

## The variability in the prevalence of subclinical and clinical mastitis and its impact on milk yield of Holstein and Simmental cows as a result of parity

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### ABSTRACT

This study analysed over 10 million test-day records collected between January 2005 and December 2022, to determine the prevalence of subclinical and clinical mastitis in Holstein and Simmental cows, and its impact on subsequent milk production. The daily somatic cell count (SCC) served as an indicator of subclinical (200,000/ml to 400,000/ml) and clinical (>400,000/ml) mastitis. The study found that mastitis-related problems increased with parity, and were more frequent in Holstein cows than in Simmental cows. The study identified factors affecting daily milk yield, including lactation stage, age at first calving, farm size, recording season, and breeding region. It was also found that subclinical and clinical mastitis significantly impacted milk production. The day when subclinical/clinical mastitis was detected (D-0) had the lowest milk yield, followed by an increase in milk production in the subsequent recordings. The increase in production varied depending on the form of mastitis, cow's breed (Holstein or Simmental), and cow's parity. The first successive milk recording (A-1) showed the greatest increase in daily milk yield across both breeds and all parities. The increase in daily milk yield was higher following clinical than subclinical mastitis, and was more pronounced in Holstein than in the Simmental breed. After the initial increase in milk production, almost all cows had a decline in milk yield in the subsequent period after subclinical mastitis. Further, a continued increase was observed in all cows after clinical mastitis. The analysis of monthly and total differences in milk production indicated that cows had the highest monthly increase in milk production in the first month after subclinical/clinical mastitis. The highest total increase in milk production was observed in Holstein primiparous cows, which amounted to 44.91 kg after clinical mastitis. In comparison to the Holstein breed, Simmental cows had a lower increase in milk production. Generally, Holstein cows had a better production increase or recovery potential than Simmental cows, particularly after clinical mastitis. Primiparous Holsteins and Simmentals exhibited the highest total production increase in the second parity. The analysis of the animal's recovery capability after the occurrence of a mastitis-related problem revealed that primiparous cows had the lowest initial increase in milk production, but they were able to regenerate more in the total period compared to the older cows with higher initial but lower total increase.

**Key words:** dairy cows; subclinical and clinical mastitis; prevalence; effect; milk production

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## Introduction

Dairy cattle are prone to various disorders, which can impact their milk production and well-being, and the efficiency of dairy farms (HOGEVEEN et al., 2019). Common disorders include mastitis, metabolic disorders, lameness, digestive disorders, reproductive disorders, respiratory diseases, parasitic infections, and heat stress. Preventive measures, routine health checks, and appropriate treatments are essential for maintaining the health and productivity of dairy cattle (NIR, 2003). Mastitis, an inflammatory disease of the mammary gland, is a widespread and economically significant health issue in dairy cattle (SEEGERS et al., 2003; HALASA et al., 2007; BENIĆ et al., 2018; HADRICH et al., 2018; MAČEŠIĆ et al., 2022; CHEN et al., 2023), affecting various breeds, including Holstein and Simmental cows. Although the general characteristics of mastitis are comparable across breeds, there may be some variations in its manifestations or the prevalence of certain pathogens (NÓBREGA and LANGONI, 2011). Holstein cows are recognized for their high milk production. However, this increased production may make them more vulnerable to mastitis due to the stress imposed on their udders. Holsteins may be more prone to environmental mastitis pathogens, such as *Escherichia coli* and environmental *streptococci*. BENIĆ et al. (2018) noted that bovine mastitis is often infectious, and can be caused by some 150 microbial species, while the *Staphylococcus aureus* is the most common pathogen isolated from the mammary gland capable of causing health disorders in humans. Furthermore, the same authors stated that the environment of the dairy cow is heavily colonized by microbiota, and pathogens usually enter the udder through contaminated bedding or mud. Holsteins with mastitis may exhibit clinical signs such as swelling, redness, and heat in the udder. Milk from affected quarters may appear abnormal, with changes in colour and consistency. Furthermore, Holsteins typically have larger udders and longer teats than other dairy breeds, which can make them more vulnerable to injury and contamination during the milking process, leading to mastitis. Simmental cows are known for their adaptability to different environments. They may encounter

specific management practices and environmental conditions, which may affect the risk of mastitis. Simmentals have diverse genetic backgrounds, and individual animals may vary in their susceptibility to mastitis. Genetic factors can influence immune response and resistance to infections. Similar to Holsteins, Simmental cows with mastitis may show signs such as udder swelling, redness, and changes in milk appearance. The severity and manifestation of clinical signs can vary between individuals. Additionally, the size and conformation of the teats in Simmental cows can vary, requiring proper teat care and milking practices to prevent injuries and reduce the risk of mastitis. Both Holstein and Simmental cows benefit from good management practices, including proper hygiene during milking, regular udder health checks, and prompt treatment of any signs of mastitis.

