Enhancement of Dairy Cow Milk Quality with Probiotic and Inorganic Selenium Supplementation

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Abstract: Selenium (Se) is an essential micronutrient crucial in various metabolic processes. Dairy production is continually expanding and can supplement the population of regions with low Se. Adding live yeast concentrate (LYC) to cows’ diets can influence milk production, composition, and quality by modulating ruminal microbiota, resulting in increased milk yield and improved nutritional content. This study aimed to assess the enrichment of milk with selenite LYC for a subsequent increase in quality and production in supplemented lactating cows. Twenty-six cows were separated into three groups supplemented with different concentrations of Se and LYC. The milk was evaluated using physical–chemical, microbiologic, and toxicologic parameters according to Brazilian legislation. The addition of Se and yeast did not alter milk production; however, the application did lead to an increase in milk fat concentration compared with the control group, and no significant variations were observed in other physical–chemical parameters. Regarding the microbiological and toxicological analyses, all the samples presented satisfactory hygienic and sanitary conditions. The Somatic Cells Count from all treatments remained below 500,000 somatic cells mL−1, representing a positive effect of Se. The milk Se content was expected in residual form with the organic selenium being the more bioavailable form throughout the processing chain. The supplementation yielded results similar to those in the literature, highlighting the potential for customized technology and processes in dairy farming in ways that improve production, quality, and sanitation.

Keywords: sodium selenite; Saccharomyces cerevisiae; probiotic; milk production

1. Introduction

Selenium (Se) plays a crucial role in various metabolic processes in animals and humans, serving as an essential micronutrient that plays a role as a component of 25 selenoproteins identified in animals. Selenium deficiency is linked to a predisposition towards developing diseases such as cancer, viral diseases, diabetes, and cardiovascular diseases, among others, due to its significant role in regulating immune and antioxidant activities [1–3].

The intake of this micronutrient is directly related to its presence in soils and its incorporation into foods. However, the concentration of Se in soils can be influenced by
various factors, including volcanic activity, fertilizer use, the presence of water sources, pH, and soil moisture [4,5].

The supplementation of Se in Brazil is important because few studies have been conducted to quantify and qualify the concentrations in soil and human blood due to this country’s vast territorial expanse. The differences in selenium concentrations appear to be discrepant even within Brazilian states. An example is the state of São Paulo, which has been shown in studies to feature large variations in selenium concentration in its soil, ranging between 0.093 and 0.127 mg kg\(^{-1}\); therefore, it is considered a soil with low levels of selenium, and thus as deficient [6,7].

Research conducted in São Paulo indicated that the majority of daily dietary selenium intake ranges observed were below the estimated average requirement values. This underscores the significance of seeking out new sources for supplementation [8]. The application of selenium-enriched fertilizers in soil represents a viable strategy for augmenting the levels of this micronutrient in cereals, grains, and animal-derived commodities consumed in Europe [9]. Another form of supplementation to enrich products of animal origin, specifically milk, is through the addition of selenium to the diet of lactating cows. Studies show that supplementation with selenized yeast, sodium selenite (Na\(_2\)SeO\(_3\)), or sodium selenate (Na\(_2\)SeO\(_4\)), forms most commonly found in food supplements, causes an average increase of around 16 µmol L\(^{-1}\) of milk [10].

Dairy production is constantly expanding, represented by an increase not only in consumption, but also in the entire economy. Global milk production increased by approximately 1.1% in 2021, with the majority being sourced from cows (81% from cow’s milk, 15% from buffalo milk, and 4% from goat, sheep, and camel milk). The forecast projections of governmental agencies for growth in global milk production for the next decade surpass those of most agricultural commodities, with an expectation of 1060 Mt by 2031. Similarly, alongside the growth in milk production, there is a global expectation of an increase in the per capita consumption of dairy derivatives in the next decade [11–13]. Milk is an essential component of the diet, accounting for a significant portion of human nutrition [9]. The supplementation of micro minerals in cows’ diets can enhance milk’s nutritional value and its derivatives [11]. Thus, enriching it can naturally supplement Se, reinforcing the importance of this food in the diet.

