. 19(1-2), 50. DOI: 10.3329/bjlr.v19i1-2.26427.
- Ahmad, S., Hossain, F.M.A., Islam, N., 2011. Effect of lactation number and different stage of lactation on milk yield of indigenous and crossbred cows in Bangladesh. Int. J. Nat. Sci. 1(1), 31–34.
- Alpan, O., Aksoy, A.R., 2015. Cattle breeding and fattening (in Turkish). In: Turkey: 7<sup>th</sup> ed. Favori Offset Press. Istanbul. pp168–182.
- Andrýšek, J., Chládek, G., Javorová, J., Milena, V., Milan, V., Daniel, Falta., 2014. The effect of milk yield on conversion coefficients between lactations of Czech Fleckvieh cows. In: Mendel Net Conference. Czech. Proceeding. Czech Republic: Brno. pp 34–137.
- Atashi, H., Abdolmohammadi, A., Dadpasand, M., Asaadi, A., 2012. Prevalence, risk factors and consequent effect of dystocia in Holstein dairy cows in Iran. Asian-Australas. J. Anim. Sci. 25(4), 447–451. <https://doi.org/10.5713/ajas.2011.11303>.
- Ayalew, W., Aliy, M., Neguessie, E., 2015. Milk production performance of Holstein Friesian dairy cows at Holetta Bull dam farm, Ethiopia. Livest. Res. Dev. 27(9).
- Barrier, A.C., Ruelle, R., Haskell, M.J., Dwyer, C.M., 2012. Effect of a difficult calving on the vigour of the calf, the onset of maternal behaviour, and some behavioural indicators of pain in the dam. Prev. Vet. Med. 103(4), 248–256. doi: 10.1016/j.prevetmed.2011.09.001.



- Berry, D.P., Lee, J.M., Macdonals, A., Roche, J.R., 2007. Body condition score and body weights on dystocia and stillbirths and consequent effects on postcalving performance. *J. Dairy Sci.* 90(9), 4201-4211. DOI: 10.3168/jds.2007-0023.
- Beth, M.L., Brian, W.K., 2018. Short communication: Heritability of twinning rate in Holstein cattle. *J. Dairy Sci.* 101(5), 4307–4311. <https://doi.org/10.3168/jds.2017-13660>.
- Bicalho, R.C., Galvao, K.N., Warnick, L.D., Guard, C.L., 2008. Stillbirth parturition reduces milk production in Holstein cows. *Prev. Vet. Med.* 84(1-2), 112–120. <https://doi.org/10.1016/j.prevetmed.2007.11.006>.
- Bolacali, M., Öztürk, Y., 2018. Effect of non-genetic factors on milk yields traits in Simmental cows raised subtropical climate condition. *Arq. Bras. Med. Vet. Zootec.* 70(1), 297–305. <http://dx.doi.org/10.1590/1678-4162-9325>.
- Caraviello, D.Z., Weigel, K.A., Fricke, P.M., Wiltbank, M.C., Florent, M.J., Cook, N.B., Nordlund, K.V., Zwald, N.R., Rawson, C.M., 2006. Survey of management practices on reproductive performance of dairy cattle on large US commercial farms. *J. Dairy Sci.* 89, 4723–4735. doi: 10.3168/jds.S0022-0302(06)72522-X.
- Carvalho, P.D., Santos, V.G., Fricke, H.P., Hernandez, L.L., Fricke, P.M., 2019. Effect of manipulating progesterone before timed artificial insemination on reproductive and endocrine outcomes in high-producing multiparous Holstein cows. *J. Dairy Sci.*, 102, 7509–7521. doi: 10.3168/jds.2019-16536.
- Çilek, S., Tekin, M.E., 2005. Environmental factors affecting milk yield and fertility traits of Simmental cows raised at the Kazova state farm and phenotypic correlations between these traits. *Turk. J. Vet. Anim. Sci.* 29(4), 987–993.
- Çilek, S., Tekin, M.E., 2006. Calculation of adjustment factors for standardizing lactations to mature age and 305-day and estimation of heritability and repeatability of standardized milk yield of Simmental cattle reared on Kazova state farm. *Turk. J. Vet. Anim. Sci.* 30, 283–289.
- Czerniawska-Piatkowska E., Szewczuk M., Chocilowicz E., Konstancik N., 2012. Comparison of Limousin and Simmental primiparous cows based on the variability of age at first calving, body weight, and the analysis of their growth and development. *Electronic J. Polish Agric. Univ. Ser. Anim. Husbandry.* 15.
