Mechanical Engineering Advances 2024, 2(2), 1752.
https://doi.org/10.59400/mea1752

Article

Influence of the new wavy teat liner "Stimulor StressLess" on milk yield performance and its quality in dairy cows: Results of a field study

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CITATION

Kaskous S, Al-Najjar K, Pfaffl MW. Influence of the new wavy teat liner "Stimulor StressLess" on milk yield performance and its quality in dairy cows: Results of a field study. Mechanical Engineering Advances. 2024; 2(2): 1752. https://doi.org/10.59400/mea1752

ARTICLE INFO

Received: 20 September 2024
Accepted: 31 October 2024
Available online: 21 November 2024

Abstract: The ideal milking system meets the physiological needs of dairy cows to increase milk yield, achieve better milk quality and maintain healthy udders. Therefore, the settings of the milking machine and the properties of the teat cup liners are very important on dairy farms. The aim of the present study was to test a new teat cup liner "Stimulor StressLess" (SSL) in two commercial dairy farms and to investigate its influence on daily milk production and quality having different experimental settings. For this purpose, 40 dairy cows of different breeds in Tirol, Austria (farm 1) were investigated for 6 months, where 3 months represent the control phase (Gr 1) and milked with conventional teat liners, and the second 3 months phase (Gr 2) was the experimental phase and milked with SSL teat cup liners. On the second farm 90 dairy cows of Simmental breed in Baden-Württemberg, Germany (farm 2) were examined for one year equally divided in the first 6 months of control phase and second 6 months of treatment phase. All cows on both farms had the same stage of lactation and lactation number. During the study period, the daily milk production of each cow was recorded and milk samples were collected to determine the ingredients. The results showed that higher daily milk production and better milk quality were observed after using the new SSL teat cup liner in the existing milking machines. However, the use of the new teat cup liner SSL was more efficient in the longer treatment in farm 2 than in farm 1. In addition, after the use of the new SSL teat cup liner, the udders remained healthy throughout the study period, showing lower somatic cell counts (SCC). It can be concluded that high milk yield and better milk quality can be achieved by using SSL teat cup liners, as they are adapted to all teat shapes and dimensions.

Keywords: dairy cows; fat; lactose; liner; protein; SCC; Stimulor StressLess; teat cup; urea

1. Introduction

The aim of the milking machine is to remove the milk completely and quickly from the udder and to keep the cows healthy. To optimally fulfil this task, the characteristics of the milking machine and the milking process play an important role [1]. Many studies have shown that the shape of teat cup liners and milking machine settings have a significant impact on milking performance, milk quality, milking time, and udder health [2–7]. The teat cup liner is the sole component of the milking machine that is in direct contact with the animal [8]. Therefore, it must perfectly match the dimensions and shape of the teats in term of size, fit and material properties to ensure the best possible performance for long-term safe and efficient milking [9]. In addition, the head piece should ensure a good seal with the teat base without restricting the connection between the mammary gland and teat cistern [10]. However, the dimensions of the teat cup liners must be tailored to the size of the teat so that they can

be massaged effectively [11]. The most important of these measurements is the depth of the liner mouthpiece. Several pieces of evidence suggest that incorrectly used teat cup liners can lead to the following problems: 1) Adhesion problems of the teat cups, resulting in air slurping and falling off, 2) Increasing restlessness of the animals during milking, 3) Incomplete emptying of the udder, 4) Significant extension of milking time and, 5) Acute disorders of the teat condition after milking [6,9,10,12–14]. In addition, selecting the appropriate teat liner as well as the correct vacuum level and pulsation settings is very important to achieve the basic goals of fast, gentle, and complete milking [11,15]. There is an almost optimal milking machine available that corresponds to the physiological, anatomical, and morphological characteristics of the cow's udder. Its name is MultiLactor (ML) (Siliconform, Germany). ML is a semi-automatic milking system that is technically different from conventional milking machines, based on a quarter-individual milking system compared to conventional milking machines with claw piece [16–19]. In addition, ML is more efficient than conventional milking systems in terms of the positive stimulation effect, because ML has an established stimulation program optimize to the physiological needs of the milking cow [20]. The working vacuum level in the ML is 34 kPa compared to the conventional milking system with 45 kPa [17]. Furthermore, it is noteworthy that ML has an excellent cleaning and disinfection system after milking each cow and after milking all cows, compared to traditional milking machines that are only cleaned after milking all cows [21].

The teats of the cows in a herd have usually different sizes and shapes. Many farmers make a compromise when choosing teat cup liners for their cows. This can mean that teat cup liners seal too tightly on large and wide teats, while allowing air to enter on small and slim teats. Therefore, it is a challenge to find the right one with the best teat fitting [22]. However, there is currently a new teat cup liner on the market for all teat shapes and sizes (Stimulor StressLess, SSL, Siliconform, Germany) [6]. The wave-shaped construction of the SSL features an adaptive lip. This means that different teat sizes can be milked with the same teat liner [23]. This prevents excessive head vacuum, stops the sliding up of the teat cup and reduces stress on the teat tissues [24]. In other words, they are ideal for maintaining teat health during milking for all lactating cows. In this context, the objective of this study was to investigate the influence of liner design "Stimulor StressLess" in routine milking in two commercial dairy farms. We tested the hypothesis whether the new teat liner design affects udder health as well as milking performance and therefore daily milk yield and milk quality.

2. Material and methods

The experiments were carried out on commercial dairy farms in Tirol, Austria (farm 1) and Baden-Württemberg, Germany (farm 2), which is why the requirements of the animal protection laws of both countries were adhered to. The farms owners were informed about experimental procedures and gave informed consent.

2.1. Animals and housing

Farm 1: 40 dairy cows of different breeds (Brown Swiss, Simmental, and Holstein) were examined in two equal phases (called Gr 1 and Gr 2) of 3 months. The first phase of 3 months (Gr 1) was the control phase where animals were milked with MultiLactor milking system (Siliconform, Germany) and normal teat cup liner. The second phase, also 3 months (Gr 2), was the experimental phase, where animals were milked with MultiLactor milking system and SSL teat cup liners (**Figure 1**). Both phases were similar at the start of the experiment in term of milking day, number of lactations and daily milking performance (**Table 1**).

Table 1. Average daily milk yield, average days in milk and, average number of lactations in (Gr 1) and (Gr 2) of the dairy cows examined at baseline in farm 1.