However, ruminants are less efficient in absorbing Se than monogastric animals [14,15]. It is known that Se has an absorption efficiency in humans of around 80% in its inorganic form, which can exceed 90% in its organic form [16]. The lower absorption by ruminants can be attributed to the fact that part of the Se in the rumen is converted into insoluble forms [11]. Mapelli [17] in 2011 demonstrated the conversion of inorganic Se into selenoproteins using Saccharomyces cerevisiae. The use of selenized yeasts led to a significant increase in the excretion of this mineral in milk, although several factors may influence the serum Se responses of different cows, such as forage types and sources, ruminal environment, fat, dietary calcium, trace metals, and genetics [11,18,19].

The literature indicates that the supplementation of dairy cows with organic selenium and probiotics in combination can enhance feed efficiency, milk production, milk composition, and overall performance. Additionally, dietary supplementation with selenized Saccharomyces cerevisiae positively affects blood selenium status, thereby contributing to the production of milk with higher selenium content [20,21].

The hypothesis of this experiment is that the enrichment of milk with selenium (Se) and live yeast concentrate (LYC) will augment milk quality and production in supplemented lactating cows. Selenium is known to play an important role in the body’s antioxidant defense mechanisms. Administering this micronutrient to dairy cows may yield improvements in milk quality by reducing somatic cell counts and increasing milk oxidative stability. Additionally, the utilization of LYC aims to manipulate ruminal fermentation, fostering improvements in animal performance. The present study aims to assess the enrichment of milk with selenite and live yeast concentrate (LYC), aiming for a subsequent increase in quality and production potential in supplemented lactating cows.
2. Materials and Methods

2.1. Experimental Design and Diets

The experiment was developed with the herd and facilities at the Fazenda Escola Cachoeiras de Macacu (FECM), at the Universidade Federal Fluminense (UFF) in Brazil. Twenty-six (26) crossbred cows of commercial lineage were used in this study. The animals were a mix of Gir and Holsteins dairy cows. The animals weighed 400 to 540 kg, with body condition scores ranging from 2.5 to 4.0, aged between 4 and 6 years, in their third to fourth parity. The milking period lasted 60 days (milked twice per day), and all animals allocated to the experiment were in the initial third of their lactation period. Supplementation began on the first day and, after 15 days of adaptation, continued for 30 days of experimentation. After selection, the animals were separated into three groups (n = 8) with two animals as backup during the adaptation period. The groups were homogenized based on the average milk production. The animals received a complete diet—total mixed ration (TMR), like all groups of animals on the farm.

The animals were divided into the following groups with the proposed treatments: Treatment 1 (T1) Control Group, with just dry matter intake (DMI); Treatment 2 (T2) DMI + 0.3 mg kg$^{-1}$ DMI of inorganic selenium + live yeast concentrate (LYC); Treatment 3 (T3) DMI + 0.6 mg kg$^{-1}$ DMI of inorganic selenium + LYC. The LYC was composed of a commercial strain, lyophilized *Saccharomyces cerevisiae*, with a concentration of 10$^8$ CFU g$^{-1}$, being added at 5.0 mg kg$^{-1}$ DMI. The DMI (kg d$^{-1}$) per treatment was T1 10.92 ± 1.41, T2 11.59 ± 1.54 and T3 11.59 ± 1.14.