- Dahl, G.E., Buchanan, B.A., Tucker, H.A., 2000. Photoperiodic effects on dairy cattle: A review. *J. Dairy Sci.* 83(4), 885–893. [https://doi.org/10.3168/jds.S0022-0302\(00\)74952-6](https://doi.org/10.3168/jds.S0022-0302(00)74952-6).
- De Amicis, I., Veronesi, M.C., Robbe, D., Gloria, A., Carluccio, A., 2017. Prevalence, causes, resolution, and consequences of bovine dystocia in Italy. *Theriogenology.* 107, 104–108. <https://doi.org/10.1016/j.theriogenology.2017.11.001>.
- Duncan, D.B., 1955. Multiple range and multiple F tests. *Biometrics.* 11(1), 1–42.
- Eaglen, S.A.E., Coffey, M.P., Woolliams, J.A., Mrode, R., Wall, E., 2011. Phenotypic effects of calving ease on the subsequent fertility and milk production of dam and calf in UK Holstein–Friesian heifers. *J. Dairy Sci.* 94(11), 5413–5423. <https://doi.org/10.3168/jds.2010-4040>.
- Echternkamp, S.E., Gregory, K.E., 2002. Reproductive, growth, feedlot, and carcass traits of twin vs. single births in cattle. *J. of Anim. Sci.* 80(2), E64. doi.org/10.2527/animalsci2002.80E-Suppl\_21b.
- Eddy, R.G., Davies, O., David, C. 1991. An economic assessment of twin births in British dairy herds. *Vet. Rec.* 129(24), 526–529. PMID: 1788917.
- Emal, H., Ibrahim, Q.M., Noorullah, A., Nasrin, S., Zaher, S.M., 2021. Effect of season and lactation number on milk production of Holstein–Friesian cows in Kabul Bini-Hesar dairy farm. *Open J. Anim. Sci.* 11(3), 369–375. doi.org/10.4236/ojas.2021.113026.
- Fernando, L.G., Irina, G.I., Sergi, G., Robert, W., Daniel, J.W., Fernando, A.D.C., 2023. Effect of genetic and environmental factors on twin pregnancy in primiparous dairy cows. *Animals.* 13(12), 2008. doi.org/10.3390/ani13122008.
- Fodor, I., Kern, L., Varga-Balogh, O.G., Gábor, G., Ózsvári, L. 2019. The prevalence and impact of twinning and stillbirth on reproductive performance and their economic losses in large Hungarian dairy herds. *Magy. Allatorvosok Lapja.* 141(1), 3–10. doi/10.15407/animbiol21.02.021.
- Fourichon, C., Seegers, H., Bareille, N., Beaudeau, F., 1999. Effects of disease on milk production in the dairy cow: A review. *Prev. Vet. Med.* 41(1), 1–35. doi: 10.1016/s0167-5877(99)00035-5.
- Fricke, P.M., 2001. Twinning in dairy cattle. *The Professional Animal Scientist.* 17(2), 61–67. doi.org/10.15232/S1080-7446(15)31599-0.
- Gaafar, H.M.A., Shamiyah, S.M., Abu-El, M.A., Shitta, A.A., Tag, El-Din MA., 2011. Dystocia in Friesian cows and its effects on postpartum reproductive performance and milk production. *Trop. Anim. Health. Prod.* 43(1), 229–234. doi: 10.1007/s11250-010-9682-3.
- Garcia, I.I., López, G.F., 2018. The effects of a single or double GnRH dose on pregnancy survival in high producing dairy cows carrying singletons or twins. *J. Reprod. Dev.* 64(6), 523–527. doi: 10.1262/jrd.2018-057.
- Gaspárdy, A., Sheridan, J., Ari, M., Gulyás, L., 2018. Twin calving and its connection to other economically important traits in dairy cattle. In: Abubakar M., editor. *Ruminants: The Husbandry, Economic, and Health Aspects.* InTech, Rijeka, Croatia. pp. 61–82.
- Gheise, N.J.E., Riasi, A., Shahneh, A.Z., Celi, P., Ghoreishi, S.M. 2017. Effect of pre-calving body condition score and previous lactation on BCS change, blood metabolites, oxidative stress, and milk production in Holstein dairy cows. *Ital. J. Anim. Sci.* 16, 474–483. doi: 10.1080/1828051X.2017.1290507.
- Gladden, N., Ellis, K., Martin, J., McKeegan, D., 2021. Administration of ketoprofen affects post-partum lying behaviours of Holstein dairy cows regardless of whether parturition is assisted. *Vet. Rec.* 189(6), e300. doi.org/10.1002/vetr.300.
- Gorgulu, O., 2011. Path analysis on effective factors affecting 305-D milk yield in Simmental cattle. *Int. J. Agric. Biol.* 13, 381–385.