Mastitis is a significant concern for the dairy industry, as it can lead to various negative outcomes that affect both individual cows and overall herd productivity (HALASA et al., 2007; HADRICH et al., 2018; CHEN et al., 2023). The consequences of this disease can be divided into several categories, including decreased milk production, altered milk composition, economic loss, quality issues, increased veterinary costs, reproductive issues, culling of cows, elevated somatic cell count (SCC), animal welfare concerns, contagious spread, and long-term impact on udder health (HALASA et al., 2007). Mastitis can result in a reduced milk yield due to the inflammatory response and damage to the udder tissue. Additionally, milk from affected cows may have changes in composition, including increased SCC, elevated levels of white blood cells, and altered levels of milk components such as fat and protein. The economic impact of mastitis is significant, with costs associated with decreased milk production, veterinary treatments, discarded milk, and potential culling of chronically infected cows, all contributing to financial losses. Moreover, managing mastitis requires veterinary intervention, including the use of antibiotics and other treatments, which can be substantial. Mastitis also poses reproductive issues, including delayed oestrus, reduced conception rates, and increased days open, which can compromise fertility due to

the systemic effects of the inflammatory response. Furthermore, culling of cows may be necessary in cases of chronic or severe mastitis to prevent the spread of infection and minimize economic losses. Additionally, mastitis can cause discomfort and pain for affected cows, highlighting the importance of proper management practices and timely treatment to alleviate suffering. Contagious spread of mastitis can lead to a higher prevalence of the disease within the herd if appropriate control measures are not implemented. Furthermore, chronic mastitis can have long-term effects on udder health, potentially leading to permanent damage and reduced milk production, even after treatment.

The timely identification of mastitis in dairy cattle is essential for effective management and treatment (ARGAW, 2016). A range of methods and technologies are available for detecting mastitis in its early stages (NARVÁEZ-SEMANATE et al., 2022). The following are some of the most common approaches to mastitis detection in dairy cattle: clinical observation, SCC monitoring, milk electrical conductivity, udder temperature monitoring, milk culture, California mastitis test, automated milk analysers, machine milking parameters, rapid mastitis tests, teat-end health monitoring, and automated health monitoring systems. Furthermore, the onset of mastitis is mediated by vitronectin, making it a valuable marker for the diagnosis of the subclinical form of the disease according to TURK et al. (2012). NEDIĆ et al. (2019) have shown that subclinical mastitis reduces paraoxonase-1 (PON1) activity in the blood and milk of affected cows due to oxidative stress and inflammatory reactions. KOVAČIĆ et al. (2019) have confirmed that PON1 activity is significantly lower in both the subclinical and clinical mastitis groups compared with the control, highlighting its potential as a biomarker for diagnosing the subclinical form of the disease. CVETNIĆ et al. (2016) emphasized the importance of systematic mastitis control in dairy herds to decrease mastitis occurrence and its significance in dairy production.

SCC represents an essential indicator of mastitis since SCC is associated with inflammatory processes, making it a useful diagnostic method for assessing udder health (SCHUKKEN et al.,

2003.; IVANOV et al., 2016; PETZER et al., 2017). BLACKBURN (1966) stated that the higher SCC during successive lactations was the result of an increase in polymorphonuclear leukocyte (PMN) count. However, an increase in SCC during any lactation was associated with an increase in both PMN and other milk somatic cells. SMITH et al. (2001) stated that a healthy udder has an SCC of less than 100,000/ml, while MIKÓ et al. (2016) noted that healthy milk should contain from 20,000 to 100,000 SCC/ml. MIKÓ et al. (2016) and HADRICH et al. (2018) pointed out that an increase in SCC can result in a loss of milk production, leading to economic losses for dairy farmers. The total milk losses in the herd depend on the distribution of SCC at the cow level and parity within the herd (CHEN et al., 2023). Furthermore, PFÜTZNER and ÓZSVÁRI (2016) emphasized that the extent of the decline in milk production was directly associated with the degree of SCC. Cows with SCC levels from 50,000 to 100,000 cells/ml exhibited a loss greater than 8%, while those with average SCC levels from 100,000 to 250,000 cells/ml displayed a decrease in milk production by more than 15%, and even up to 18%. GANTNER et al. (2011; 2017) analysed the impact of heat stress on SCC in Holstein cows and reported SCC variations due to the temperature-humidity index value, daily production level, breed, and parity. Furthermore, the effect of lactation stage, parity, season, and region on SCC was confirmed by several studies (BLACKBURN, 1966; SEEGERS et al., 2003; NÓBREGA and LANGONI, 2011; YANG et al., 2013; TOMAZI et al., 2018; WEBER et al., 2020; STOCCO et al., 2023; CHEN et al., 2023).

This study aimed to determine the variability in the prevalence of subclinical and clinical mastitis as well as its impact on subsequent milk production of Holstein and Simmental cows due to parity, using the test-day records.

## Materials and methods

The statistical analysis conducted in this study was based on *test-day records* of Holstein and Simmental dairy cows. The data was collected over a period of 17 years, from January 2005 to December 2022, during regular milk recording.

The milk recording process adhered to the guidelines set by the International Committee for Animal Recording (ICAR, 2017) for alternative milk recording methods (AT4/BT4). This method involves measuring milk yield and collecting milk samples from each cow during morning or evening milking every four weeks. These activities were carried out either by trained officers from the Croatian Agency for Agriculture and Food (HAPIH) for the AT4 method, or by trained employees at the farm for the BT4 method. The milk samples were then transported to the accredited Central Laboratory for Milk Quality Control (SLKM) of HAPIH for analysis, again following the ICAR (ICAR, 2017) procedures. The chemical quality of the milk samples (the content of milk fat, protein, lactose, dry matter, dry matter without fat, urea, casein, free fatty acids, pH value, ketone bodies, and freezing point) was determined using infrared spectrophotometry on MilkoScan analysers. Additionally, somatic cell counts were determined using Fossomatic analysers and the fluoro-opto-electronic method.

Before the logical data control process, the dataset obtained from HAPIH contained more than 10 million test-day records. As part of the logical data control process, test-day records that did not meet the ICAR (2017) standards were deleted from the database. Additionally, records with missing or incorrect values for breed, farm code, breeding region, lactation stage, parity, age at first calving, calving date, and milk recording date were also deleted from the database. After the logical control, the database contained 3,953,637 test-day records of Holstein and 4,922,751 test-day records of Simmental cows.