Experimental cows had similar lactation numbers (T1 3.0 ± 0.4; T2 3.0 ± 0.3; T3 3.0 ± 0.5), lactation days (T1 34 ± 4; T2 36 ± 3; T3 34 ± 5), and production levels (Kg) at the beginning of the experiment (T1 18.1 ± 3.0; T2 18.2 ± 4.0; T3 17.7 ± 2.5). All cows had free access to water and vitamin/mineral mix (Purina Agribrands, Campinas, São Paulo, Brazil). Before each milking, each cow received corn silage and a mixture of grain (corn kernels and dried malt bagasse) according to their production. This diet was assessed weekly and all groups had access to pasture (*Panicum maximum*) for 8 h daily. The quantity and quality of the available pasture were evaluated biweekly and estimated as an excess of nutrients for lactating cows.

The experimental cows were milked twice daily, at 06:30 a.m. and 2:30 p.m. During the milking process, all 4 glands were first tested and then milked completely using a closed mechanical milking system. The milking operation was performed using a 1 × 10 low-line Casse system milking parlor featuring 10 milking stalls and 2 milking clusters. The milking machine (Westfalia Elk Grove Village, IL, USA) was set at 120 pulsations/min in a 50:50 ratio at a vacuum of 36 kPa. The milking routine included teat cleaning and drying, and foremilk hand milking on a back cup to ensure the absence of milk flocks as pre-stimulation practices during approximately 1 min. Teat cups were attached thereafter and manually detached after the visual observation of milk flow cessation. Machine stripping was not applied. The milking routine was constant, and the same person performed all experimental milkings.

The experiment was performed in accordance with Law 11.794 of 8 October 2008, Decree 6899 of 15 July 2009, as well as with the rules issued by the National Council for Control of Animal Experimentation (CONCEA), and was approved by the Ethics Committee on Animal Use of the Universidade Federal Fluminense (CEUA/UFF). The nutritional protocol for evaluating the animals was submitted to the animal ethics committee and was authorized under the UFF registration (CEUA nº 1101203021).

The treatments were provided to the animals via mixing into the total diet, aiming to reach the stipulated concentration. During the milking procedures, the animals’ zootechnical performance parameters were measured using the “dark bottom mug” test and California Mastitis Test (CMT), as well as milk production indices. For sampling, representative samples from each group were obtained from a mixture of two daily milkings to evaluate milk quality and fixed selenium levels. The analyses were carried out in the laboratories of partner institutions, with physical–chemical and microbiological analyses.
carried out in the Centro Estadual de Pesquisa em Qualidade de Alimentos (CEPQA-RJ), belonging to the Empresa de Pesquisa Agropecuária do Rio de Janeiro (PESAGRO-RJ). In contrast, the quantification of residual selenium in milk was carried out at the Laboratório de Micologia e Micotoxinas (LAMICO) of the Universidade Federal de Minas Gerais (UFMG). Also, all animals were given a basic diet for maintenance during the lactation period. This diet is described in Table 1.

Table 1. Ingredients and nutritional composition of the experimental diets (g kg\(^{-1}\) of dry matter).

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Diets (g kg(^{-1}) of Dry Matter)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Corn silage</td>
<td>110</td>
</tr>
<tr>
<td>Corn kernels</td>
<td>110</td>
</tr>
<tr>
<td>Dried malt bagasse</td>
<td>60</td>
</tr>
<tr>
<td>Mineral premix (^\text{(1)})</td>
<td>22</td>
</tr>
<tr>
<td>Dicalcium phosphate</td>
<td>5</td>
</tr>
</tbody>
</table>

\(^{1}\) Mineral premix chemical composition (quantities/kg of commercial product): Ca, 230.0 g; P, 50.0 g; Co, 30.0 mg; Mg, 8.0 g; Mn, 1000.0 mg; Zn, 1875 mg; Se, 12.0 mg; I, 30.0 mg; S, 15.0 g; Fe, 500.0 mg; Cu, 255.0 mg; Na, 55.0 g.

