








Review Article

Application of disinfectants in the prevention and control of bovine mastitis

Aplicação de desinfetantes na prevenção e controle da mastite bovina

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Abstract

A literature review was conducted to assess global trends and scientific contributions related to the use of disinfectants in the control of bovine mastitis. Bovine mastitis, a major inflammatory disease affecting dairy cows, is predominantly caused by bacterial infections and remains a significant concern for global dairy production. Among the preventive and control measures, the application of disinfectants plays a crucial role, particularly in pre- and post-milking hygiene protocols. To provide a more structured overview, this work maps publication trends, regional contributions, and evaluates the efficacy and diversity of disinfectants tested *in vivo* and *in vitro*. This review aimed to explore and synthesize the current scientific knowledge regarding the use of disinfectants in the control and prevention of bovine mastitis, focusing on the main chemical classes, mechanisms of antimicrobial action, efficacy, and emerging alternatives. Scientific articles were selected from PubMed, Web of Science, Scopus, and SciELO databases up to December 2024. From a total of 309 initially identified records, 86 relevant studies were included based on thematic alignment with disinfectant application in dairy production. Most publications originated from developed countries, particularly the United States (34.9%) and Germany (3.6%), with Brazil (19.3%) standing out in the South American context. A steady annual growth of approximately 5.6% in publication volume was observed, with increased scientific attention to sustainable alternatives from 2007 onwards. Synthetic disinfectants such as iodophors, chlorhexidine, and sodium hypochlorite remain widely studied due to their high *in vivo* and *in vitro* efficacy. In contrast, natural and biotechnological alternatives—such as essential oils, silver nanoparticles, and polyhexamethylene biguanide (PHMB)—have shown promising potential but require further validation. Key challenges identified include the need for standardization of concentrations, clear application protocols, and comprehensive safety assessments. This review emphasizes the importance of broadening research efforts in diverse geographic regions and of developing standardized, cost-effective, and environmentally friendly disinfection strategies, contributing to both animal health and the sustainability of global dairy production. The combination of scientometric and analytical review approaches contributes to both understanding research dynamics and identifying gaps in disinfection strategies.

Keywords: animal health, mammary inflammation, milk production, antimicrobials.

Resumo

Foi realizada uma revisão de literatura para avaliar as tendências globais e as contribuições científicas relacionadas ao uso de desinfetantes no controle da mastite bovina. A mastite bovina, uma importante doença inflamatória que afeta vacas leiteiras, é predominantemente causada por infecções bacterianas e continua sendo uma preocupação significativa para a produção leiteira global. Entre as medidas de prevenção e controle, a aplicação de desinfetantes desempenha um papel crucial, especialmente nos protocolos de higiene pré e pós-ordenha. Para oferecer uma visão mais estruturada, este trabalho mapeia as tendências de publicações, contribuições regionais e avalia a eficácia e a diversidade dos desinfetantes testados *in vivo* e *in vitro*. Esta revisão teve como objetivo explorar e sintetizar o conhecimento científico atual sobre o uso de desinfetantes no controle e prevenção da mastite bovina, com foco nas principais classes químicas, mecanismos de ação antimicrobiana, eficácia e alternativas emergentes. Artigos científicos foram selecionados nas bases de dados PubMed, Web of Science, Scopus e SciELO até dezembro de 2024. De um total de 309 registros inicialmente identificados, 86 estudos relevantes foram incluídos com base na afinidade temática com a aplicação de desinfetantes na produção leiteira. A maioria das publicações teve origem em países desenvolvidos, particularmente os Estados Unidos (34,9%) e a Alemanha (3,6%), com destaque para o Brasil (19,3%) no contexto sul-americano. Observou-se um crescimento anual constante de aproximadamente 5,6%

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no volume de publicações, com maior atenção científica voltada para alternativas sustentáveis a partir de 2007. Desinfetantes sintéticos como iodóforos, clorexidina e hipoclorito de sódio continuam sendo amplamente estudados devido à sua elevada eficácia *in vivo* e *in vitro*. Em contraste, alternativas naturais e biotecnológicas—como óleos essenciais, nanopartículas de prata e biguanida polixametileno (PHMB)—demonstram potencial promissor, mas ainda requerem validação adicional. Os principais desafios identificados incluem a necessidade de padronização das concentrações, protocolos de aplicação bem definidos e avaliações abrangentes de segurança. Esta revisão enfatiza a importância de ampliar os esforços de pesquisa em diferentes regiões geográficas e de desenvolver estratégias de desinfecção padronizadas, economicamente viáveis e ambientalmente sustentáveis, contribuindo tanto para a saúde animal quanto para a sustentabilidade da produção leiteira global. A combinação de abordagens de revisão cienciométrica e analítica contribui tanto para a compreensão da dinâmica da pesquisa quanto para a identificação de lacunas nas estratégias de desinfecção.

Palavras-chave: saúde animal, inflamação mamária, produção leiteira, antimicrobianos.

1. Introduction

Bovine mastitis is one of the most prevalent and economically significant diseases affecting dairy farming, characterized by inflammation of the mammary gland caused by pathogenic microorganisms, physical trauma, or irritating chemical agents (Gazzola et al., 2024; Yamauchi et al., 2022). It manifests clinically—with signs such as swelling, pain, and altered milk—or subclinically, detected through laboratory tests such as the California Mastitis Test and somatic cell count (SCC) (Sharun et al., 2021).

The disease compromises both the quality and volume of milk produced, resulting in considerable economic impact. The main etiological agents include *Staphylococcus spp.*, *Streptococcus spp.*, *Escherichia coli*, *Enterococcus spp.*, *Corynebacterium bovis*, and opportunistic fungi and algae such as *Candida spp.* and *Prototheca spp.* (Kybartas et al., 2023; Alves et al., 2017).