Regarding the recording date, the test-day records were categorized into four recording seasons: winter (December, January and February), spring (March, April, and May), summer (June, July, and August), and autumn (September, October, and November). Additionally, the test-day records were grouped into three breeding regions: Central, Eastern, and Mediterranean, based on the farm's location. The number of cows on the farm was also factored in, with Simmental cows being grouped into five classes (ranging from less than 5 cows to 200-500 cows) and Holstein cows into six

classes (ranging from less than 5 cows to over 500 cows). The cows were divided into four different classes based on parity: I., II., III., and IV. + (cows in fourth and higher lactation).

The daily SCC served as an indicator of subclinical and clinical mastitis. Cows were classified into the mastitis classes on the basis of their SCCs, with SCCs lower than 200,000/ml indicating healthy animals, SCCs ranging from 200,000/ml to 400,000/ml indicating subclinical mastitis, and SCCs higher than 400,000/ml indicating clinical mastitis. The *prevalence of subclinical and clinical mastitis* was calculated as the percentage of cows in a specific mastitis class from the total number of animals. The prevalence rate was calculated separately for each breed and parity class.

To analyse the *effect of subclinical or clinical mastitis prevalence* on successive milk production, only cows with confirmed subclinical or clinical mastitis were included in further analysis. The daily milk yield measured on the test day when subclinical or clinical mastitis was observed was taken as the starting value. The mastitis index was defined on the basis of the duration of the period after determination of subclinical or clinical mastitis, with D-0 being the starting test day, A-1 being within 35 days, A-2 being between 36 and 70 days, A-3 being between 71 and 105 days, and A-4 being more than 105 days. The effect of the mastitis index on daily milk yield was tested using a statistical model that accounted for the variability due to the lactation stage (as lactation curve), age at first calving, farm size, recording season, breeding region, and mastitis index. The significance of the differences between estimated LsMeans was tested by Scheffe's method of multiple comparisons in PROC MIXED (SAS Institute Inc., 2019). This analysis was performed separately for each breed and parity class.

*Estimated differences in daily milk yield* between the analysed milk recordings (A-1 – D-0, A-2 – A-1; A3 – A2; and A4 – A3) after the subclinical or clinical mastitis prevalence was analysed and graphically presented by parity class separately for each breed. Furthermore, based on the estimated daily differences and the intervals between successive milk recordings, *the monthly*



and total difference in milk production over four successive milk recordings (from D-0 to A-4) after subclinical or clinical mastitis was calculated separately by parity class and breed.

### Results

The present study provides an in-depth analysis of the prevalence of subclinical and clinical mastitis in two different breeds of dairy cows, namely Holstein and Simmental. The results of the analysis are depicted in Fig. 1 and 2 for Holstein and Simmental breeds, respectively. As per the findings, 35.4% of Holstein cows and 34.3% of Simmental cows exhibited mastitis-related problems, either subclinical or clinical. The prevalence of subclinical

mastitis in Holstein cows was found to range from 11.4% in first parity cows to 16.9% in cows in the fourth or higher parities. A similar trend was observed in the Simmental breed. Furthermore, the prevalence of clinical mastitis was found to range from 14.6% in the first parity to 33.1% in cows in the fourth or higher parities in the Holstein breed, and from 13.3% in the first parity to 26.1% in cows in the fourth or higher parities in Simmental breed. Finally, the prevalence of mastitis-related problems increased with parity in both breeds. These findings provide valuable insights into the prevalence and trends of mastitis in dairy cow populations, which could enable the development of effective management and preventive strategies to mitigate the prevalence of mastitis in dairy herds.

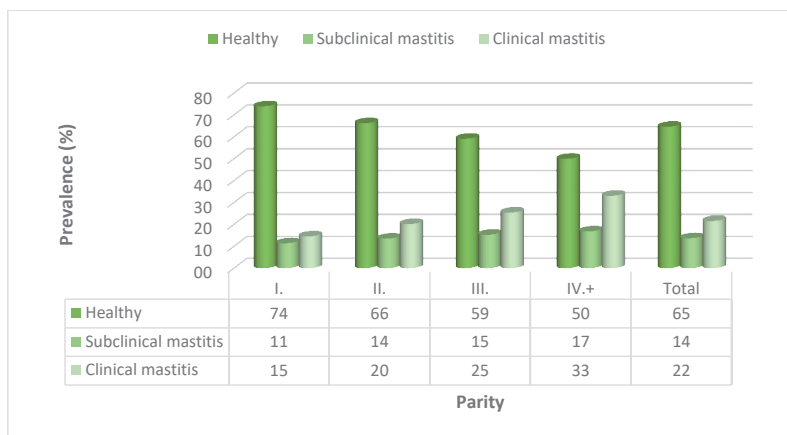


Fig. 1. Prevalence of subclinical and clinical mastitis in Holstein cows regarding the parity class (I., II., III., and IV. +)

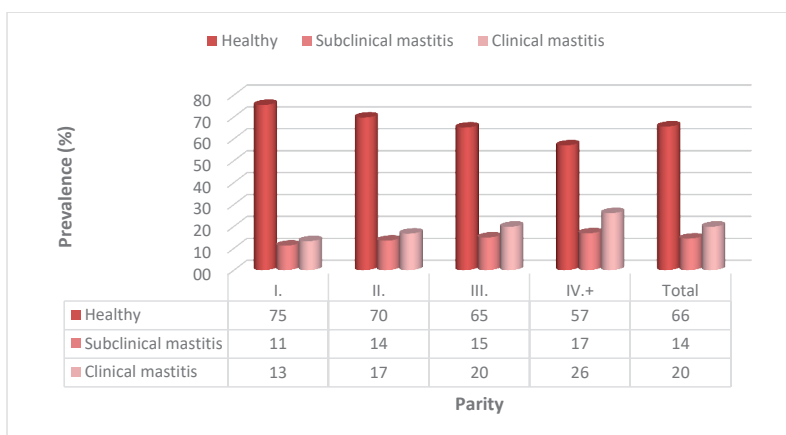


Fig. 2. Prevalence of subclinical and clinical mastitis in Simmental cows regarding the parity class (I., II., III., and IV. +)