- Grosu, H., Oltenacu, P.A., 2005. Programe de ameliorare genetica in zootehnie. Ed. Ceres Publishing, Bucharest (Romania). pp.186–187.
- Grubbs, F., 1969. Procedures for detecting outlying observations in samples. *Technometrics*. 11(1), 1–21. doi.org/10.2307/1266761.
- Habibi, E., Qasimi, M., Ahmadzai, N., Stanikzai, N., Sakha, M., 2021. Effect of season and lactation number on milk production of Holstein–Friesian Cows in Kabul Bini-Hesar dairy farm. *Open J. Anim. Sci.* 11, 369–375. doi: 10.4236/ojas.2021.113026.
- Hill, T., Lewicki, P., 2007. *Statistics: Methods and Applications*, Stat Soft, Tulsa, OK, USA.
- Hossein-Zadeh, N.G., 2010. The effect of twinning on milk yield, dystocia, calf birth weight, and open days in Holstein dairy cows of Iran. *J. Anim. Physiol. Anim.* 94(6), 780–787. doi: 10.1111/j.1439-0396.2009.00963.x.
- Hossein-Zadeh, N.G., Nejati-Javaremi, A., Miraei-Ashtiani, S., Kohram, H., 2008. An observational analysis of twin births, calf stillbirth, calf sex ratio, and abortion in Iranian Holsteins. *J. Dairy Sci.* 91(11), 4198–4205. doi: 10.3168/jds.2008-1079.
- Huyghe, C., de Vlieghe, A., van Gils, B., Peeters, A., 2014. Grasslands and herbivore production in Europe and effects of common policies. Versailles Cedex, France: Quæ. pp.48.
- Jolanta, K., Zbigniew, D., 2002. Genetic aspects of twinning in cattle. *Review J. Appl. Genet.* 43(1), 55–68. PMID: 12084971.
- Kamal El-Den, M.A., Safaa S.S., Refaey, A.K., 2020. Genetic evaluation for milk production traits a herd of Friesian cattle raised in Egypt. *J. An Poult Prod. Mansoura Univ.* 11(10), 405–409. doi:10.21608/jappmu.2020.123628.
- Kinsel, M.L., Marsh, W.E., Ruegg, P.L., Etherington, W.G., 1998. Risk factors for twinning in dairy cows. *J. Dairy Sci.* 81(4), 989–993. doi: 10.3168/jds.S0022-0302(98)75659-0.
- Lopez, H., Caraviello, D.Z., Satter, L.D., Fricke, P.M., Wiltbank, M.C., 2005. Relationship between level of milk production and multiple ovulations in lactating dairy cows. *J. Dairy Sci.* 88(8), 2783–2793. doi.org/10.3168/jds.S0022-0302(05)72958-1.
- Lopez-Gaitus, F., Andreu-Vazquez, C., Mur-Novales, R., Cabrera, V., Hunter, R., 2017. The dilemma of twin pregnancies in dairy cattle. *Livest. Sci.* 197, 12–16. doi.org/10.1016/j.livsci.2017.01.001.
- Mayakrishnan, V., Ji, H.P., Kwang, S.K., Dong, H.L., Sang, B.K., Seong, M.P., Ha, Y.J., Beom, Y.P., Tae, I.K., 2017. The effect of lactation number, stage, length, and milking frequency on milk yield in Korean Holstein dairy cows using automatic milking system. *Asian-Australas. J. Anim. Sci.* 30(8), 1093–1098. doi.org/10.5713/ajas.16.0882.
- Mellado, M., Antonio-Chirino, E., Meza-Herrera, C., Veliz, F.G., Arevalo, J.R., Mellado J., de Santiago, A., 2011. Effect of lactation number, year, and season of initiation of lactation on milk yield of cows hormonally induced into lactation and treated with recombinant bovine somatotropin. *J. Dairy Sci.* 94(9), 4524–4530. doi: 10.3168/jds.2011-4152.
- Meyer, C.L., Berger, P.J., Koehler, K.J., Thompson, J. R., Sattler, C.G., 2001. Phenotypic trends in incidence of stillbirth for Holsteins in the United States. *J. Dairy Sci.* 84, 515–523. doi.org/10.3168/jds.S0022-0302(01)74502-X (2001).
- Miglior, F., Sewalem, A., Jamrozik, J., Lefebvre, D.M., Moore, R.K., 2006. Analysis of milk urea nitrogen and lactose and their effect on longevity in Canadian dairy cattle. *J. Dairy Sci.* 89, 4886–4894. doi: 10.3168/jds.S0022-0302(06)72537-1.