Parameters farm 1	**Control phase (Gr 1)**	**Experimental phase (Gr 2)**
Number of animals	40	40
Daily milk yield (kg/day)	21.56	21.61
Days in milk (day)	164.96	162.15
Lactation number	2.60	2.63

The evaluation took place between July and December 2022. Both groups were subjected to the same nutritional and environmental conditions, housed in a free stall system. The supply of diet was grass-and corn-silage, hay, and concentrate, where all nutrients were adapted to the cow's lactation stage and milk yield. The ration mixture consisted of the following feedstuffs: grass silage: 45%, maize silage: 28%, hay: 14%, straw: 4%, rapeseed meal: 6%, grain maize: 2% and minerals: 0.9%. The water was provided ad libitum, and the chemical composition of the diet supplied to dairy cows met the required nutritional standards [25]. The animals were proved to be healthy throughout the course of the experiment.

Farm 2: 90 dairy cows of the Simmental breed were used and examined in two phases (Gr 1, Gr 2), like in farm 1. The first phase (6 months) (Gr 1) was the control phase and the animals were milked with a conventional milking system and normal teat cup liners. The second phase (6 months) (Gr 2) was the experimental phase and the animals were milked with a conventional milking system and SSL teat cup liners. Both animal phases were similar at the start of the experiment in terms of milking day, number of lactations and daily milking performance (**Table 2**).

Table 2. Average daily milk yield, average days in milk and, average number of lactations in Gr 1 and 2 of the dairy cows examined at baseline in farm 2.

Parameters farm 2	**Control phase (Gr 1)**	**Experimental phase (Gr 2)**
Number of animals	90	90
Daily milk yield (kg/day)	20.69	20.64
Days in milk (day)	140.02	138.73
Lactation number	3.03	3.05

The experiment took place between September 2022 and August 2023. The cows were kept in a loose housing system. All animals received the same compound feed

ration (grass-and corn-silage, hay, and concentrate) depending on performance and maintenance requirements [25]. The ration mixture consisted of the following feedstuffs: grass silage: 38%, maize silage: 35%, hay: 11%, straw: 5%, rapeseed meal: 7%, grain maize: 3% and minerals: 0.9%. Drinking water was administered ad libitum. The animals were proved to be healthy throughout the course of the experiment.

2.2. Milking equipment

2.2.1. Properties of the teat liner "Stimulor StressLess" used in both farms

The new teat liners "Stimulor StressLess" have unique properties that are excellent for maintaining teat health during machine milking in all dairy cows [6]. One of its distinguishing features is the presence of a wave-shaped design of its mouthpiece, which allows it to adapt well to the different teat sizes in a herd, thus ensuring consistent milking of the entire herd. The wave designed of the lip reacts to the pressure difference in the liner and, if necessary, allows the outside air inlet to flow in to compensate. This prevents excessive head vacuum and reduces tissue stress. In addition, with this liner, the head vacuum will be kept stable in the physiological range, and the teats adhere particularly well and do not cause any slurry noise during milking. For dairy cows, this means protection of the sensitive teat tissue and more comfort when milking (**Figure 1**) [1].

Figure 1. Teat cup liners (Stimulor StressLess) used by the two farms for dairy cows.

2.2.2. Milking equipment and milking routine on farm 1

The experimental dairy cows (both phases) were milked twice daily (starting 5 AM and 4 PM) in a tandem milking parlour (2 × 3 places) equipped with MultiLactor (ML) milking system (Siliconform, Germany), based on a quarter-individual milking equipment. This means that teat cups work completely independently of each other (without a claw) (**Figure 2**). The working "vacuum" level was 36 kPa and sequential pulsation (25% offset quarter to quarter) was adopted. The pulsation rate was 60 cycles per minute and the pulsation ratio was 60/40 during the milking time. The milking process took place in both phases according to the usual routine on the farm. At the time of milking with ML the milking routine started with pre-milking preparations, which consisted of fore-stripping of one or two squirts of milk from each teat and then cleaning the udder and teats. After that, the milking unit swing directly in front of the

cow's udder. The teats cups were pulled out of the storing individually or in pairs and manually attached to the teats. After this step, the system is started on the control display and the pre-stimulation began. The pre-stimulation is structured to be intensively activated with a normal pulse rate (60 cycles/min) and reduced the milking phase (b-phase) of 10% over a period of 50 s. At the same time, intensive movement of the teats cups is regulated as an additional stimulation by an actuator. This is an arm on which four milk tubes lie. During the pre-stimulation and the milking time, this arm moves up and down. This movement is transferred to the teats cups and vibrate the teats. When the milk flow reaches 250 g/min during milking, then the milking process automatically ends with the detachment of the milking unit and each teat is dipped with a solution containing Chlorhexidin. At the same time teat cups are cleaned and disinfected automatically with water and per acetic acid solution (0.5%) after each cow and milking.

(A) **(B)**

Figure 2. Milking parlor. **(A)** Milking cup with SSL teat liner; **(B)** multiLactor milking machine (Siliconform, Germany).

2.2.3. Milking equipment and milking routine on farm 2

Experimental dairy cows (both phases) were milked twice daily (starting 6:30 AM and 4:30 PM) in a steeply herringbone milking parlour (60 degrees) (2 × 10 places) equipped with conventional milking system. In this milking system, each milking unit consists of four teat cups and a claw piece. The working "vacuum" level was 40 kPa and alternating cyclic pulsation between the front and back were adopted. The pulsation rate was 60 cycles per minute and the pulsation ratio was 60/40 during the milking time. The milking process were performed according to the usual routine of the farm. At the time of milking the milking routine started with pre-milking preparations, which consisted of fore-stripping of one or two squirts of milk from each teat and then cleaning the udder and teats. After that, the teat cups were manually attached to the teats. The milking process was observed and if no milk came, the teat cup was removed and milking stopped. Each teat was then dipped in a chlorhexidine solution. It is noteworthy that in the first phase (Gr 1), in some animals, an additional piece was attached to the teat cup to prevent the teat cup from sliding up.