The statistical analysis showed that all independent variables accounted for in the statistical model, including lactation stage, age at first calving, farm size, recording season, and breeding region, as well as mastitis index (D-0, A-1, A-2, A-3, A-4), had a statistically highly significant effect ( $P < 0.001$ ) on daily milk yield for both breeds and all parity classes. In Table 1, the LsMeans of daily milk yield across analysed milk recordings (D-0 – A-4) regarding the form of mastitis (subclinical or clinical), cow's

breed (Holstein or Simmental), and cow's parity (I., ..., IV. +) are presented. For Holsteins in their first parity, LsMeans ranged from 21.70 kg/day at D-0 to 22.31 kg/day at A-4 after subclinical mastitis, and from 18.92 kg/day at D-0 to 20.43 kg/day at A-2 after clinical mastitis. In the case of Simmental cows in their first parity, LsMeans ranged from 16.24 kg/day at D-0 to 16.64 kg/day at A-4 after subclinical, and from 14.45 kg/day at D-0 to 15.28 kg/day at A-2 after clinical mastitis.

Table 1. LsMeans of daily milk yield at analysed milk recordings (D-0, A-1, A-2, A-3, A-4) after the determination of subclinical and clinical mastitis regarding the breed (Holstein and Simmental and parity class (I., II., III., and IV. +))

Holstein breed		Subclinical mastitis				Clinical mastitis			
MR	Parameter	I.	II.	III.	IV. +	I.	II.	III.	IV. +
D-0	Estimate	21.70	23.44	23.84	23.05	18.92	20.66	21.05	20.28
D-0	StdErr	0.04	0.06	0.05	0.05	0.09	0.11	0.12	0.09
A-1	Estimate	22.09	23.78	24.23	23.35	20.25	22.06	22.68	21.62
A-1	StdErr	0.04	0.06	0.06	0.05	0.10	0.12	0.13	0.10
A-2	Estimate	22.09	23.96	24.38	23.42	20.43	22.11	22.73	21.61
A-2	StdErr	0.04	0.06	0.05	0.05	0.10	0.12	0.13	0.10
A-3	Estimate	22.21	23.99	24.50	23.46	20.42	21.99	22.66	21.60
A-3	StdErr	0.04	0.06	0.06	0.06	0.10	0.13	0.13	0.10
A-4	Estimate	22.31	24.12	24.69	23.59	20.42	21.92	22.47	21.32
A-4	StdErr	0.04	0.05	0.05	0.05	0.08	0.11	0.11	0.08
Simmental breed		Subclinical mastitis				Clinical mastitis			
MR	Parameter	I.	II.	III.	IV. +	I.	II.	III.	IV. +
D-0	Estimate	16.24	16.60	17.41	16.61	14.45	16.19	15.37	15.29
D-0	StdErr	0.05	0.06	0.07	0.05	0.17	0.18	0.18	0.10
A-1	Estimate	16.43	16.75	17.63	16.78	15.20	17.00	16.20	15.87
A-1	StdErr	0.05	0.06	0.07	0.05	0.18	0.18	0.18	0.11
A-2	Estimate	16.47	16.74	17.62	16.76	15.15	16.96	16.12	15.85
A-2	StdErr	0.05	0.06	0.07	0.05	0.18	0.18	0.18	0.11
A-3	Estimate	16.54	16.82	17.67	16.81	15.23	17.04	16.20	15.83
A-3	StdErr	0.06	0.06	0.07	0.05	0.18	0.18	0.19	0.11
A-4	Estimate	16.65	16.87	17.70	16.83	15.28	17.05	16.14	15.89
A-4	StdErr	0.05	0.06	0.07	0.05	0.17	0.17	0.18	0.10

\* MR – milk recording, \*D-0 – milk recording when the subclinical/clinical mastitis prevalence was determined; A-1, A-2, A-3, A-4 – successive milk recordings

In Holstein cows, the highest starting daily milk yield (D-0) with subclinical and clinical mastitis was observed in cows in their third lactation, with yields of 23.84 kg/day and 21.05 kg/day, respectively. Similarly, in Simmental cows, the highest starting daily milk yield (D-0) with subclinical mastitis, in the amount of 17.41 kg/day, was observed in cows in their third lactation, while in the case of clinical mastitis, the highest starting yield of 16.19 kg/day was observed in cows in their second lactation.

The lowest daily milk yield was at D-0 (the test day when subclinical or clinical mastitis was determined) for both subclinical and clinical mastitis, both breeds and all parity classes, followed by an increase in milk production at successive milk recordings. However, the extent of increase in production, and the animal's ability to recover after the occurrence of mastitis-related problems, varied depending on the form of mastitis (subclinical or clinical), the cow's breed (Holstein or Simmental), and the cow's parity (I., ..., IV.+). Furthermore, lower daily milk yields at D-0 recording were determined in Simmental than in Holstein cows, as well as in the case of clinical than subclinical mastitis.

The estimated differences in daily milk yield between the analysed milk recordings (D-0&A-1, A-1&A-2, A-2&A-3, and A-3&A-4) after the subclinical and clinical mastitis prevalence are presented regarding the parity class and breed. Fig. 3 and 4 represent the data for Holstein breed cows, while fig. 5 and 6 represent the data for Simmental breed cows. The estimated differences in daily milk

yield were found to be dependent on the form of mastitis, the breed of the cow, and the cow's parity.