2.2. Evaluation of the Identity Standards, Physicochemical and Toxicological Milk Quality

For the analysis of milk quality and composition, the reference standards established in Normative Instruction No. 76 [22], dated 28 November 2018, were followed. The evaluation techniques adhered to the technical guidelines of the Ministério de Agricultura, Pecuária e Abastecimento (MAPA) and were carried out according to the methodology of the Adolfo Lutz Institute [23]. The following assessments were performed: fat content, acidity, relative density at 15 °C, cryoscopy index, non-fat solids, total protein, and alizarol stability. Selenium in the milk was assessed using the ICP-MS method following the AOAC methodology [24].

Toxicological evaluations were realized considering the health risks associated with the presence of these substances. The staphylococcal enterotoxin was evaluated as required by legislation. The screening for specific genes responsible for staphylococcal enterotoxin was performed in the way Pyzik et al. [25] described. Aflatoxin M1 detection and quantification were performed in raw materials as described by Moebus et al. [26].

2.3. Evaluation of the Identity Standards and Microbiological Quality of Milk

Microbiological analysis of the milk was carried out to confirm the quality of the product after the administration of the supplements. At the end of supplementation, the three treatments were subjected to microbiological analyses to certify the quality of the final product. The microbiological and health quality parameters evaluated were the mesophilic bacterial count and somatic cell count (SCC) as recommended in the Brazilian guidelines for dairy products established in Normative Instruction No. 76 [22] according to the methodologies described in the Compendium of Methods for the Microbiological Examination of Foods (APHA) for bacterial counting and identification [27]. Furthermore, total coliforms, coliforms at 45 degrees Celsius, coagulase-positive staphylococci, lactic acid bacteria, and Salmonella spp. were also analyzed, aiming to ensure the safety of the end product for consumption and its potential suitability for dairy product production [28].

Serial dilutions were prepared at a ratio of 1:1000 from 25 g of each sample. Each dilution was inoculated into Petri dishes containing specific culture media for each microorganism investigated. Viable cell counts after incubation were performed through plate counts using a Biocell Biocc150-Bi colony counter (Prolab®, São Paulo, Brazil).

Somatic cell counts were determined using the flow cytometry-type method (DeLaval cell counter 15600, AKSO, DeLaval International AB, Tumba, Sweden) and were reported in integer units to the nearest 100,000 cells/mL. All milk samples were heated in a water bath at 40 °C for 15 min before analysis. The instruments were operated according to the manufacturer’s recommendations; adjustments to the samples were performed as part
of the starting procedure to verify the correct alignment of the laser and detectors on the equipment. All the analyses were performed in triplicate.

2.4. Statistical

The data analyses were conducted through analysis of variance (ANOVA) and performed using the PROC GLM computational program in the Statistical Analysis System (SAS) (SAS Institute, Cary, NC, USA). The fixed interaction between treatment and sampling time is the random effect of cows tested within the treatment. The same model was used to analyze milk production, milk characterization, and body weight changes with supplementation. The fixed effects of time and their interactions were removed from the ANOVA model. Previous lactation yield, milk production range, milk parameters range, and the concentrations of residual milk Se indices obtained during 21 days relative to the expected calving date were used as covariates, and covariates were excluded from the model if they were not significant \( p > 0.1 \). Comparisons were also made among the variables obtained in the in vivo experiment, including fat, acidity, relative density at 15 degrees Celsius, cryoscopic index, non-fat solids, total protein, alizarol stability, SCC, TBC, coagulase-positive \textit{Staphylococcus}, lactic acid bacteria, and total and thermotolerant coliforms, as well as the quantification of selenium in milk. The data are reported as LSM, and statistical significance was indicated at \( p \leq 0.05 \), with trends toward significance assessed using Tukey’s multiple comparison test.

3. Results and Discussion

3.1. Milk Yield, Physicochemical and Toxicological Parameters

After the cow experimentation period, the collected milk was analyzed, and the physical–chemical parameters are arranged in Table 2 below with the principal nutrient composition.