In major milk-producing countries—such as the United States, India, Germany, and Brazil—mastitis is recognized as a leading cause of financial losses in the dairy sector. In Brazil, the average SCC in 2024 reached 531,000 cells/mL, and with an estimated annual production of 35.4 billion liters, losses from subclinical mastitis were projected at 2.12 billion liters/year (Brasil, 2024).

The global impact of mastitis has prompted scientific efforts aimed at developing new antimicrobials and improving prevention and control protocols (EMBRAPA, 2022; Langoni et al., 2017; Kybartas et al., 2023). Among these measures, the use of disinfectants stands out as a central strategy—particularly in pre- and post-milking routines—to reduce microbial contamination of teats, udders, and equipment. Compounds such as iodophors, chlorhexidine, and sodium hypochlorite are widely used and demonstrate high antimicrobial efficacy when properly applied *in vivo* and *in vitro* (Gonçalves et al., 2015; Medeiros et al., 2009).

However, the effectiveness of these compounds varies according to formulation, concentration, and target microorganism. For instance, iodine alone showed less than 50% efficacy, while combinations of Nonoxinol-9, iodine, and chlorhexidine surpassed 90% inhibition against *S. aureus* (El Behiry et al., 2012). Similarly, chlorhexidine showed variable results—from <50% to >90%—depending on the microbial strain (Kybartas et al., 2023).

Polyhexamethylene biguanide (PHMB) has demonstrated potent activity, with MIC₉₀ values as low as 1 µg/mL against

Prototheca spp. and excellent efficacy when incorporated into nanoparticle systems (Leite et al., 2021; Lu et al., 2023). Other alternatives—such as essential oils, silver nanoparticles, and probiotics like *Lactococcus cremoris*—have also shown promise, although further standardization and safety validation are required.

In addition to international efforts, Brazilian research has played a significant role in advancing sustainable and biologically inspired antimicrobial strategies. Peron (2020) emphasized the potential of natural agents in animal health, while Barbosa (2018) investigated microbiological practices particularly suited to tropical environments. Together, these contributions strengthen the pursuit of innovative and eco-friendly alternatives to conventional disinfectants.

Despite the growing number of options, the use of disinfectants still faces limitations related to standardization, application practices, and risks such as milk contamination and skin irritation in animals (Berg et al., 2014). Additionally, overuse or misuse can contribute to microbial resistance and environmental harm.

Given these challenges, this review aims to synthesize and critically discuss the current scientific knowledge on the use of disinfectants for the prevention and control of bovine mastitis, addressing their chemical classifications, mechanisms of action, effectiveness, advantages, and limitations. The goal is to guide the development of more effective and sustainable practices in dairy production.

2. Materials and Methods

Reviews typically comprise three main phases: planning, conducting, and reporting results. The protocol by Silva Neto et al. (2022) was adopted to guide this review (Figure 1), along with an analysis to assess trends and impact of the literature. The approach integrates scientometric indicators (geographical and temporal trends, database mapping) with structured qualitative assessment of article content. Scientific articles available in the databases were used, with no time frame up to December 2024.

In addition to descriptive statistics, the study followed key elements of systematic review structure, including: definition of inclusion and exclusion criteria, independent assessment by pairs of reviewers, and extraction of standardized data variables. However, it was not registered as a PRISMA review.

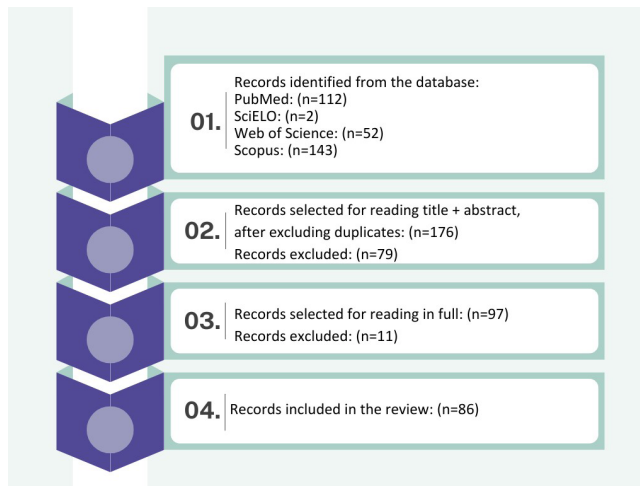


Figure 1. Methodological approach of the literature review on the use of disinfectants in the prevention and control of bovine mastitis. Flowchart of the process for identifying and selecting the studies included in the review (n=86).

The following databases were used: Web of Science, Scopus, e *SciELO* and National Library of Medicine (PubMed). A set of keywords were used in the database search process: “Disinfectants,” “Biocide,” “Biocides,” “Disinfectant,” “Mastitis, Bovine,” and “Bovine Mastitis” were combined using the Boolean operators AND and OR to refine the results. As a result, 143 studies were identified in Scopus, 52 in Web of Science, 112 in PubMed, and 2 in SciELO, totaling 309 articles. After removing duplicates and applying the inclusion criteria, 212 articles were excluded.

The inclusion criteria encompassed original research on the evaluation of disinfectants for the prevention and treatment of bovine mastitis, case reports, *in vitro* studies, and clinical trials, with no language restrictions. Studies unavailable in full text, those investigating disinfectants not directly related to bovine mastitis, and research evaluating products not classified as disinfectants were excluded.

After applying these criteria, 86 articles were selected for full-text review and data extraction. Each article was initially assigned a code, after which the specific information to be extracted was defined, including: year of publication, country where the study was conducted, type of study (*in vivo* and/or *in vitro*), compounds evaluated, concentrations and efficacy, sample collection sites, and microorganisms identified.