2.3. Recording of milk yield, taking of milk samples and analysis

Milk yield was recorded and milk samples (50 ml bottle) were collected monthly for qualitative analysis in both farms and groups Gr 1 and Gr 2 from each cow during

the study period. The milk samples were preserved and then, they sent to the laboratory. The milk composition was determined by the Tiroler State control association, Innsbruck, Austria (farm 1) and in LKV Baden-Württemberg, Hohenstein, Germany (farm 2). The absolute values of fat (%), protein (%) and lactose (%) in the milk were determined by infrared-spectroscopy (MilkoScan, Foss, Denmark) and the somatic cell count (SCC) was determined by fluorescence optical counting (Fossomatic, Foss, Denmark). An infrared measurement with partial least-squares (PLS) calibration model was used to determine the urea in the milk. It should be mentioned at this point that no lactose was determined in the milk from farm 1.

2.4. Statistical analysis

Program SAS 9.3 was used for statistical evaluation in both farms [26]. GLM was used to determine the effect of the teat liner type on daily milk production, composition of cows according to the following linear models:

Model 1: $Y_{ij} = \mu + G_i + e_{ij}$, where, Y_{ij} =daily milk (kg), fat (%), protein (%), SCC, lactose (%), and urea of ij^{th} record. μ = overall mean. G_i = effect of i^{th} teat liner type coded as i = 1 (normal teat liner), and i = 2 (SSL teat liner). e_{ij} = random error term associated with Y_{ij} observations with zero mean and variance $I\sigma^2$e.

Model 2: $Y_{ij} = \mu + G_i(Ls)_j + e_{ij}$, where, $(Ls)_j$ = effect of j^{th} stage of lactation within teat liner type coded as j = 1 (< 100 days), j = 2 (101–200 days), and j = 3 (> 200 days).

Model 3: $Y_{ij} = \mu + G_i(Par)_j + e_{ij}$, where, $(Par)_j$= effect of j^{th} parity within teat liner type

coded as j = 1, 2, ..., and 6 for first, second, ... and sixth, respectively.

Model 4: $Y_{ij} = \mu + G_i(scl)_j + e_{ij}$, where, $(scl)_j$ = effect of j^{th} somatic cell counts classes within teat liner type coded as j = 1 (< 50×10^3), 2 (50×10^3–100×10^3), 3 (101×10^3–150×10^3), 4 (151×10^3–200×10^3), 5 (201×10^3–250×10^3), and 6 (> 250×10^3). Since the number of animals on farm 1 was too small, the following somatic cell count classes were carried out. 1 (< 50×10^3), 2 (50×10^3–100×10^3), 3 (101×10^3–150×10^3), 4 (151×10^3–300×10^3), 5 (> 300×10^3).

To determine significant differences between least square means (LSM) of the group effects, Duncan's multiple range test was used. The results were presented as LSM ± SE.

3. Results

Since different milking systems were used on farm 1 and 2 in this field study, the results were treated separately for each farm.

3.1. Farm 1

3.1.1. General mean values of the examined parameters in Gr 1 and Gr 2

The results in **Table 3** showed that the daily milk yield in Gr 2 increased compared to the milk production in Gr 1 during the study period, but the increase was not significant ($P > 0.05$). Conversely, it was shown that the fat (%) and protein (%) increased significantly ($P < 0.001$) after using the new SSL teat cup liner (Gr 2) in

comparison to Gr 1. The values were 4.31 ± 0.06% versus 4.03 ± 0.06% for fat and 3.83 ± 0.04% versus 3.51 ± 0.04% for protein in Gr 2 and Gr 1, respectively. It is noteworthy that after using a new SSL teat cup liner, the SCC in the milk decreased from 126.69 ± 23.24 × 10^3 to 95.06 ± 12.97 × 10^3 in Gr 1 and Gr 2, respectively, but the difference was not significant. According to the urea content in the milk, after using the new SSL teat cup liner (Gr 2), it was in the ideal concentration (15.03 ± 0.54 mg/dl) compared to Gr1 (24.98 ± 0.78 mg/dl).

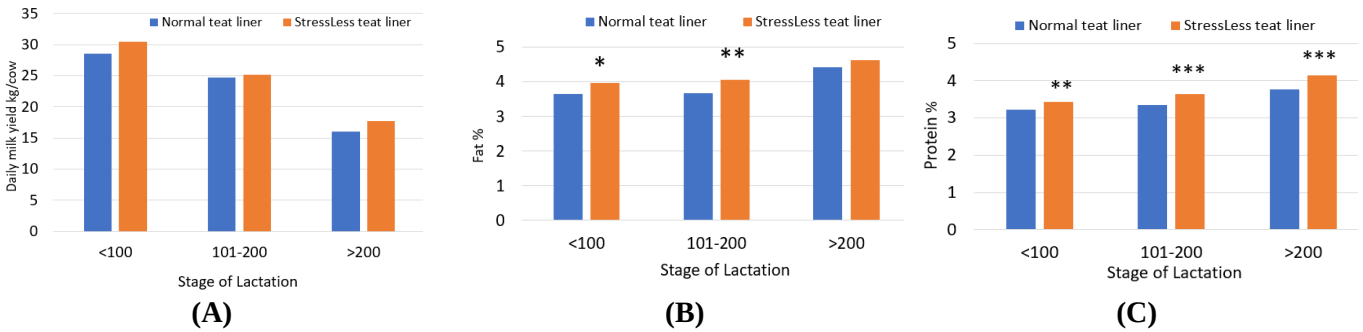
Table 3. The average (LSM ± SE) of the milk parameters of the examined farm 1 during the investigation period.

Parameters farm 1	Normal teat liner (Gr 1)	StressLess teat liner (Gr 2)	Significance (*p*-value)
Milk yield (kg/day/animal)	21.57 ± 0.66	22.95 ± 0.61	0.1259
Fat (%)	4.03 ± 0.06	4.31 ± 0.06	0.0001
Protein (%)	3.51 ± 0.04	3.83 ± 0.04	0.0001
SCC (× 10^3 cells/ml)	126.69 ± 23.24	95.06 ± 12.97	0.2214
Urea (mg/dL)	24.98 ± 0.78	15.03 ± 0.54	0.0001

Gr 1: Milking the dairy cows with normal teat cup liners. Gr 2: Milking the dairy cows with SSL teat cup liners.