<table>
<thead>
<tr>
<th>Brazilian Legislation</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>( p )-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Milk yield (kg·d(^{-1}))</td>
<td>7–8</td>
<td>8.50 ± 1.11</td>
<td>9.50 ± 0.91</td>
<td>8.50 ± 1.12</td>
</tr>
<tr>
<td>ECM (kg·d(^{-1})) ( ^2 )</td>
<td>-</td>
<td>16.71 ± 0.90</td>
<td>16.23 ± 0.90</td>
<td>16.70 ± 0.90</td>
</tr>
<tr>
<td>ECM/DMI -</td>
<td>1.56</td>
<td>1.40</td>
<td>1.44</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td>Fat (g·kg(^{-1}))</td>
<td>35–53</td>
<td>28.91 ± 0.75 ( ^{A} )</td>
<td>31.12 ± 0.89 ( ^{B} )</td>
<td>32.28 ± 0.90 ( ^{B} )</td>
</tr>
<tr>
<td>Protein (g·kg(^{-1}))</td>
<td>27–45</td>
<td>32.33 ± 1.22</td>
<td>33.30 ± 1.02</td>
<td>33.32 ± 1.01</td>
</tr>
<tr>
<td>Casein (g·kg(^{-1}))</td>
<td>18–29</td>
<td>22.3 ± 1.06</td>
<td>22.3 ± 1.11</td>
<td>22.3 ± 1.12</td>
</tr>
<tr>
<td>Lactose (g·kg(^{-1}))</td>
<td>34–54</td>
<td>46.5 ± 1.45</td>
<td>46.5 ± 1.00</td>
<td>46.5 ± 1.02</td>
</tr>
</tbody>
</table>

ECM: Energy-corrected milk. ECM/DMI: Efficiency of milk production corrected for energy. \(^{A}\) Raw milk quality standard according to the Ministry of Agriculture, Livestock and Supply (MAPA). \(^{B}\) ECM = (milk yield \times (0.0929 \times % \text{fat} + 0.0547 \times % \text{protein} + 0.0395 \times % \text{lactose}) \times 4.187)/2.931. Averages in the same line followed by the same letters do not differ from each other at 5% significance by Tukey test \( (p < 0.05) \).

The inclusion of selenium (Se) and \textit{Saccharomyces cerevisiae} had no impact on milk yield. Changes in the physical–chemical parameters of milk can be observed mainly in the concentration of fat. However, from what we could observe of other physical–chemical parameters, no significant variations were observed in levels of lactose and casein, the general appearance of the milk, or even in the oxidation-reductive potential.

The effects of organic mineral supplementation on milk production and composition reported in the literature vary considerably. Some authors report the effects of organic trace mineral supplementation on milk production, with no change in their composition \([4,5,16,29]\); the results of the present work show that variation in the study model, diet, and production period can significantly influence the positive response to supplementation. The study evaluates the perspective of Brazilian small farms, and the average milk production in small family farms varies significantly due to factors such as...
socioeconomic limitations, which often prevent small-scale producers from investing in advanced technologies and infrastructure.

Regarding the parameters manifesting significant changes, the addition of Se and *S. cerevisiae* leads to an increase in milk fat concentration compared to the control group. The literature indicates that the supplementation of selenium and LYC in dairy cow diets significantly enhances the fatty acid composition of milk, particularly increasing the percentages of polyunsaturated fatty acids in milk fat [30,31]. This enhancement is not solely attributed to DMI, although DMI is a key indicator of volatile fatty acid production in the rumen, which is directly linked to acetate production—the primary precursor of milk fat. This suggests that dietary selenium can modulate milk fat synthesis through multiple pathways, highlighting the complex interplay between dietary components and milk fat synthesis beyond the scope of DMI alone. While no significant differences were observed among the energy conversion parameters, these findings underscore the intricate relationship between dietary components and milk fat synthesis.