The assessment of the articles was conducted independently, in pairs, and blinded, using Excel software. In cases where consensus could not be reached between the reviewers, a third collaborator was consulted. Only articles that fully met the inclusion criteria were retained in the study.

Statistical analyses were performed using R software (R Core Team, 2024). To assess temporal trends in the number of publications and the association between different types of compounds (synthetic, animal, plant-based, and others), Pearson’s correlation was applied using the `perm.cor.test` function from the `RVAideMemoire` package (Hervé, 2023). The significance of the correlation was estimated through Monte Carlo simulation with 999 permutations, considering a *p*-value < 0.05 as statistically significant.

A contingency table was constructed using the `ftable` function, and the Chi-square test was conducted with the `chisq.test` function, both from the `stats` package (R Core Team, 2024). In addition to these analyses, descriptive data such as mean, mode, and percentages were compiled, along with graphical representations of the results. The Chi-square test was also employed to determine whether the efficacy of the compounds varied across studies and to assess the association between compound types and the experimental strategy used (*in vivo* or *in vitro*).

3. Results and Discussion

The literature review examines the temporal and geographical evolution of research related to the different classes of disinfectants used in the control of bovine mastitis, strengthening the scientific consistency of the study by enabling a comprehensive understanding of the field and a critical synthesis of the available experimental evidence regarding their effectiveness.

Bovine mastitis remains one of the most critical diseases affecting dairy production worldwide, causing significant economic losses and compromising milk quality. Among the strategies adopted for its prevention and control, the use of disinfectants stands out, with their efficacy widely investigated, especially when associated with proper management and milking protocols.

3.1. Geographical distribution and temporal trends

Figure 2 presents the geographical distribution of studies on the use of disinfectants in the prevention and treatment of bovine mastitis, showing a predominance of publications from North America and Western Europe. The United States leads with 34.9% of the total studies, followed by Canada (7.2%), the United Kingdom (4.8%), Germany, Australia, Japan, and New Zealand (3.6% each). In Latin America, Brazil stands out with 19.3% of the publications,

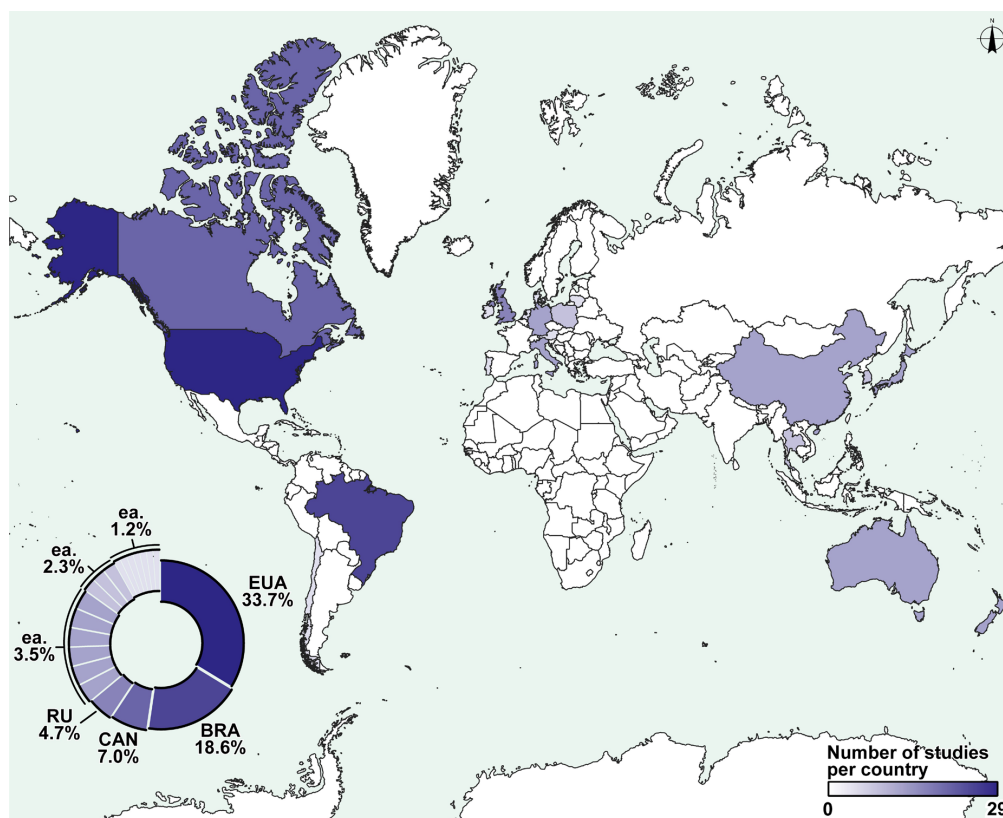


Figure 2. Geographical distribution of publications on disinfectants used in bovine mastitis control (1968–2024).

reflecting its global prominence in milk production and increasing investment in animal health research.

Although global milk production has expanded, research remains concentrated in developed countries with greater investments in science and technology. Regions such as Africa and parts of the Middle East are underrepresented, possibly due to limited research infrastructure or lower sanitary prioritization (Addis et al., 2024; Gazzola et al., 2024).

The temporal trend of publications is shown in Figure 3, revealing a progressive increase over time ($r_s = 0.46$; $p < 0.004$), with an average annual growth rate of 2.90%. From 2007 onward, there is a significant inflection point ($p < 0.05$), marking the beginning of a new phase, likely driven by growing concerns about antimicrobial resistance and the pursuit of more sustainable practices (Castro et al., 2023).