3.1.2. Examined milk parameters in Gr 1 and Gr 2 according to the stage of lactation

Figure 3 shows the tested milk parameters in Gr 1 and Gr 2 based on the lactation stage in farm 1. In all stages of lactation, daily milk production was numerically higher in Gr 2 than in Gr 1 after using the new teat liners (SSL). However, the difference was not significant ($P > 0.05$) (**Figure 3A**). After using a new SSL teat cup liner, a significant increase in milk fat % was observed in Gr 2 compared to Gr 1, in the first and second stages of lactation ($P < 0.05$; $P < 0.01$, respectively) (**Figure 3B**). Similar results were clearly observed for milk Protein (%), which was higher in Gr 2 than in Gr 1 in all lactation stages, and the difference was highly significant ($P < 0.001$) (**Figure 3C**). Regarding the urea content of the milk, it was found that after using the new SSL teat cup liner (Gr 2) a normal urea content (approx. 15 mg/dl) was observed compared to that during Gr 1 (**Figure 3D**). Although the SCC in Gr 2 was lower than in Gr 1 after using the new SSL teat liner, no significant difference was observed ($P > 0.05$) (**Figure 3E**).



(A) **(B)** **(C)**

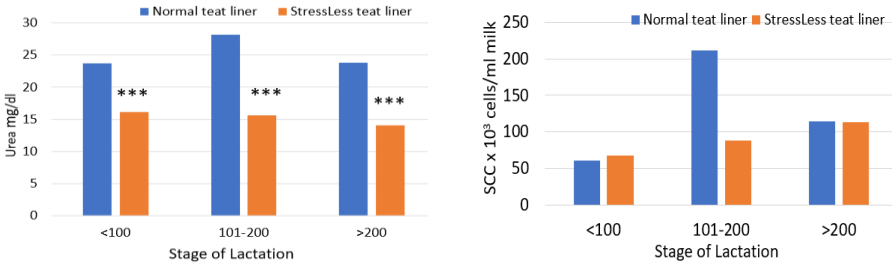


Figure 3. Average (LSM) milk parameters in Gr 1 (normal teat cup liner) and Gr 2 (SSL teat cup liner) with significant differences (*p*-value): * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$, according to the stage of lactation in farm 1. **(A)** Daily milk yield (kg/cow); **(B)** fat (%); **(C)** protein (%); **(D)** urea (mg/dl); **(E)** somatic cell count (SCC) (× 10^3 cells/ml).

3.1.3. Examined milk parameters in Gr 1 and Gr 2 according to the lactation numbers

Table 4 showed that the daily milk production after using a new teat liner (SSL) in Gr 2 compared to Gr 1 was increased. However, the significant increase was only observed in the sixth lactation and above ($P < 0.05$). Interestingly, the fat content (%) in milk increased significantly in the first, third and fifth lactations in the Gr 2 compared to Gr 1. It is noteworthy that the protein content (%) of milk increased significantly in Gr 2 compared to Gr 1 at all lactation numbers ($P < 0.05$). Current results on the cell count in milk after use of a new teat liner (Gr 2) showed no significant differences ($P > 0.05$) in all lactation numbers compared to (Gr 1). The results for the urea concentrations in the milk showed better values after using new teat cup liner (Gr 2) compared to Gr 1. However, the difference was significant only in the first, second and third lactation numbers.

Table 4. The average (LSM ± SE) milk parameters of the examined farm 1 in the investigation period according to lactation numbers and significances (*p*-values).

Parameters	**Group**	**Lactation number**					
		1 (29)*	**2 (25)**	**3 (21)**	**4 (18)**	**5 (14)**	**≥ 6 (13)**
Milk yield (kg/day/cow)	Gr 1	15.49 ± 0.69	21.44 ± 0.93	25.73 ± 0.93	24.07 ± 0.58	23.80 ± 0.88	22.93 ± 0.82
	Gr 2	16.19 ± 0.48	22.09 ± 0.88	26.40 ± 0.89	25.14 ± 0.57	25.15 ± 0.74	24.45 ± 0.74
significance		0.4155	0.6136	0.6311	0.6592	0.0659	0.0256
Fat (%)	Gr 1	4.16 ± 0.14	4.12 ± 0.09	3.72 ± 0.11	4.07 ± 0.12	4.23 ± 0.22	3.73 ± 0.10
	Gr 2	4.77 ± 0.17	4.28 ± 0.09	4.21 ± 0.09	4.68 ± 0.29	4.71 ± 0.03	4.03 ± 0.16
significance		0.0134	0.2235	0.0009	0.1747	0.0496	0.2145
Protein (%)	Gr 1	3.52 ± 0.52	3.57 ± 0.06	3.37 ± 0.05	3.51 ± 0.10	3.35 ± 0.06	3.52 ± 0.06
	Gr 2	4.11 ± 0.09	3.84 ± 0.07	3.71 ± 0.06	4.06 ± 0.13	4.02 ± 0.03	3.87 ± 0.02
significance		0.0001	0.0048	0.0004	0.0282	0.0001	0.0013
SCC (× 10^3 cells/ml)	Gr 1	92.33 ± 15.31	111.28 ± 31.19	132.04 ± 36.67	126.67 ± 21.73	134.67 ± 22.10	167.67 ± 19.91
	Gr 2	84.33 ± 11.83	78.09 ± 13.42	115.20 ± 32.70	86.00 ± 16.37	90.25 ± 5.34	153.50 ± 16.80
significance		0.7348	0.3277	0.7468	0.1952	0.1721	0.6166

Table 4. (*Continued*).

Parameters	Group	Lactation number					
		1 (29)*	**2 (25)**	**3 (21)**	**4 (18)**	**5 (14)**	**≥ 6 (13)**
Urea (mg/dl)	Gr 1	21.22 ± 1.76	25.83 ± 0.10	24.25 ± 1.74	32.67 ± 1.84	26.33 ± 0.38	26.33 ± 0.13
	Gr 2	10.67 ± 2.00	15.12 ± 0.79	15.52 ± 0.90	18.20 ± 2.06	18.00 ± 3.49	11.00 ± 2.65
significance		0.0012	0.0001	0.0001	0.0848	0.1568	0.0718

Gr 1: Milking the dairy cows with normal teat cup liners. Gr 2: Milking the dairy cows with SSL teat cup liners. * The number in brackets indicates the number of milk samples in each group within each lactation number.

3.1.4. Somatic cell classes in Gr1 and Gr2 during the study period

The results of the statistical analysis clearly showed that after using the new teat cup liner, the cell count content of the milk was lower compared to before (**Figure 4**). This means that 80% of the milk samples after using the new teat cup liner (Gr 2) were below 100 × 10^3 cells/ml compared to Gr 1 (69%). Furthermore, it was observed that after using the new teat cup liner, only 12.48% of milk samples were above 150 × 10^3 cells/ml compared to Gr 1 (18.26%) (**Figure 4**).