Zhang et al. [32] showed that the ability to positively modulate the microbiota of the gastrointestinal tract, because of Se and LYC supplementation, allows a better response in the quality of milk produced by lactating females. Faccendaa et al. [16] affirmed that this supplementation must be associated with a good energy balance in the diet. The response in milk quality is directly proportional to a balanced diet, achieving the minimum DMI requirements, considering that supplementation will allow the better use of nutrients, as it positively stimulates the ruminal microbiota.

Faccenda et al. [16] similarly found that *S. cerevisiae* and Se had no impact on milk urea nitrogen levels. This underscores the potential for yeast supplementation to enhance the growth of rumen fibrolytic bacteria with a preference for ammonia, thereby enhancing the utilization of ammoniacal nitrogen and minimizing its excretion. However, the absence of enhancements in fiber digestibility, microbial protein synthesis, and milk yield suggests a lack of influence on ruminal bacteria and, consequently, nitrogen utilization.

The findings of the present study align with the common perspective that dietary supplementation is unlikely to significantly impact milk yield and encourage implementing the feeding strategy or ripening time. As expected, the only differences involved the increase in dry matter at the end of the ripening period.

### 3.2. Microbiological and Toxicological Analysis

After carrying out the experimentation, the collected milk was analyzed, and the microbiological and toxicological parameters are arranged in Table 3.

**Table 3.** Average and standard deviation of microbiological counts from milk collected from cows fed diets containing the addition of selenium supplementation and *Saccharomyces cerevisiae*.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>SSC ¹</th>
<th>Aerobic Mesophilic ²</th>
<th>Total Coliforms ²</th>
<th>Thermotolerant Coliforms ²</th>
<th>Acid Lactic Bacteria ²</th>
<th>Salmonella spp.</th>
<th>Aflatoxin M1 ³</th>
</tr>
</thead>
<tbody>
<tr>
<td>Brazilian legislation</td>
<td>500.00 ⁴</td>
<td>5.70 ⁴</td>
<td>2.0 ⁵</td>
<td>1.0 ⁵</td>
<td>-</td>
<td>Abs. ⁵</td>
<td>0.5</td>
</tr>
<tr>
<td>T1</td>
<td>155.60 ± 12.00 ³</td>
<td>5.00 ± 0.77 A</td>
<td>1.30 ± 0.23</td>
<td>&lt;1.0</td>
<td>4.30 ± 1.00</td>
<td>Abs.</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>T2</td>
<td>126.40 ± 9.00 B</td>
<td>4.00 ± 0.73 B</td>
<td>1.20 ± 0.26</td>
<td>&lt;1.0</td>
<td>4.20 ± 0.55</td>
<td>Abs.</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>T3</td>
<td>122.00 ± 11.00 B</td>
<td>4.00 ± 0.75 B</td>
<td>1.20 ± 0.24</td>
<td>&lt;1.0</td>
<td>4.30 ± 0.45</td>
<td>Abs.</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

Values represent the average microbial load by log colony-forming units per gram of 3 different treatments collected during the study period. Abs.: Absence. ¹ Somatic cell count (thousand cell mL⁻¹). ² Microbiological counts (log₁₀ CFU mL⁻¹) with the detection limit being ≤0.001 ng mL⁻¹. ³ Raw milk quality standard according to the Ministry of Agriculture, Livestock and Supply (MAPA). ⁴ Microbiological quality standard for dairy products according to the Agência Nacional de Vigilância Sanitária (ANVISA). Limit of detection: 1.0 log₁₀ CFU mL⁻¹. Averages in the same column followed by the same letters do not differ from each other at 5% significance by Tukey test (p < 0.05).

The choice of applying a quality standard to the raw materials for products aimed at human consumption is related to the need to ensure final product safety, as crude milk quality is an important factor in how the product is manipulated. Thermotolerant coliforms,
Salmonella spp., and Staphylococcus aureus, were not present in the samples, demonstrating that the milking procedure was performed aseptically, thus remaining within Brazilian legislation standards from the handling stage to the storage period [22]. Although not described in the quality standard, the lactic acid bacteria count is a strong indicator used in the production of dairy products. A high count of lactic acid bacteria can cause pre-fermentation in the milk, making its disposal unfeasible.