3.2. Chemical classification, efficacy, and mechanisms of action

The compounds evaluated over the past decades include synthetic disinfectants (halogens, biguanides, quaternary ammonium compounds), natural products (essential oils, plant extracts), and biological agents (probiotics, bacteriocins, phages), applied in different concentrations and formulations. Table 1 summarizes the efficacy of each class against the main etiological agents, with inhibition

rates classified as very high (>90%), high (75–90%), moderate (50–75%), or low (<50%).

No statistically significant differences were observed among compound types ($X^2 = 0.78$; $p = 0.99$), nor between *in vitro* and *in vivo* approaches ($X^2 = 3.35$; $p = 0.605$). However, *in vivo* studies have predominated since the 1960s, offering realistic evaluations under field conditions. In recent decades, the number of *in vitro* studies has grown, enabling controlled testing of novel compounds.

Among synthetic compounds, iodophors (0.25–5%) (Grindal and Priest, 1989), sodium hypochlorite (Park and Han, 2002), chlorhexidine (Schwenker et al., 2022), quaternary ammonium compounds (Arabe Filho et al., 2023), and metal nanoparticles (Fernandes et al., 2021) have been widely tested, with efficacy ranging from moderate to very high. *In vivo* studies involving direct application to udders and teats have shown high efficacy, especially against *Staphylococcus aureus*, *Streptococcus agalactiae*, and *S. dysgalactiae* (Lopez-Benavides et al., 2009; Justice-Allen et al., 2010).

Although natural and biological compounds are less represented, they have gained attention. Essential oils from *Lippia origanoides*, citronella, and basil (0.78–6.25 $\mu\text{L/mL}$) have demonstrated very high efficacy against *S. aureus* and *Escherichia coli* (Marcelo et al., 2020). Probiotics such as *Lactococcus cremoris* and phage therapy have also shown high antimicrobial potential (Gazzola et al., 2024; Xue et al., 2024).

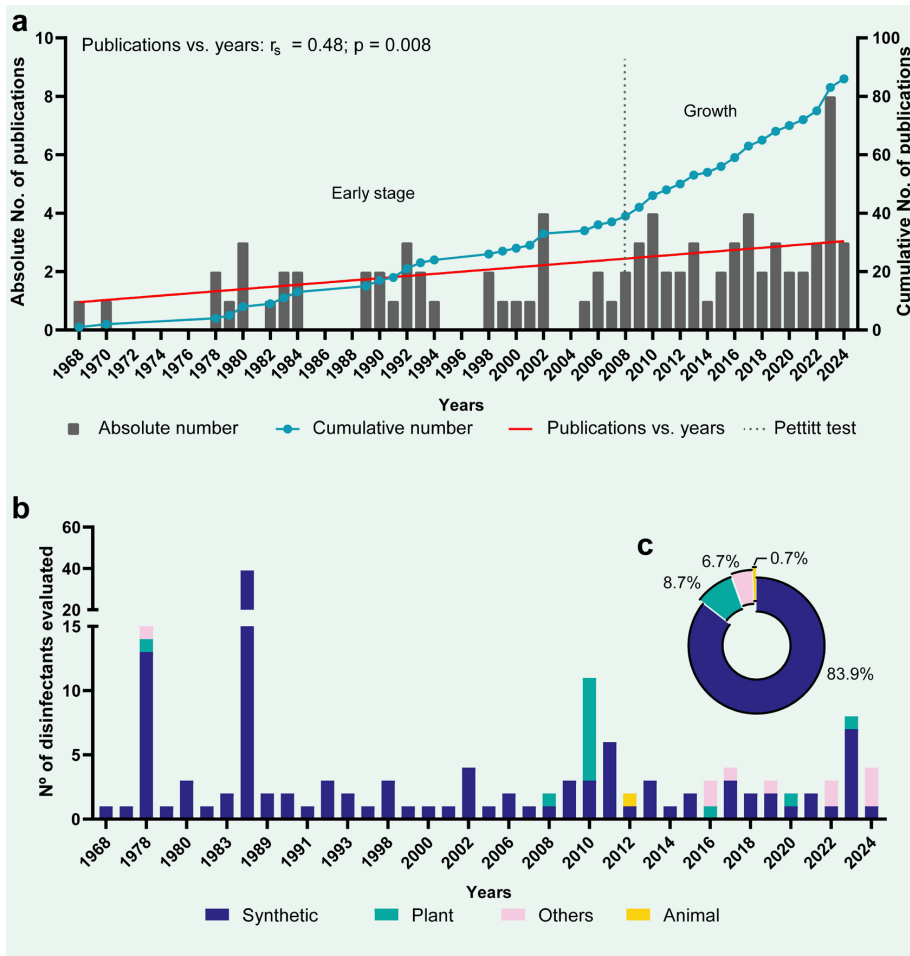


Figure 3. Temporal trend of publications on disinfectants used in bovine mastitis control (1968–2024). **(a)** Absolute and cumulative number of publications over the years, showing the early and growth stages of research development. **(b)** Annual distribution of publications according to the origin of disinfectants investigated, classified as synthetic, plant-derived, animal-derived, or others. **(c)** Percentage proportion of disinfectant categories evaluated, highlighting the predominance of synthetic compounds.

3.3. Efficacy variability and methodological challenges

Several studies reported discrepancies between *in vitro* and *in vivo* outcomes. For instance, chlorhexidine and quaternary ammonium compounds showed high efficacy *in vitro* but moderate results under field conditions, suggesting the importance of environmental factors and application protocol adherence (Schwenker et al., 2022).

Phenolic compounds and simple chlorine-based disinfectants, when applied without standardized protocols, showed low efficacy (<50%) (Oliver et al., 2001; Peters et al., 2000), reinforcing the need for proper concentration and application standardization.