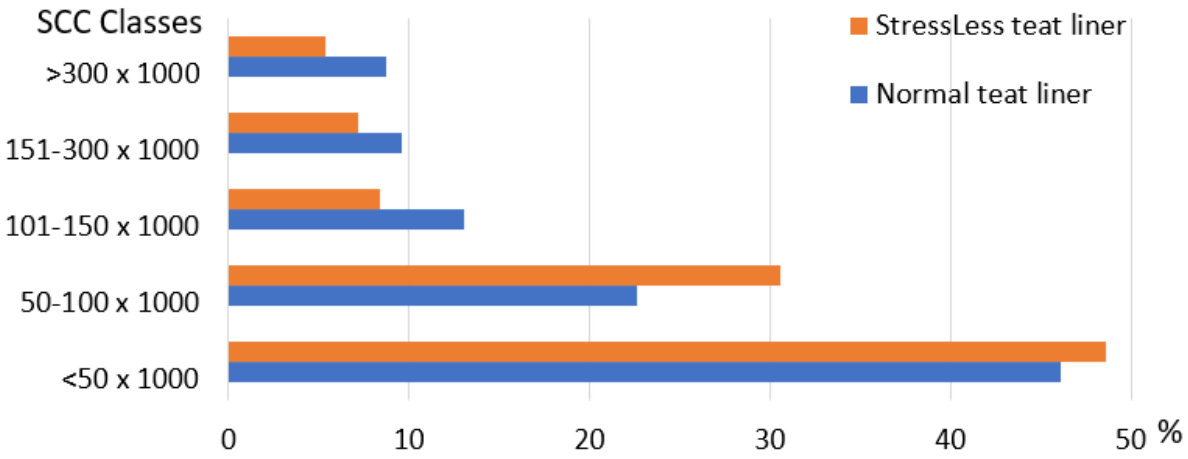


Figure 4. Average somatic cell classes (cells/ml) before and after using the SSL new teat cup liner in all dairy cows examined in farm 1.

3.2. Farm 2

3.2.1. General mean values of the examined parameters in Gr 1 and Gr 2

The results in **Table 5** clearly showed that the daily milk yield in Gr 2 increased significantly ($P < 0.001$) when using the new teat cup liner StressLess and the value has increased by around 2.0 kg per animal. The average fat concentration of dairy cows in Gr 1 was 3.91 ± 0.03% and increased significantly ($P < 0.01$) to 4.04 ± 0.03% in Gr 2. The same trend was observed in protein content and the values increased from 3.22 ± 0.01% in Gr 1 to 3.42 ± 0.02% in Gr 2 and the difference was highly significant ($P < 0.001$). It is noteworthy that the SCC in milk in Gr 2 decreased significantly ($P < 0.001$) compared to Gr 1 and the values were 99.66 ± 3.45 × 10^3 cells/ml and 121.54± 3.39 × 10^3 cells/ml respectively. An interesting aspect was the lactose concentration in dairy cows on this farm and the values increased significantly ($P < 0.001$) from 4.73 ± 0.01% in Gr 1 to 4.81 ± 0.01% in Gr 2. In addition, the urea concentration also

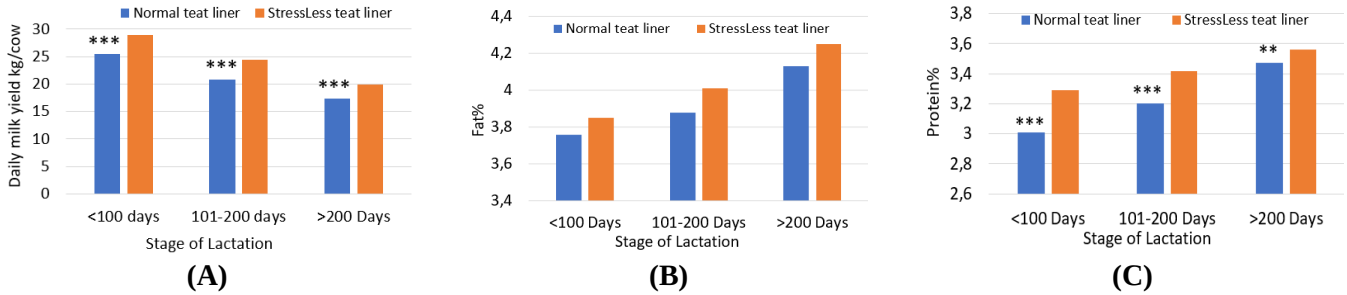
increased after using the new teat cup liner and the values were 12.21 ± 0.30 mg/dl and 14.30 ± 0.32 mg/dl in Gr 1 and 2, respectively.

Table 5. The average (LSM ± SE) of the milk parameters of the examined farm 2 during the investigation period.

Parameters farm 2	Normal teat liner (Gr 1)	StressLess teat liner (Gr 2)	Significance (*p*-value)
Milk yield (kg/day/cow)	21.21 ± 0.27	23.30 ± 0.27	0.001
Fat (%)	3.91 ± 0.03	4.04 ± 0.03	0.004
Protein (%)	3.22 ± 0.01	3.42 ± 0.02	0.001
SCC (× 10^3 cell/ml)	121.54 ± 3.39	99.66 ± 3.45	0.001
Lactose (%)	4.73 ± 0.01	4.81 ± 0.01	0.001
Urea (mg/dl)	12.21 ± 0.30	14.30 ± 0.32	0.001

3.2.2. Examined milk parameters in Gr1 and Gr2 according to the stage of lactation

Figure 5 shows the tested milk parameters in Gr 1 and Gr 2 related to the lactation stage. In all lactation stages, daily milk yield was in Gr 2 significantly higher than in Gr 1. The percentage increases in Gr 2 in lactation stages 1, 2 and 3 were 14%, 17% and 14%, respectively (**Figure 5A**). Similar results were shown for protein (%) (**Figure 5C**). Despite an increase in the percentage fat content in Gr 2 compared to Gr 1 after using the SSL teat liner, no significant difference was found ($P > 0.05$) (**Figure 5B**). **Figure 5D** clearly showed an increase in lactose content (%) in the milk of Gr 2 compared to Gr 1 in the second and third stages of lactation ($P < 0.001$). However, the increase in lactose content (%) in the first 100 days of lactation in Gr 2 was not significant compared to Gr 1. Furthermore, it has been shown, that SCC in Gr 2 was numerically lower than in Gr 1 in all lactation stages and the difference was highly significant in the third lactation stage (103.12 ± 6.03 versus 146.85 ± 6.06; $P < 0.001$) (**Figure 5F**). It is noteworthy that urea content in the milk increased significantly after using the new StressLess teat liner in Gr 2 compared to Gr 1 ($P < 0.001$) in lactation stages 1 and 2. The values were 14.91 ± 0.59 compared to 11.46 ± 0.39 and 15.08 ± 0.64 compared to 12.61 ± 0.37 in lactation stages 1 and 2 respectively (**Figure 5E**).