In general, the first successive milk recording (A-1) showed the highest increase in daily milk yield for both breeds and all parities. The data revealed that the increase in daily milk yield was higher after clinical mastitis than subclinical mastitis, and it was higher in Holstein breed cows compared to Simmental breed cows. The data also showed that the estimated differences in daily milk yield varied depending on the cow's parity.

The study showed that Holstein cows in their first and third lactation experienced the highest increase in daily milk yield, with an average increase of 0.39 kg/day after the occurrence of subclinical mastitis. On the other hand, the cows in their fourth or later lactation (IV. + parity) had the lowest production increase of 0.30 kg/day in the first month after the subclinical mastitis. It was also observed that after the initial increase in milk production, almost all Holstein cows experienced a decrease in milk production in the following period (from the second to the fourth milk recordings) after subclinical mastitis. This decline in milk production was consistent across all lactations.

In the case of clinical mastitis, the increase in daily milk yield in the first month was found to be higher, ranging from 1.33 kg/day in primiparous cows to 1.63 kg/day in cows in their third lactation. The increase in milk production observed predominantly continued in the successive milk recordings, except for A-2 in cows in their first and third lactation.

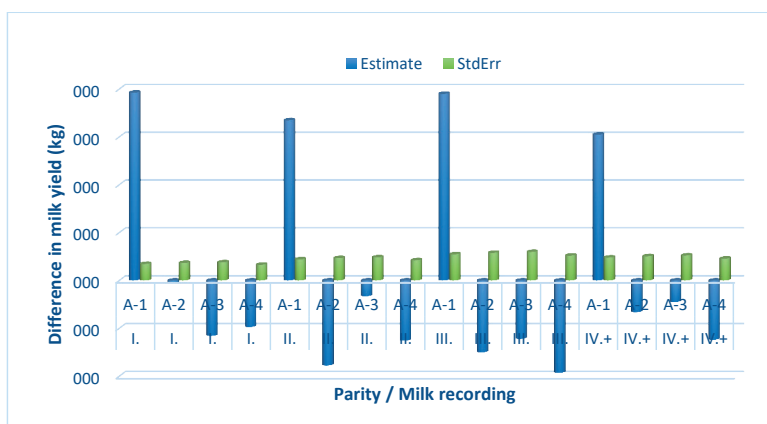


Fig. 3. Estimated differences in daily milk yield between the analysed milk recordings after subclinical mastitis in Holstein cows in relation to parity

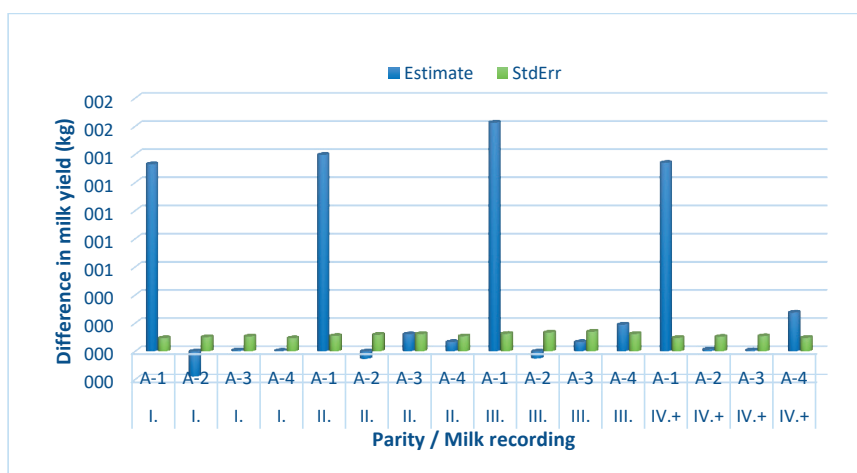


Fig. 4. Estimated differences in daily milk yield between the analysed milk recordings after clinical mastitis in Holstein cows in relation to parity

The study found that Simmental cows in the third parity showed the highest increase in daily milk yield, with an average increase of 0.22 kg per day in the first month after the occurrence of subclinical mastitis. On the other hand, the cows in the second parity showed the lowest increase in

daily milk yield, with an average increase of 0.15 kg per day. This pattern was similar to that observed in the Holstein breed. Following the initial phase of regeneration and increase in milk production, daily milk production generally decreased in the following months, except for A-2 in older cows in parities II, III, and IV+.

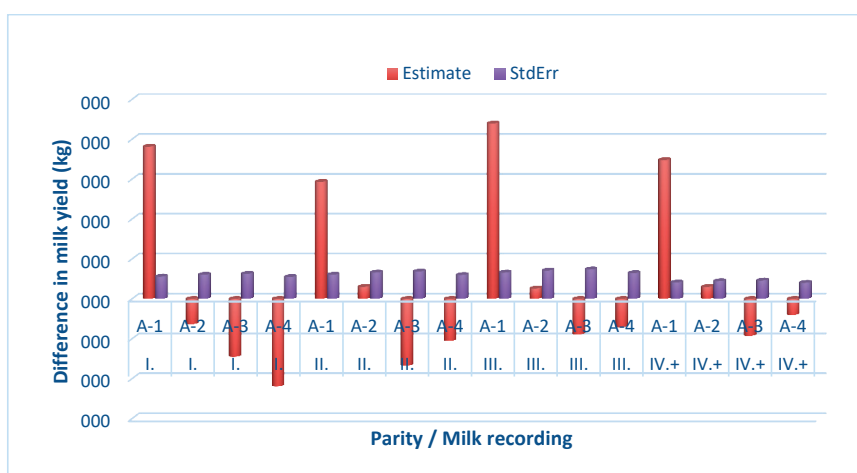


Fig. 5. Estimated differences in daily milk yield between the analysed milk recordings after subclinical mastitis in Simmental cows in relation to parity