3.4. Average costs, environmental impacts, and sustainability

Although specific economic data remain limited, synthetic compounds such as iodophors tend to be cost-effective and widely accessible. However, their indiscriminate use can generate significant environmental

impacts, including selection of resistant strains and contamination of soil and water (Aiemsaard et al., 2023; Arabe Filho et al., 2023).

Natural and biological alternatives are less toxic to the environment but require higher investment in formulation stability, standardization, and scalability (Dias, 2019; Spilki, 2021). Balancing efficacy, cost-efficiency, and sustainability remains a growing challenge in dairy production systems.

3.5. Advantages and disadvantages of different approaches

Synthetic disinfectants: high efficacy, low cost, readily available, but associated with environmental toxicity and risk of antimicrobial resistance.

Natural disinfectants: environmentally safer, good efficacy, but higher production cost and limited standardization.

Biological agents: promising against multidrug-resistant strains, though still largely experimental and facing regulatory hurdles.

Table 1. Efficacy of disinfectant compounds against key mastitis-causing pathogens.

Type of Study	Evaluated Compounds	Tested Concentrations	Sample Collection Site	Evaluated Microorganisms	Efficacy	Authors
In vivo	Iodine	-	Bovine milk	<i>Staphylococcus aureus</i> , <i>Streptococcus dysgalactiae</i>	Very High	Sheldrake and Hoare (1980)
In vivo	Iodine	1000 mg av/L and 5000 mg av/L	Bovine udder	<i>Staphylococcus aureus</i> , <i>Streptococcus dysgalactiae</i>	Not specified	Sheldrake et al. (1980a)
In vivo	Iodine	-	Bovine udder	<i>Staphylococcus aureus</i> , <i>Streptococcus agalactiae</i>	Moderate	Grindal and Priest (1989)
In vivo	Iodine	0.25%	Bovine milk	<i>Staphylococcus aureus</i> , <i>Escherichia coli</i>	High	Oliver et al. (1991)
In vivo	Iodine	2.5% or 5% w/w; Chlorhexidine 0.3% w/w; Salicylic acid 0.1% w/w	Bovine milk/Bovine udder	<i>Streptococcus uberis</i>	Moderate	Lopez-Benavides et al. (2009)
In vivo	Iodine-containing gel	0.0002%	Bovine milk/Bovine udder	<i>Staphylococcus aureus</i> , <i>Streptococcus dysgalactiae</i>	Very High	Ingawa et al. (1992)
In vivo	Iodine, iodine and glycerin, iodine and emulsified paraffin	-	Bovine udder	<i>Staphylococcus aureus</i> , <i>Streptococcus dysgalactiae</i>	Very High	Sheldrake et al. (1980b)
In vivo	Iodine with a high level of free iodine	-	Bovine milk	<i>Streptococcus</i> spp., <i>Corynebacterium</i> spp.	Low	Martins et al. (2017)
In vivo	Iodine complex and skin conditioning agents	0.5% and 1.0% iodine	Bovine milk	<i>Staphylococcus aureus</i> , <i>Streptococcus agalactiae</i>	High	Fox et al. (2006)
In vitro	Iodine, Chlorhexidine	-	Bovine milk	Coagulase-negative <i>Staphylococcus</i> spp.	Very High	Tremblay et al. (2014)
In vitro	Iodine, Lactic acid, Chlorhexidine digluconate, Salicylic acid, Ammonium lauryl sulfate, Diamine, Chlorine dioxide, Didecyl dimethylammonium, Povidone-iodine	Lactic acid 2.5% or 5% w/w, chlorhexidine 0.3% w/w or salicylic acid 0.1% w/w, Dichloroisocyanurate: ≥500 µg/mL; Povidone-iodine: ≥8,000 µg/mL	Bovine milk	<i>Enterococcus faecalis</i> , <i>Hafnia alvei</i> , <i>Serratia marcescens</i> , <i>Serratia liquefaciens</i> , <i>Aerococcus viridans</i> , <i>Lactococcus lactis</i> , <i>Staphylococcus chromogenes</i> , <i>Staphylococcus devriesii</i> , <i>Staphylococcus epidermidis</i> , <i>Staphylococcus haemolyticus</i> , <i>Staphylococcus hominis</i> , <i>Staphylococcus xylosum</i>	Low	Fitzpatrick et al. (2019)

Effectiveness of the tested products expressed as percentage of inhibition: < 50% – Low effectiveness; 50 to 75% – Moderate effectiveness; 75 to 90% – High effectiveness; > 90% – Very high effectiveness.

Table 1. Continued...