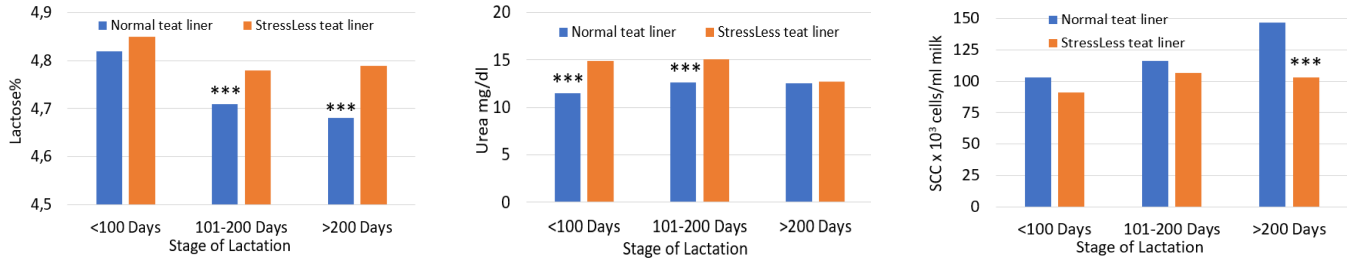


Figure 5. Average (LSM) milk parameters in Gr 1 (normal teat cup liner) and Gr 2 (SSL teat cup liner) with significant differences (*p*-value): * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$, according to the stage of lactation in farm 2. **(A)** Daily milk yield (kg/cow); **(B)** fat (%); **(C)** protein (%); **(D)** lactose (%); **(E)** urea (mg/dl); **(F)** somatic cell count (SCC) (× 10^3 cells/ml).

3.2.3. Examined milk parameters in Gr 1 and Gr 2 according to the lactation numbers

Table 6 shows that all tested parameters were improved after using the SSL teat cup liner in all lactation numbers. Daily milk production was higher in Gr 2 than in Gr 1 at all lactation numbers and the difference was significant except in the fourth lactation. Milk fat content (%) was numerically increased at all lactation numbers after using the new teat cup liner, but it was only significant in the sixth lactation and above. Statistical analysis clearly showed that after using new teat cup liners (Gr 2), the protein content (%) of milk increased significantly at all lactation numbers compared to Gr 1. Similar results were shown for lactose content (%) of milk. Based on the SCC content in milk after using a new teat cup liner (Gr 2), it was lower than Gr 1 for the first and second lactation. The results in **Table 6** also showed that the urea content in milk in Gr 2 was higher than that before using SSL teat cup liners (Gr 1). However, the differences between Gr 1 and Gr 2 were significant only in the first, second, third and sixth and above lactations.

Table 6. The average (LSM ± SE) of the milk parameters of the examined farm 2 in the investigation period according to lactation numbers and significances (*p*-values).

Parameters	**Groups**	**Lactation number**					
		1 (120)*	**2 (110)**	**3 (90)**	**4 (85)**	**5 (80)**	**≥ 6 (55)**
Milk yield (kg/day)	Gr 1	17.81 ± 0.44	20.70 ± 0.46	22.43 ± 0.62	25.13 ± 0.71	23.87 ± 0.75	22.37 ± 0.50
	Gr 2	20.12 ± 0.50	23.38 ± 0.52	24.61 ± 0.71	26.49 ± 0.75	25.06 ± 0.60	24.15 ± 0.64
significance		0.001	0.001	0.004	0.281	0.005	0.001
Fat (%)	Gr 1	3.96 ± 0.06	4.01 ± 0.06	3.93 ± 0.09	3.98 ± 0.09	3.80 ± 0.14	3.65 ± 0.08
	Gr 2	4.02 ± 0.06	4.10 ± 0.06	4.13 ± 0.08	4.21 ± 0.10	3.91 ± 0.10	3.89 ± 0.07
significance		0.456	0.270	0.109	0.094	0.540	0.024
Protein (%)	Gr 1	3.17 ± 0.03	3.23 ± 0.03	3.33 ± 0.04	3.20 ± 0.04	3.20 ± 0.07	3.13 ± 0.04
	Gr 2	3.39 ± 0.03	3.49 ± 0.03	3.48 ± 0.04	3.41 ± 0.05	3.37 ± 0.05	3.37 ± 0.03
significance		0.001	0.001	0.014	0.001	0.012	0.001

Table 6. (*Continued*).

Parameters	Groups	Lactation number					
		1 (120)*	**2 (110)**	**3 (90)**	**4 (85)**	**5 (80)**	**≥ 6 (55)**
SCC (× 10^3 cells/ml)	Gr 1	88.37 ± 6.66	129.32 ± 7.00	133.16 ± 9.45	135.17 ± 10.92	151.40 ± 11.48	128.38 ± 7.58
	Gr 2	69.16 ± 5.85	78.42 ± 6.10	132.35 ± 8.24	132.12 ± 8.70	151.77 ± 13.38	114.18 ± 7.45
significance		0.028	0.001	0.952	0.836	0.982	0.184
Lactose (%)	Gr 1	4.84 ± 0.02	4.74 ± 0.02	4.68 ± 0.03	4.70 ± 0.03	4.67 ± 0.03	4.69 ± 0.02
	Gr 2	4.90 ± 0.01	4.80 ± 0.01	4.79 ± 0.02	4.74 ± 0.02	4.76 ± 0.03	4.75 ± 0.02
significance		0.006	0.009	0.001	0.234	0.031	0.060
Urea (mg/dl)	Gr 1	12.68 ± 0.44	12.10 ± 0.46	12.02 ± 0.62	11.83 ± 0.72	13.66 ± 0.75	11.39 ± 0.50
	Gr 2	14.57 ± 0.70	14.42 ± 0.73	14.60 ± 0.10	13.71 ± 1.04	13.55 ± 1.60	14.10 ± 0.89
significance		0.030	0.005	0.025	0.121	0.947	0.009

Gr 1: Milking the dairy cows with normal teat cup liners. Gr 2: Milking the dairy cows with SSL teat cup liners. * The number in brackets indicates the number of milk samples in each group within each lactation number.