Also, there were no detected levels of staphylococcal enterotoxin and aflatoxin M1 that related to the legislation’s limits. Staphylococcal enterotoxins, primarily associated with Staphylococcus aureus, constitute a significant cause of food poisoning, particularly in milk and dairy products [33,34]. Appropriate milking procedures associated with pasteurization serve as an effective measure for diminishing microbial concentrations in milk and its derivatives. Nevertheless, the detection of staphylococcal enterotoxins is imperative due to their thermoresistant nature, potentially leading to food poisoning outbreaks even in the absence of the pathogenic microorganism [33,34].

The quantification of aflatoxin M1 in raw materials and final products, particularly in the dairy industry, is crucial to ensuring consumer safety and well-being. AFM1, being thermoresistant and globally prevalent, possesses carcinogenic, cytotoxic, teratogenic, and genotoxic properties. Prolonged exposure to AFM1 can result in chronic conditions such as immunosuppression, hepatocarcinoma, and impaired growth in children [35–37].

The absence of aflatoxin M1 in the samples not only attests to the safety of the product for consumption, but also reinforces the need to provide high-quality feed for animals, since lactating animals subjected to the continuous ingestion of aflatoxin B1 through fungal contamination can also produce and excrete aflatoxin M1 in milk [5,8,9,11]. Therefore, all samples presented satisfactory hygienic–sanitary conditions, with no significant handling and processing inferences concerning product safety.

According to Andrade et al. [38], the supply of Se and Saccharomyces cerevisiae in dairy cows did not affect milk yield and composition; however, it promoted a reduction in both the somatic cell count as well as the incidence of subclinical mastitis. This nutrient decreases oxidative damage and enhances the bactericidal capability of neutrophils by activating glutathione peroxidase.

This was corroborated by the research conducted by Facchinetti et al. [16] and Cortinhas et al. [29], which identified the effects of Se supplementation on somatic cell count (SCC) and immune response, correlating with Se levels ranging from 1.6 to 4.8 mg per day. The SCC of milk from all treatments remained below 500,000 somatic cells mL\(^{-1}\) [22], suggesting that the health of the mammary gland is possibly related to the positive effects of selenium. However, no reduction in SCC was observed as a result of the supplementation with live selenized yeast in comparison to other treatments.

3.3. Milk Total Organic Selenium Residual Concentration

After 30 days of supplementation adaptation, the animals began to be monitored during daily milkings, with weekly sample evaluation for parameters and levels of selenium incorporation in the milk. For 21 days (Table 4), the animals were monitored daily. This organic selenium ends up allowing this residual selenium to become more bioavailable throughout the processing chain.

The milk was expected to contain Se, independent of the dose, in a residual form of sodium selenite or selenate and Se yeast. Organic forms of selenium, such as selenomethionine and selenocysteine, are absorbed more efficiently compared to inorganic forms like selenite. This enhanced absorption occurs particularly when selenomethionine and selenocysteine are integrated into non-functional proteins. In the experiment, the use of sodium selenate at a concentration of 0.6 mg kg\(^{-1}\) of DM in association with the LYC of S. cerevisiae was able to reduce SCC levels and increase fat content when compared to lower concentrations. Selenium concentration did not generate significant results regarding production, protein, defatted dry extract, or lactose.
Table 4. Average and standard deviation of residual selenium and fat from milk of cows fed diets containing the addition of sodium selenite supplementation and *Saccharomyces cerevisiae*.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>7 Days (mg L⁻¹)</th>
<th>15 Days (mg L⁻¹)</th>
<th>21 Days (mg L⁻¹)</th>
<th>Fat (g kg⁻¹)</th>
<th>SSC</th>
</tr>
</thead>
<tbody>
<tr>
<td>T1</td>
<td>0.0034 ± 0.0008</td>
<td>0.0033 ± 0.0007</td>
<td>0.0035 ± 0.0009</td>
<td>28.91 ± 0.75</td>
<td>155.60 ± 12.00</td>
</tr>
<tr>
<td>T2</td>
<td>0.059 ± 0.008</td>
<td>0.058 ± 0.0081</td>
<td>0.057 ± 0.0083</td>
<td>31.12 ± 0.89</td>
<td>126.40 ± 9.00</td>
</tr>
<tr>
<td>T3</td>
<td>0.061 ± 0.0081</td>
<td>0.064 ± 0.0082</td>
<td>0.063 ± 0.0084</td>
<td>32.28 ± 0.90</td>
<td>122.00 ± 11.00</td>
</tr>
</tbody>
</table>