Type of Study	Evaluated Compounds	Tested Concentrations	Sample Collection Site	Evaluated Microorganisms	Efficacy	Authors
In vitro	Iodine, Sodium hypochlorite, Sodium dichloro-s-triazinetriene, Quaternary ammonium, Chlorhexidine, Cetylpyridinium chloride, Bronopol, 8-Hydroxyquinoline sulfate, Pine oil, Hexachlorophene, Udder antibodies	0.0390625–0.15625%	Not specified	<i>Staphylococcus aureus</i> , <i>Streptococcus agalactiae</i>	Very High	Philipot et al. (1978)
In vitro	Iodine, Quaternary ammonium Dodecylbenzenesulfonic acid	Lactic acid 2.5% or 5% w/w, and Chlorhexidine 0.3% w/w or Salicylic acid 0.1%	Bovine milk	<i>Prototheca zopfii</i>	Low	Lassa et al. (2011)
In vivo	Iodine (post-milking), Chloramine-T (pre and post-milking)	-	Bovine milk	<i>Staphylococcus aureus</i> , <i>Streptococcus uberis</i> , <i>Corynebacterium</i> spp., and Coagulase-negative <i>Staphylococcus</i> spp.	Low	Williamson and Lacy-Hulbert (2013)
In vitro	Iodine, H ₂ O ₂ , Chlorine dioxide	Iodophors: 0.5% and 0.25%	Bovine milk	<i>Mycoplasma bovis</i>	Very High	Enger et al. (2015)
In vivo	Iodophor, Hexachlorophene, Chlorhexidine,		Bovine milk, Bovine udder	<i>Staphylococcus haemolyticus</i> <i>Staphylococcus</i> spp.	High Low	Edwards and Smith (1970)
In vitro	Iodophor, Hypochlorite, Chlorhexidine, Iodine (in oil), Sodium dichloro-s-triazinetriene, Quaternary ammonium, Dodecyl benzene sulfonic acid	Iodophor: 1% and 0.5%, Dodecyl benzene sulfonic acid 2%	Bovine milk	<i>Staphylococcus aureus</i> <i>Streptococcus agalactiae</i>	High Moderate	Philipot and Pankey (1978)
In vivo	Iodophor solution, Sodium hypochlorite solution	Iodophor solution: not specified; sodium hypochlorite solution: MIC between 0.25 and 8 mg/mL	Bovine milk/Bovine udder	<i>Staphylococcus aureus</i>	Very High	King et al. (1979)
In vitro	Iodophor, Chlorhexidine, Quaternary ammonium, Cetylpyridinium chloride, Benzyl alcohol	Iodophor (0.2%) / Chlorhexidine (0.2%) / Quaternary ammonium (0.2%) / Cetylpyridinium chloride / Benzyl alcohol	Not specified	<i>Staphylococcus aureus</i> , <i>Streptococcus agalactiae</i>	Very High	Watts et al. (1984)

Effectiveness of the tested products expressed as percentage of inhibition: < 50% – Low effectiveness; 50 to 75% – Moderate effectiveness; 75 to 90% – High effectiveness; > 90% – Very high effectiveness.

Table 1. Continued...

Type of Study	Evaluated Compounds	Tested Concentrations	Sample Collection Site	Evaluated Microorganisms	Efficacy	Authors
In vivo	Iodophor solution, Iodine	-	Bovine milk	<i>Streptococcus uberis</i> , <i>Staphylococcus aureus</i> , <i>Mycoplasma bovis</i> , Coagulase-negative <i>Staphylococcus</i> , <i>Escherichia coli</i>	Not specified	Hillerton et al. (1993)
In vivo	Iodophor (Full-Bac), Iodophor (Bovadine)	0.001 to 0.035%	Bovine milk/Bovine udder	<i>Staphylococcus aureus</i> , <i>Streptococcus agalactiae</i>	Low	Leslie et al. (2005)
In vitro	N, N-Dimethyldodecanamine ¹	-	Not specified	<i>Streptococcus agalactiae</i> , <i>Escherichia coli</i>	High	Turck et al. (1982)
In vivo	Quaternary ammonium	-	Bovine milk	<i>Streptococcus agalactiae</i>	Moderate	Pankey and Watts (1983)
In vitro	Quaternary ammonium	8-35 ppm	Bovine milk	<i>Prototheca bovis</i>	-	Arabe Filho et al. (2023)
In vivo	Chlorous acid gel and chlorine dioxide	-	Bovine milk	<i>Staphylococcus aureus</i> , <i>Streptococcus dysgalactiae</i>	Moderate	Oliver et al. (1989)
In vivo	Chlorous acid - chlorine dioxide	-	Bovine milk/Bovine udder	<i>Streptococcus uberis</i> , <i>Corynebacterium bovis</i>	Low	
In vivo	Chlorous acid - chlorine dioxide, Sodium hypochlorite	0.5% and 0.25%	Bovine milk	<i>Staphylococcus aureus</i>	High	Drechsler et al. (1990)
In vitro	Chlorine, Iodine, Chlorhexidine, Quaternary ammonium, Lactic acid	0.0024- 0.0190%	Bovine milk	<i>Streptococcus agalactiae</i>	Moderate	
In vitro	Chlorine, Iodine, Chlorhexidine, Quaternary ammonium, Lactic acid	-	Bovine milk	<i>Staphylococcus aureus</i> , Coagulase-positive <i>Staphylococcus</i>	Very High	Boddie et al. (1998)
In vivo	Chlorine dioxide (new formulation), Chlorine dioxide (established formulation)	-	Bovine milk	<i>Staphylococcus aureus</i> , Coagulase-positive <i>Staphylococcus</i>	Very High	Medeiros et al. (2009)
In vivo	Chlorine, Hibitane, Iodovet, Iosan	-	Bovine milk	Not specified	Moderate	Krömker et al. (2023)
In vivo	Chlorine dioxide, Glycerol	Glycerol 10% and chlorine dioxide 120 ppm	Teat cup	<i>Staphylococcus spp.</i> and <i>Streptococcus spp.</i>	Very High	Saucier et al. (1968)
In vivo	Chlorine dioxide, Glycerol	-	Bovine udder	<i>Streptococcus uberis</i> , <i>Staphylococcus aureus</i>	Low	Rasmussen and Larsen (1998)

Effectiveness of the tested products expressed as percentage of inhibition: < 50% – Low effectiveness; 50 to 75% – Moderate effectiveness; 75 to 90% – High effectiveness; > 90% – Very high effectiveness.

Table 1. Continued...