- Miller, A.R.E., Stanisiewski, E.P., Erdman, R.A., Douglass, L.W., Dahl, G.E., 1999. Effects of long daily photoperiod and bovine somatotropin on milk yield in cows. *J. Dairy Sci.* 82, 1716–1722. doi.org/10.3168/jds.S0022-0302(99)75401-9.
- Moioli, B., Steri, R., Marchitelli, C., Catillo, G., Buttazzoni, L., 2017. Genetic parameters and genome-wide associations of twinning rate in a local breed, the Maremmana cattle. *Animal*. 11(10), 1660–1666. doi: 10.1017/S1751731117000283.
- Moore, R.K., Kennedy, B.W., Schaeffer, L.R., Moxley, J.E., 1991. Relationships between age and body weight at calving and production in first lactation Ayrshires and Holsteins. *J. Dairy Sci.* 74, 269–278. doi: 10.3168/jds.S0022-0302(91)78170-8.
- Mostafa, S., 2009. Twinning in dairy cattle and its effect on milk yield, lactation length, dry period length, and calf performance. *BS. Vet. Med.* 19(2), 13–18. doi: 10.21608/JVMR.2009.77812.
- Mur-Novales, R., Lopez-Gatius, F., Fricke, P.M., Cabrera, V.E., 2018. An economic evaluation of management strategies to mitigate the negative impact of twinning in dairy herds. *J. Dairy Sci.* 101(9), 8335–8349. doi: 10.3168/jds.2018-14400.
- Naceur, M., Mahdi, B., Saoussen, F., Yosra, R., Satinder, K.B., Mohamed, B.H., 2012. Effects of environmental factors on milk yield, lactation length and dry period in Tunisian Holstein Cows. Chaiyabutr, Narongsak, In: *An Up-to-Date Overview of Animal Nutrition, Management, and Health*. Chapter 8. Ed. Milk Production - InTech. http://dx.doi.org/10.5772/50803
- Nardone, A., Ronchi, B., Lacetera, N., Ranieri, M.S., Bernabucci, U., 2010. Effects of climate changes on animal production and sustainability of livestock systems. *Livest. Sci.* 130, 57–69. doi: 10.1016/j.livsci.2010.02.011.
- Nielen, M., Schukken, Y.H., Scholl, D.T., Wilbrink, H.J., Brand, A., 1989. Twinning in dairy cattle: A study of risk factors and effects. *Theriogenology*. 32(5), 845–862. doi.org/10.1016/0093-691X(89)90473-1.
- Oltenacu, P.A., Broom, D.M., 2010. The impact of genetic selection for increased milk yield on the welfare of dairy cows. *Anim. Welfare*. 19 (S), 39–49. doi:10.1017/S0962728600002220.
- Özden, Ç., 2010. Twinning in cattle: Desirable or undesirable? *J. Biol. Environ. Sci.* 4(10), 1–8.

- Özkan, M., Güneş, H., 2011. Effects of some factors on milk yield characteristics of Simmental cows on commercial farms in Kayseri. *J. Fac. Vet. Med. Istanbul Üniv.* 37(2), 81–88.
- Pantelić, V., Aleksic, S., Stojic, P., Durdevic, R., Ljiljana, S., Dobrila, J., Niksic, D., 2010. The effect of breeding region and year on milk traits of Simmental bull dams. *Biotechnol. Anim. Husbandry.* 26, 287–295. doi:10.2298/BAH1006287P.
- Pantelic, V., Ružic-Muslic, D., Petrovic, M.M., Nikšić, D., Ostojic A.D., Aleksic, Stevica., Lazarevic, M., 2013. The phenotypic variability of production traits in the population of Simmental cows (Serbia). In: *Proceedings of the 10th International Symposium Modern Trends in Livestock Production*, Belgrade, Serbia, October 2–4, 2013, 26–36
- Petrovic, D.M., Bogdanovic, V., Petrovic, M., Snežana, B.B., Radojica, D., Radica, D., Simeon, R., 2015. Effect of non-genetic factors on standard lactation milk performance traits in Simmental Cows. *Ann. Anim. Sci.* 15, 211–220. doi: 10.2478/aoas-2014-0073.
- Pohl, A., Burfeind, O., Heuwieser, W., 2015. The associations between *postpartum* serum haptoglobin concentration and metabolic status, calving difficulties, retained foetal membranes, and metritis. *J. Dairy Sci.* 98, 4544–4551. doi: 10.3168/jds.2014-9181.