Values represent the average microbial load by log colony-forming units per gram of 3 different treatments collected during the study period; Turkey test *p* > 0.05. 1 Somatic cell count (thousand cell mL⁻¹). Averages in the same column followed by the same letters do not differ from each other at 5% significance by Tukey test (*p* < 0.05).

In his work, Ceballos et al. [10] reported that the administration of Se (6.0 mg per head per day) associated with yeast resulted in milk with a Se concentration of 0.37 µmol L⁻¹ greater than the concentration in cows supplemented only with sodium selenite or selenate 75 days after starting supplementation.

In another study, Ianni et al. [30] reported that dietary Se supplementation in cows appears to also lead to an increase in conjugated linoleic acid (CLA) in milk and cheese. It was reported that myristoleic acid (C14:1), linoleic acid (C18:2), and rumenic acid (CLA cis-9 trans-11) played an important role in antioxidant activity. These fatty acids protected bovine mammary epithelial cells from lipoperoxidation and mitigated levels of reactive oxygen species, leading to improved mammary gland function. The increase in CLA in milk also represents an increase in the consumer’s quality of life, since dairy products represent the main food source of CLA for humans. Possibly, as reported in the literature, this may be accompanied by a total increase in fat levels. As a result, there is not only a significant increase in fatty acids with functional potential, but also an increase in beneficial microbiota, evidenced by the decrease in deteriorating contaminants and the maintenance of lactic acid bacteria. Therefore, selenium supplementation would not only have a direct effect on residual levels in milk but also mainly impact the health of the mammary glands.

Dietary supplementation with selenium (Se) and *S. cerevisiae* shows promise in enhancing the nutritional and nutraceutical qualities of cow milk. Furthermore, it offers significant insights into enhancing desaturation mechanisms within the bovine mammary gland. Our primary discoveries highlight increased levels of linoleic acid and conjugated linoleic acid in milk and cheese from cows supplemented with Se, leading to a reduction in saturated fatty acids. This reflects improvements in the health properties of these dairy products, with noteworthy implications for human consumption. Furthermore, the aromatic profile of dairy products was also positively affected by the diet, enabling the application of this milk in the production of dairy products with nutraceutical and prebiotic potential, in addition to serving as a substrate for the multiplication of probiotic microorganisms in these products.

4. Conclusions

Supplementing lactating cows with both sodium selenite and *S. cerevisiae* consistently demonstrates advantageous effects on milk quality in numerous studies. While no significant impact on milk yield was observed, higher milk fat concentrations were noted with the inclusion of Se and *S. cerevisiae*. Moreover, the study highlights the importance of a balanced diet in conjunction with supplementation, emphasizing its positive effects on milk quality, including reduced somatic cell count and improved immune response.

New studies should be conducted to optimize the processes of supplementing probiotics and Se. Encouraging the supplementation of food in dairy cattle farming will not only directly improve milk quality and sanitary conditions but also emphasize the necessity of the development of new technologies and processes adjusted to each circumstance.

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