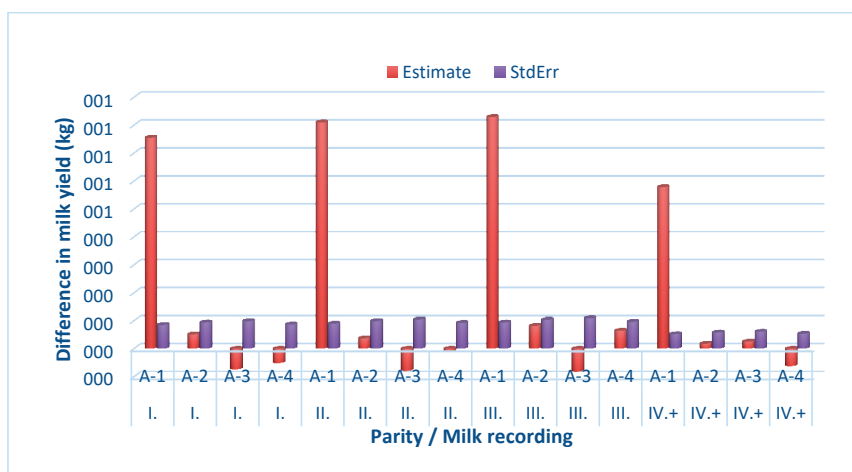


Fig. 6. Estimated differences in daily milk yield between the analysed milk recordings after clinical mastitis in Simmental cows in relation to parity

The study also found that in the first month after clinical mastitis, the cows were able to recover their production potential and increase their daily milk production by 0.58 kg per day for cows in parity IV +, and up to 0.83 kg per day for those in parity III. This increase in production continued until the 2<sup>nd</sup> or 3<sup>rd</sup> milk recording, particularly for cows in parity IV +.

The estimated monthly and total differences in milk production at and over four successive milk recordings (from D-0 to A-4) after subclinical or clinical mastitis are presented in Table 2. The results indicate that Holstein cows showed a higher monthly increase in milk production in the first month after clinical mastitis, particularly cows in the third lactation, with an increase of 48.80 kg/month. In contrast, cows in parity IV + after subclinical mastitis had the lowest increase in milk production, which was 9.14 kg/month. It was also observed that cows in parity IV + experienced the highest decrease in milk production, with a decline of 8.29 kg/month in the period between the third and fourth milk recordings after the clinical mastitis. Finally, the highest total increase in milk production, in the amount of 44.91 kg, was determined in primiparous Holstein cows after clinical mastitis. The highest total increase after subclinical mastitis was determined in Holstein cows in the third lactation.

The analysis of the animals' recovery capability revealed that primiparous cows had the lowest initial increase, but they were able to regenerate more in the total period compared to the older cows with higher initial but lower total increase.

In comparison to the Holstein breed, Simmental cows had a lower increase in milk production, followed by the highest total increase after clinical mastitis, of 25.81 kg, observed in the second parity. The oldest Simmental cows (IV. +) had the lowest total increase compared to younger cows, which was determined to be 6.77 kg after subclinical mastitis.

Generally, Holstein cows had a better production increase or recovery potential than Simmental cows, particularly after clinical mastitis. Furthermore, younger animals, such as primiparous Holsteins and Simmentals in their second parity, exhibited the highest total production increase.

Table 2. The total difference in milk production in the analysed period of four successive milk recordings (from D-0 to A-4) after the determination of subclinical and clinical mastitis in relation to breed (Holstein and Simmental) and parity class (I., II., III., and IV. +)

Holstein	Subclinical mastitis					Clinical mastitis				
Parity	A-1	A-2	A-3	A-4	Total	A-1	A-2	A-3	A-4	Total
I.	11.78	0.19	3.47	2.92	18.36	39.93	5.39	-0.26	-0.15	44.91
II	10.05	5.32	0.99	3.77	20.13	41.93	1.59	-3.65	-2.00	37.87
III.	11.71	4.51	3.66	5.79	25.66	48.80	1.54	-1.99	-5.73	42.63
IV. +	9.14	1.99	1.35	3.71	16.19	40.24	-0.41	-0.26	-8.29	31.27
Simmental	Subclinical mastitis					Clinical mastitis				
Parity	A-1	A-2	A-3	A-4	Total	A-1	A-2	A-3	A-4	Total
I.	5.72	0.96	2.18	-3.29	12.14	22.66	-1.55	2.22	1.55	24.89
II	4.40	-0.45	2.51	-1.58	8.04	24.29	-1.13	2.37	0.27	25.81
III.	6.59	-0.39	1.34	-1.07	8.61	24.89	-2.46	2.46	-1.94	22.95
IV. +	5.22	-0.45	1.39	-0.61	6.77	17.36	-0.54	-0.77	1.88	17.92

\* A-1, A-2, A-3, A-4 – successive milk recordings after the subclinical/clinical mastitis

## Discussion

This study analysed mastitis prevalence and its effect on milk production in Holstein and Simmental dairy cows. 35.4% of Holstein and 34.3% of Simmental cows showed mastitis-related issues. The prevalence of subclinical mastitis increased with parity, ranging from 11.4% to 16.9% in Holstein and from 11.2% to 16.9% in Simmental cows. Furthermore, clinical mastitis also increased with parity, and ranged from 14.6% to 33.1% in Holstein and from 13.3% to 26.1% in Simmental cows. Finally, the prevalence of mastitis-related problems increased with the animal's age (parity), and was more frequent in Holstein than in the Simmental breed.