- Porter, P.A., Luhman, C.M., 2002. Changing photoperiod improves persistency in high producing Holstein cows. Abstract # 49 presented at the Midwest Meeting of the American Society of Animal Science /American Dairy Science Association.
- Proudfoot, K.L., Huzzey, J., von Keyserlingk, M.A.G., 2009. The effective of dystocia on the dry matter intake and behavior of Holstein cows. *J. Dairy Sci.* 92(10), 4937–4944. doi: 10.3168/jds.2009-2135.
- Rajala, P.J., Gröhn, Y.T., 1998. Effects of dystocia, retained placenta, and metritis on milk yield in dairy cows. *J. Dairy Sci.*, 81, 3172–3181. doi: 10.3168/jds.S0022-0302(98)75883-7.
- Rodrigo, C.B., Soon, H.C., Kliks, N.G., Lorin, D.W., Charles, L.G., 2007. Effect of twin birth calving on milk production, reproductive performance, and survival of lactating cows. *J. Am. Vet. Med. Assoc.* 231(9), 1390–1397. doi: 10.2460/javma.231.9.1390.
- Rodriguez-Venegas, R., Meza-Herrera, C.A., Robles-Trillo, P.A., Angel-Garcia, O., Legarreta-Gonzalez, M.A., Sánchez-Vocanegra, HF., Rodriguez-Martinez, R., 2023. Effect of THI on milk production, percentage of milking cows, and time lying in Holstein Cows in northern-arid Mexico. *Animals.* 13(10), 1715. doi.org/10.3390/ani13101715.
- Sangsrivavong, S., Combs, D.K., Sartori, R., Armentano, L., Wiltbank M.C., 2002. High feed intake increases liver blood flow and metabolism of progesterone and 17-estradiol in dairy cows. *J. Dairy Sci.* 85(11), 2831–2842. doi: 10.3168/jds.S0022-0302(02)74370-1.
- Sawa, A., Bogucki, M., G?owska, M., 2015. Effect of single and multiple pregnancies on performance of primiparous and multipa rous cows. *Arch. Tierzucht.* 58, 43–48. doi.org/10.5194/aab-58-43-2015.
- Shock, D.A., Renaud, D.L., Roche, S.M., Poliquin, R., Thomson, R., Olson, M.E., 2018. Evaluating the impact of meloxicam oral suspension administration at parturition on subsequent production, health, and culling in dairy cows: A randomized clinical field trial. *PLoS ONE.* 13(12), e0209236. doi: 10.1371/journal.pone.0209236.
- Silva, del R.N., Stewart, S., Rapnicki, P., Chang Y.M., Fricke, P.M., 2007. An observational analysis of twin births, calf sex ratio, and calf mortality in Holstein dairy cattle. *J. Dairy Sci.* 90(3), 1255–1264. doi.org/10.3168/jds.S0022-0302(07)71614-4.
- Steven, M.R., Joseph, A.R., Crystal, S., Kendall, B., Sjoert, Z., Brenda, R., Nick, A., Merle, O., 2023. Impact of dystocia on milk production, somatic cell count, reproduction and culling in Holstein dairy cows. *Animals.* 13(3), 346. doi.org/10.3390/ani13030346.
- Stevenson, J.S., Portaluppi, M.A., Tenhouse, D.E., 2007. Factors influencing upfront single- and multiple-ovulation incidence, progesterone, and luteolysis before a timed insemination resynchronization protocol. *J. Dairy Sci.* 90(12), 5542–5551. doi: 10.3168/jds.2007-0475.
- Tietjen, G.L., Moore, R.H., 1972. Some Grubbs-Type statistics for the detection of outliers. *Technometrics.* 14(3), 583–597. doi.org/10.2307/1267287.
- Victor, E.C., Paul, M. F., 2021. Economics of twin pregnancies in dairy cattle. *Animals.* 11(2), 552. doi: 10.3390/ani11020552.
- Wall, E., Mrode, R., Banos, G., Coffey, M., 2010. Development of calving-ease evaluations for UK Holstein–Friesian cows. *Proc. BSAS and ARF. Br. Soc. Anim. Sci., Belfast, UK (2010).* pp. 69.
- Yujie, W., Pengju, H., Yukun, S., Yonggen, Z., 2019. Effects of body condition score changes during peripartum on the postpartum health and production performance of primiparous dairy cows. *Animals (Basel).* 9(12), 1159. doi: 10.3390/ani9121159.
- Zukiewicz, A., Gtzesiak, W., Szatkowska, I., Blaszczyk, P., Dybus, A., 2012. Genetics Factors of milk yield in dairy cattle: Advances in the quest for universal markers. *Israel J. of Vet. Med.* 67(2), 82–91.