3.2.4. Somatic cell classes in Gr 1 and Gr 2 during the study period

It was found that 60% of SCC in milk after using a new teat cup liner had less than 100 × 10^3 cells/ml, compared to only 45% for Gr1 (**Figure 6**). An interesting aspect is that after using the new teat cup liner, only 4.25% of milk samples were above 250 × 10^3 cells/ml, compared to 7.76% in Gr 1.

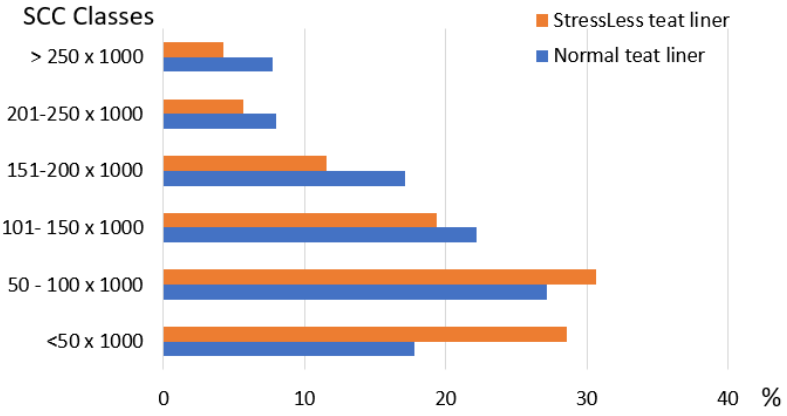


Figure 6. Average somatic cell classes (cells/ml) before and after using the SSL new teat cup liner in all dairy cows examined.

4. Discussion

This study in the two commercial dairy farms clearly showed that higher daily milk production, better milk quality and less SCC were observed after using the new SSL teat cup liner in the existing milking machines. In any case, it has been shown that the SSL teat cup liner plays an important role in optimal milk removal during the milking process. It was observed that a well-designed milking machine harvests milk quickly, gently, and maintains the health of the animals' udders and is easy to clean and disinfect [7,13]. Thus, for optimal milk removal, the continuous availability of milk from the cluster attachment to the point of extraction is crucial. Based on that, the liner plays an important and central role in this as its movement provides

continuous stimulation during milking, resulting in oxytocin release from the pituitary gland and continuous milk ejection until the teat cups are removed [27,28]. This process works well and optimally if, in addition to the properties of the milking machine, the teat cup liner used is tailored to the dimensions of the udder teats [1]. Notably, if the dimensions between the teat and teat cup liner are different, the teat cups can climb up, which can lead to an obstruction of the milk flow by disability the connection between the gland cistern and the teat cistern [8]. It should be noted that the shape of the teats and teat dimensions vary greatly within different dairy cows of the same herd, since dairy cows have been primarily are selected for their higher milking speed and for their milking parlour throughput [29,30]. This is exactly what we found during our experiment, particularly on farm 2, very different shapes, lengths, and diameters of udder teats (**Figure 7**).

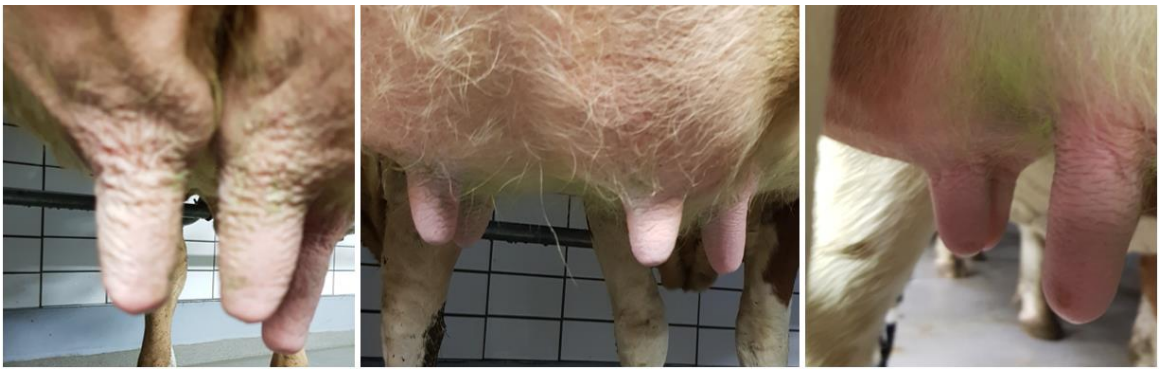


Figure 7. Some teats with different shapes, lengths, and diameters in farm 2.

From a practical point of view, every cow owner uses one type of teat cup liner for all lactating cows and he cannot use additional teat cup liners for each lactating cow with different teat sizes on the farm. Hence individual dairy cows suffered from unsuitable fitting teat liners. Normally, the selected teat cup liner size only fits optimally for around 80% of the lactating cows in a herd [31]. This means that the remaining 20% of cows have more or less problems with this chosen teat cup liner. The properties of the new SSL teat liner (presence of mouthpiece with wavy construction design) were therefore particularly noticeable in the farms examined and had a very good effect because they fit all teat shapes. With large or wide teats, the teat cup liners often fit tightly to the teats and the negative pressure of the milking machine hinders blood and lymph circulation, which puts a lot of strain on the teat tissue. This causes pain and induces mammary immune reactions by increased SCC in the milk produced, as observed in this study before using the SSL teat cup liner. These changes in the circulatory system damage the teat ends, reduce the effectiveness of the udder defense, and can cause in the long-range severe mastitis [15,32,33]. Hillerton [34] found that teat liners had a significant impact on milking performance, cow behaviour during milking, udder health and teat immune reaction. In addition, if the teats are small, air gets into the teat cup and the cluster falls off. In such situations, there may be problems with the attachment of the teat cup or poor adhesion to the teat during milking. The wave-shaped construction of the new liner used (Stimulor StressLess) has an adaptive lip. This means that with the integrated adaptive lip,

different teat sizes can be milked with the same liner [6]. These new teat cup liners do not only improve milking performance, but also positively support the well-being of dairy cows. One explanation for the increased milk yield after using the new SSL teat cup liner in our experiment is the complete emptying of the udder. It has long been known that complete emptying of the udder promotes milk synthesis and secretion, hence the milk yield increases [6]. An earlier study indicated that poor teat cup design results in cows not being fully milked [9]. Studies on the physiological relationships between complete milking and milk secretion rate show that milk secretion is controlled via local mechanisms in the udder tissue [34]. Based on the relationships described by Wilde and Peaker [35], it can be assumed that the epithelial cells in the alveoli will reach their genetically determined secretion maximum again, after the inhibitor has been completely removed. This inhibitor is a component of whey protein that acts quickly- and concentration-dependently. In any case, complete udder emptying keeps the content of effective inhibitors in the udder low, and thus stimulates milk secretion.