Type of Study	Evaluated Compounds	Tested Concentrations	Sample Collection Site	Evaluated Microorganisms	Efficacy	Authors
In vitro	Microspheres composed of poly dl-lactide-co-glycolide (DPLGA) and povidone-iodine	-	Bovine udder	<i>Staphylococcus aureus</i> , <i>Streptococcus</i> spp., <i>Streptococcus uberis</i>	Low	Oliver et al. (1993)
In vivo	Phenol at 1.6%	MIC90 \geq 0.312 g/L	Bovine milk/Bovine udder	<i>Escherichia coli</i> , <i>Staphylococcus aureus</i>	Low	Peters et al. (2000)
In vivo	Disinfectant based on a phenolic combination	MIC90 \geq 0.019 g/L	Bovine milk	Gram-negative bacteria, <i>Streptococcus dysgalactiae</i> , Coagulase-negative <i>Staphylococcus</i>	Low	Oliver et al. (2001)
In vivo	Phenolic combination	MIC90 \geq 0.625 g/L	Bovine milk	<i>Streptococcus uberis</i> , <i>Staphylococcus aureus</i>	Low	Oliver et al. (1999)
In vivo	Lactic acid and Sodium hypochlorite	0.0002%	Bovine milk	Not specified	Low	Park and Han (2002)
In vivo	Lactic acid, Lauricidin, Caprylic and Capric acids, Lauric acid	-	Bovine milk/Bovine udder	<i>Staphylococcus aureus</i> , <i>Streptococcus agalactiae</i>	High	Boddie and Nickerson (1992)
In vivo	Bismuth subnitrate (Teatseal and Cepravin Dry Cow)	-	Bovine milk	<i>Escherichia coli</i> , Enterobacteriaceae, Other pathogens	High	Huxley et al. (2002)
In vivo	Acidified chlorite with sodium dodecylbenzene sulfonic acid	-	Bovine milk/Bovine udder	<i>Staphylococcus aureus</i> , <i>Streptococcus agalactiae</i>	High	Oura et al. (2002)
In vivo	Hydrogen peroxide	0.5%	Bovine udder	<i>Staphylococcus aureus</i> , <i>Streptococcus agalactiae</i>	Low	Leslie et al. (2006)
In vivo	Acidified sodium hypochlorite disinfectant and iodine	-	Bovine milk	Not specified	Moderate	Hillerton et al. (2007)
In vitro	Decoction of Hypericum caprifoliatum Cham. and Schlecht. - Guttiferae	500 mg/L	Not specified	<i>Staphylococcus aureus</i>	Very High	Avancini and Wiest (2008)
In vitro	Biocitro ®, Eco Plus ®, Combicid ®, Prodip G ®	500 mg/L	Bovine milk	<i>Prototheca zopfii</i>	Very High	Lopes et al., 2008
In vitro	Essential oils of citronella grass, lemongrass, kafir lime, holy basil, sweet basil, turmeric, and grapevine	MIC between 0.25 and 8 mg/mL	Not specified	<i>Pseudomonas aeruginosa</i>	Very High	Aiemsard et al. (2010)

Effectiveness of the tested products expressed as percentage of inhibition: < 50% – Low effectiveness; 50 to 75% – Moderate effectiveness; 75 to 90% – High effectiveness; > 90% – Very high effectiveness.

Table 1. Continued...

Type of Study	Evaluated Compounds	Tested Concentrations	Sample Collection Site	Evaluated Microorganisms	Efficacy	Authors
In vivo	Essential oils of clove, citronella, and basil	MIC and MBC: 0.78–6.25 µL/mL; Citronella with 0.54% iodine	Bovine udder	<i>Staphylococcus aureus</i> , <i>Streptococcus agalactiae</i> , <i>Escherichia coli</i>	Very High	Aiemsaaard et al. (2023)
In vivo	Essential oil of Lippia organoïdes	120 µL/mL	Bovine milk	<i>Staphylococcus</i> spp., <i>Streptococcus</i> spp.	High	Marcelo et al. (2020)
In vitro	Sodium hypochlorite, Iodine	Sodium hypochlorite: 0.0390625–0.15625%; Iodine: 0.15625–0.625%	Bovine milk	<i>Prototheca zopfii</i>	Very High	Salerno et al. (2010)
In vivo	Sodium dichloroisocyanurate, Bronopol, Quaternary ammonium	Sodium dichloroisocyanurate (0.25%), Bronopol (0.1%), Quaternary ammonium (0.2%)	Bovine milk	<i>Staphylococcus aureus</i> , <i>Streptococcus agalactiae</i>	Moderate	Boddie and Nickerson (2002)
In vivo	Sodium hypochlorite in combination with other compounds	-	Bovine udder	<i>Staphylococcus aureus</i> , <i>Streptococcus agalactiae</i>	Very High	Pankey et al. (1983)
In vitro	Silver ion	Less than 2 ppm	Bovine udder	<i>Staphylococcus aureus</i>	Very High	Seol et al. (2010)
In vitro	Silver nanoparticles, Silver nitrate	-	Bovine milk	<i>Staphylococcus aureus</i> , <i>Escherichia coli</i>	Very High	Fernandes et al. (2021)
In vitro	Nonoxinol-9 Iodine Complex, Chlorhexidine digluconate	MIC of 32 µM and MBC of 32 µM	Bovine milk/Bovine udder	<i>Staphylococcus aureus</i>	Very High	El Behiry et al. (2012)
In vitro	Marine sponges	MIC of 1.024 µg/mL	Bovine milk	Coagulase-negative <i>Staphylococcus</i> spp.	Moderate	Laport et al. (2012)
In vivo	Theratec Plus (Iodine)	-	Bovine milk	Not specified	Very High	Ceballos-Marquez et al. (2013)
In vitro	Peracetic acid, Sodium hypochlorite, Iodine solution	Peracetic acid: MIC90 ≥0.019 g/L; Sodium hypochlorite: MIC90 ≥0.312 g/L; Iodine solution: MIC90 ≥0.625 g/L	Bovine milk	<i>Prototheca zopfii</i>	High	Gonçalves et al. (2015)
In vivo	Peracetic acid (PAS), Plasma-activated buffered solution (PABS)	MS-TMC-Flusc: not specified, PAS: 0.5%, and PABS: not specified	Teatcup	<i>Escherichia coli</i> , <i>Staphylococcus aureus</i> , <i>Streptococcus uberis</i> , <i>Streptococcus agalactiae</i>	High	Scheib et al. (2023)
In vitro	Crude hydroalcoholic extract of <i>Achyrocline satureioides</i> (Asteraceae)	-	Bovine milk	<i>Candida albicans</i> , <i>Candida krusei</i> , <i>Candida rugosa</i>	Very High	Campos et al. (2016)