ANTANAITIS et al. (2021) and COSTA et al. (2019) reported that the number of somatic cells tends to increase, while daily lactose content decreases with increasing parity. In addition, the prevalence of mastitis has been found to increase with parity, primarily due to the deterioration of the immune system, anatomical changes in the udder and teats, and repeated exposure to milking procedures (GIRMA and TAMIR, 2022). GONCALVES et al. (2022) indicated that the incidence rate for mastitis

is higher among older cows (parity  $\geq 3$ ), and during the first 100 days of lactation. According to these authors, cows in early lactation in the first and second parity are more likely to remain in the herd after disease diagnosis compared to older cows and cows in later stages of lactation. Furthermore, NÓBREGA and LANGONI (2011) found that Holstein cows had higher somatic cell counts compared to Jersey cows, which they attributed to differences between cow breeds in some aspects of the immune response. The prevalence of mastitis is increasing in parallel to the development of new, highly-productive breeds (SHARMA et al., 2018). A significantly higher incidence of udder infections was detected in Jersey and crossbred cows than in indigenous breeds, whereas Holstein cows are more prone to mastitis due to the size of their udders, the position of the teat, and the anatomy of the teat canal (FESSEHA et al., 2021). Moreover, FESSEHA et al. (2021) explained that high-producing dairy cattle are more susceptible to udder inflammation than low-producing cows due to the consequences of possible injuries that are more likely to occur in larger udders, which

predisposes the udder to infection because it allows the entry of pathogens, as well as stress due to the large volume of milk, which can also disrupt the cow's defence system. GIRMA and TAMIR (2022) further confirmed the effect of breed, parity, and stage of lactation on the prevalence of mastitis, reporting that crossbred cows are 2.17 times more likely to have mastitis than local breeds. This may be attributed to various hereditary traits, such as the ability to produce milk, teat characteristics, and udder shape, as well as the breed's genetic ability to resist disease, difficulties in adapting to a new environment, and the anatomical size of the udder in crossbreds, which is large and easily infected with bacterial pathogens.

This study found that various factors, including lactation stage, age at first calving, farm size, recording season, and breeding region, as well as subclinical/clinical mastitis, had a significant effect on daily milk yield. The lowest milk yield was observed on the day when the subclinical/clinical mastitis was detected (D-0), followed by an increase in milk production in the subsequent recordings. Furthermore, lower daily milk yields at D-0 recording were determined in Simmental than in Holstein cows, as well as in clinical than subclinical mastitis. Holstein and Simmental cows showed the highest initial daily milk yield at subclinical and clinical mastitis in their third or second lactation (Simmental after subclinical mastitis). Furthermore, the extent of production increase varied depending on the form of mastitis (subclinical or clinical), the cow's breed (Holstein or Simmental) and the cow's parity (I., ..., IV. +). The estimated differences in daily milk yield between the analysed milk recordings (D-0&A-1, A-1&A-2, A-2&A-3, and A-3&A-4) varied in relation to the form of mastitis, the breed and the parity of the cow. The results revealed that the first successive milk recording (A-1) exhibited the greatest increase in daily milk yield across both breeds and all parities. Also, the increase in daily milk yield was higher following clinical than subclinical mastitis, and was more pronounced in Holstein than in the Simmental breed. The estimated differences in daily milk yield also varied depending on the cow's parity. The cows in their third lactation experienced the

highest increase in daily milk yield. Furthermore, after the initial increase in milk production, almost all cows experienced a decline in milk yield in the subsequent period after subclinical, and a continued increase after clinical mastitis.

According to SEEGERS et al. (2003), cows affected with mastitis, and in particular, those with a higher number of lactations, tend to produce more milk. Additionally, GIRMA and TAMIR (2022) suggest that lower milk production in older cows ( $\geq 4$ th lactation) can be attributed to anatomical changes in the udder. ATASEVER and ERDEM (2008) report that milk production losses increase proportionally with the number of lactations. DÜRR et al. (2008) observed a variation in milk losses per unit increase in ISCC among Holstein cows. In the first lactation, milk losses ranged from 0.33 to 0.52 kg/day, whereas in cows from the 2nd to 4th lactation, losses ranged from 0.76 to 1.80 kg/day. Likewise, GONÇALVES et al. (2018) found that in Holstein cows in the first lactation, the loss of milk production was 1.75 kg/day at the level of SCC 100,000/ml and 2.21 kg/day at the level of SCC 200,000/ml. Furthermore, cows in their 2<sup>nd</sup> and 3<sup>rd</sup> lactation experienced greater losses than those in their 1<sup>st</sup> lactation. At SCC levels of 200,000/ml, HAND et al. (2012) estimated milk losses of 0.35 to 0.80 kg/day for cows in their first lactation, 0.61 to 1.07 kg/day for cows in their second lactation, and 0.63 to 1.09 kg/day for cows in their third lactation. Meanwhile, at SCC levels of 500,000/ml, milk production losses were found to be greater, ranging from 0.80 to 1.87 kg/day for cows in their first lactation, 1.42 to 2.49 kg/day for cows in their second lactation, and 1.46 to 2.52 kg/day for cows in their third lactation (HAND et al., 2012). The effect of parity on SCC and milk production was also determined by YANG et al. (2013).