Milk ingredients are an indicator of the success of the milking process and allow identifying some important management errors. Therefore, in many countries the determination of fat, protein, lactose, urea, and SCC is routinely carried out on each individual cow. Milk fat content is influenced by composition of the feed ration and physical structure of the ration [36–39]. The milking machine, the teat liner fitting and the milking process have also a significant influence on the milk fat content, as this study has shown. After using the SSL teat cup liner on both farms, the udder is completely empty after milking. As a result, the fat content in the milk increased significantly on both farms. It has long been known that the fat content of milk increases continuously during the course of milking and the last portions of milk are of the highest fat content which improves the quality of the final product [38,40,41].

For the protein content of the milk, the results of the test farms showed that it increased after using the new teat cup liner. This increase in milk protein proves that the milking process after using the new teat liner was better than that of the normal teat liner. At this point, udder health and fitness plays “the central role” in the level of milk protein content and their fraction in lactating cows [42]. Many studies have shown that a negative relationship has been demonstrated between the SCC in the milk and the protein content of milk [42,43]. This condition was clearly visible in our study after using the new teat cup liner. It means that the SCC has decreased and the protein content of the milk has increased. This could be explained by the fact that decrease in protein synthesis and DNA in epithelial cells in the udder is due to the increase in SCC in milk [43,44]. Milk from mastitis udders has been observed to have greatly increased proteolytic activity [45–47]. However, plasmin is the main protease in the milk of diseased udders with increased SCC, because the enzyme plasmin is produced by leucocytes [48,49]. It is noteworthy that casein hydrolysis in the udder begins between milking times when subclinical or clinical mastitis is present [50]. The research clearly showed that higher SCC reduces the proportion of β-Casein (β-CN) and αS1-Casein (αS1-CN), which is due to the increased activity of proteolysis [43,51].

Referring to the SCC in the milk of both farms examined clearly showed that the SCC in the milk fell below 100×10^3 cells/ml after using the new teat cup liner SSL.

However, the influence of teat cup liners on teat condition, udder health, and thus on the level of SCC is well known [1]. It should be noted that the somatic cell count in milk is the most important inflammatory parameter of the cow udder [52,53]. Many studies have shown that with prolonged use of an unsuitable milking machine or improperly fitting teat cup liners, the milking machine attacks the teat tip tissue and forms a callus ring around the teat opening, resulting in hyperkeratosis [54–57]. In addition, it has been observed that the proper functioning of the milking machine and milking hygiene are necessary for reducing the risks of mastitis, and thus reducing SCC in milk [53].

The significant increase in lactose content in milk (4.81 ± 0.01 versus 4.73 ± 0.01, $P < 0.001$, **Table 5**) after using the new SSL teat liner showed that the udders of all cows tested were healthy. This means that lactose synthesis in the udder was going well since there were no secretion disorders. It is known that in clinical or subclinical mastitis, the lactose content of the milk decreases and new synthesis is reduced [58]. However, lactose is osmotically active; it is in osmotic equilibrium with sodium and chloride ions in milk. When there is an inflammation of the udder, lactose is transported back into the blood and appears in the urine. At the same time, increased amounts of sodium and chloride ions pass from blood into the milk. Therefore, lactose is very important in maintaining the osmotic pressure of milk.

Urea is the major metabolite formed from dietary proteins and tissue protein turnover and is excreted from the liver into the bloodstream and then into milk in a stable form [59]. The results of this study showed that the urea content of milk was optimized after using the new teat liner. This means that after using the new teat liner in farm 1, the urea content fell from 24.98 ± 0.78 mg/dl to 15.03 ± 0.54 mg/dl and in farm 2, the urea content increased from 12.21 ± 0.30 mg/dl to 14.30 ± 032 mg/dl. Since the situation on farm 1 was the same before and after the use of the new teat liner, it is difficult to determine which factor other than the liner influenced the urea content of the milk. This is also the case on farm 2. whether this contradiction in the results (decrease in urea concentration in milk on farm 1 and increase in urea concentration in milk on farm 2) that occurred after the use of new teat liners was coincidental or whether there were other factors that influenced these results. Nowadays, it is not yet known whether the teat liner has an influence on the urea content of the milk or not, as urea is strongly influenced by husbandry and feeding conditions, such as the lack of rumen-degradable proteins and changes in dry matter intake or energy in the feed ration [60]. Therefore, the milk urea content may serve as an on-farm indicator to guide nutritional strategies [61–65]. Further studies are needed to clearly demonstrate the influence of the use of teat liners on urea in milk. However, it has been observed that a reduction in the amount of urea in the milk of cows is associated with IMI and increased SCC [66,67]. This is exactly what was observed in farm 2, that the urea content increased after the use of new teat cup liners and the reduction of SCC in the milk. The mechanisms behind this relationship are not yet fully understood and therefore require further investigation.

5. Conclusion

- The SSL teat cup liner has a significant influence on the degree of udder emptying, the maintenance of the udder health and the quality of raw milk.
- After using the new SSL teat cup liner, the udders remained healthy throughout the study period, showing lower somatic cell counts (SCC).
- High milk yield and better milk quality can be achieved by using SSL teat cup liner as it is adapted to all teat shapes.
- Finally, the SSL teat cup liner prevents injuries, and thus makes a decisive contribution to greater animal welfare and improved animal health.

Author contributions: Conceptualization, SK; methodology, SK; software, KAN; validation, SK, KAN and MWP; formal analysis, KAN; investigation, SK; resources, SK; data curation, SK, KAN; writing—original draft preparation, SK; writing—review and editing, MWP; visualization, SK; supervision, MWP; project administration, SK; All authors have read and agreed to the published version of the manuscript.

Conflict of interest: The authors declare no conflict of interest.

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