The analysis of monthly and total differences in milk production indicates that cows had the highest monthly increase in milk production in the first month after subclinical/clinical mastitis. The highest total increase in milk production in the amount of 44.91 kg was determined in primiparous Holstein after clinical mastitis. In comparison to the Holstein breed, Simmental cows had a lower increase in milk production. Generally, Holstein

cows had a better production increase or recovery potential than Simmental cows, particularly after clinical mastitis. Furthermore, younger animals, such as primiparous Holsteins and Simmentals in the second parity, exhibited the highest total production increase. The analysis of the animal's recovery capability revealed that primiparous cows had the lowest initial increase in milk production, but they were able to regenerate more in the total period, compared to the older cows with higher initial but lower total increase.

YANG et al. (2013) found that milk yield, composition, and SCC were affected by parity and season. HUIJPS et al. (2008) discovered that the economic losses of a clinical case of mastitis amounted to €210 on average, while more than 70% of farmers underestimated their losses. The same authors emphasised that underestimating the economic impact of mastitis represents a general problem in the dairy sector, and can result in inadequate investment in prevention and control measures. The significant economic implication of subclinical or clinical mastitis prevalence was also confirmed by the studies by SEEGERS et al. (2003); HALASA et al. (2007); MIKÓ et al. (2016); PFÜTZNER ÓZSVÁRI (2016) HADRICH et al. (2018); WANI et al. (2022) and CHEN et al. (2023).

### Conclusions

The prevalence of mastitis-related problems in Holstein and Simmental cows increased with parity, with Holstein cows having a higher frequency. Various factors, including mastitis, lactation stage, age at first calving, farm size, recording season, and breeding region, had a significant effect on daily milk yield. The lowest milk yield was observed on the day of mastitis detection, followed by an increase in milk production in subsequent recordings. The increase in daily milk yield was more significant following clinical than subclinical mastitis, and was more pronounced in Holsteins than in the Simmental breed. Primiparous cows had the lowest initial increase, but they were able to regenerate more in the total period compared to the older cows with higher initial but lower total increase. Furthermore, Holstein cows had a

better production increase or recovery potential than Simmental cows, particularly after clinical mastitis. Finally, the study emphasizes the need for periodic monitoring and management of mastitis to minimize its impact on milk production, animal welfare, and farm profitability.

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### Declaration of Competing Interest

All authors declare that they have no conflicts of interest

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**GANTNER, V., I. JOŽEF, M. SAMARDŽIJA, Z. STEINER, R. GANTNER, D. SOLIĆ, K. POTOČNIK: Varijabilnost u prevalenciji subkliničkog i kliničkog mastitisa i njegov utjecaj na mliječnost krava holštajnske i simentalke pasmine kao rezultat pariteta. Vet. arhiv 94, 269-284, 2024.**

#### **SAŽETAK**

U istraživanju je analizirano više od 10 milijuna zapisa na kontrolni dan, prikupljenih između siječnja 2005. i prosinca 2022. kako bi se utvrdila prevalencija subkliničkog i kliničkog mastitisa u krava holštajnske i simentalke pasmine te njegov utjecaj na kasniju proizvodnju mlijeka. Dnevni broj somatskih stanica (SCC) služio je kao pokazatelj subkliničkog (200 000/ml do 400 000/ml) i kliničkog (>400 000/ml) mastitisa. Utvrđeno je da se problemi povezani s mastitisom povećavaju s paritetom i da su češći u holštajnske nego u simentalke pasmine. Utvrđeno je da na dnevnu količinu mlijeka utječu različiti čimbenici, uključujući stadij laktacije, dob prvog teljenja, veličinu farme, sezonu, te regiju uzgoja. Također je utvrđeno da subklinički i klinički mastitis imaju značajan utjecaj na proizvodnju mlijeka. Na dan kada je otkriven subklinički/klinički mastitis (D-0) zabilježena je najniža mliječnost, dok pri sljedećim kontrolama mliječnosti slijedi povećanje proizvodnje mlijeka. Povećanje proizvodnje variralo je ovisno o obliku mastitisa, pasmini krava (holštajnska ili simentalke) i paritetu krava. Nadalje, pri prvoj sljedećoj kontroli mliječnosti (A-1) utvrđen je najveći porast dnevne količine mlijeka kod obje pasmine i u svim laktacijama. Porast dnevne mliječnosti bio je značajniji nakon kliničkog nego subkliničkog mastitisa te je bio izraženiji u holštajnske nego u simentalke pasmine. Nakon početnog povećanja proizvodnje mlijeka, kod gotovo svih krava došlo je do pada mliječnosti u narednom razdoblju nakon subkliničkog te kontinuiranog porasta nakon kliničkog mastitisa. Analiza mjesečnih i ukupnih razlika u proizvodnji mlijeka pokazuje da su krave imale najveći mjesečni porast proizvodnje mlijeka u prvom mjesecu nakon subkliničkog/kliničkog mastitisa. Najveće ukupno povećanje proizvodnje mlijeka zabilježeno je kod prvotelke holštajnske pasmine, koje je nakon kliničkog mastitisa iznosilo 44,91 kg. U usporedbi s holštajnskom pasminom, simentalke krave imale su manji porast proizvodnje mlijeka. Općenito, holštajnske krave imale su viši porast proizvodnje ili potencijal oporavka od simentalčkih krava, osobito nakon kliničkog mastitisa. Prvotelke holštajnske i simentalke pasmine u drugom paritetu pokazale su najveći ukupni porast proizvodnje. Analiza sposobnosti oporavka životinja otkrila je da su prvotelke imale najmanji početni porast, ali su se mogle više regenerirati u ukupnom razdoblju u usporedbi sa starijim kravama s većim početnim, ali manjim ukupnim porastom.

**Cljučne riječi:** mliječne krave; subklinički i klinički mastitis; prevalencija; učinak; proizvodnja mlijeka

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