Effectiveness of the tested products expressed as percentage of inhibition: < 50% – Low effectiveness; 50 to 75% – Moderate effectiveness; 75 to 90% – High effectiveness; > 90% – Very high effectiveness.

Table 1. Continued...

Type of Study	Evaluated Compounds	Tested Concentrations	Sample Collection Site	Evaluated Microorganisms	Efficacy	Authors
In vivo	Glycolic acid, Iodine	-	Bovine milk	Not specified	Low	Godden et al. (2016)
In vivo	Glycolic acid, Iodine	0.6% diamine or 0.5% iodine	Bovine milk/Bovine udder	Coagulase-negative <i>Staphylococcus</i> , <i>Streptococcus</i> spp.	Moderate	Lago et al. (2016)
In vitro	Guanidine	-	Bovine milk	<i>Prototheca zopfii</i>	Very High	Alves et al. (2017)
In vitro	Slightly acidic electrolyzed water	-	Bovine milk, Hoses, jets, and water reservoir walls	<i>Pseudomonas</i> spp.	Very High	Kawai et al. (2017)
In vivo	Probiotic teat disinfectant based on lactobacilli	120 µL/mL	Bovine milk	<i>Pseudomonas</i> spp., <i>Acinetobacter</i> spp., <i>Streptococcus</i> spp., <i>Bacillus</i> spp.	Very High Low	Yu et al. (2017)
In vitro	SunSmile® Fruit & Vegetal	0.0024–0.0190%	Bovine milk	<i>Prototheca zopfii</i>	Low	Grzesiak et al. (2018)
In vitro	Gentisaldehyde (2,5-dihydroxybenzaldehyde), 2,3-dihydroxybenzaldehyde	500 mg/L	Bovine milk	<i>Staphylococcus aureus</i>	Moderate	Schabauer et al. (2018)
In vivo	ZincCu	-	Bovine milk/Bovine udder	<i>Streptococcus uberis</i> , <i>Corynebacterium</i> spp., Coagulase-negative <i>Staphylococcus</i>	Low	Vissio et al. (2020)
In vivo	Bactofencin A, Nisin, Reuterin, Bacteriocin	-	Bovine udder	<i>Staphylococcus</i> spp., <i>Streptococcus</i> spp.	Moderate	Bennett et al. (2022)
In vitro	Mannosylerythritol lipid-B	Above 10 ppm	Not specified	Total bacteria <i>Staphylococcus aureus</i> , <i>Escherichia coli</i>	Low Very High	Yamauchi et al. (2022)
In vitro	Polyhexamethylene biguanide	MIC90 ≥2 µg/mL and MBC90 ≥4 µg/mL for <i>P. bovis</i> ; ≥1 µg/mL for <i>P. blaschkeae</i>	Bovine milk/Bovine udder	<i>Pseudoblastesikia blaschkeae</i>	Very High	Fidelis et al. (2023)
In vitro	Polyhexamethylene biguanide (nanoparticles)	MIC <0.003 µg/mL	Bovine milk	<i>Prototheca</i> spp., <i>Prototheca bovis</i>	High	Leite et al. (2021)
In vitro	Polyhexamethylene biguanide	3 g/L	Chinese Veterinary Culture Collection Center	<i>Streptococcus agalactiae</i> , <i>Streptococcus dysgalactiae</i> , <i>Staphylococcus aureus</i> , <i>Escherichia coli</i>	Very High	Lu et al. (2023)

Effectiveness of the tested products expressed as percentage of inhibition: < 50% – Low effectiveness; 50 to 75% – Moderate effectiveness; 75 to 90% – High effectiveness; > 90% – Very high effectiveness.

Table 1. Continued...

Type of Study	Evaluated Compounds	Tested Concentrations	Sample Collection Site	Evaluated Microorganisms	Efficacy	Authors
In vivo	Chlorhexidine	8% and 50%	Bovine udder	<i>Staphylococcus aureus</i> , <i>Streptococcus uberis</i> , <i>Escherichia coli</i> , <i>Candida albicans</i>	Very High	Kybartas et al. (2023)
In vitro	Chlorhexidine	Chlorhexidine 2%	Teats	<i>Enterococcus faecalis</i> , <i>Enterococcus faecium</i> , <i>Staphylococcus intermedius</i> , <i>Streptococcus uberis</i> , <i>Streptococcus dysgalactiae</i>	Very High	Santos et al. (2023)
In vivo	Chlorhexidine	0.35%	Bovine milk/Bovine udder	<i>Streptococcus</i> spp., <i>Escherichia coli</i> , Coagulase-negative <i>Staphylococcus</i>	Low	Oliver et al. (1994)
In vitro	Chlorhexidine, Lactic acid	Chlorhexidine 0.215% and Lactic acid 3.5%	Bovine milk/Bovine udder	<i>Staphylococcus aureus</i> , <i>Staphylococcus epidermidis</i> , <i>Staphylococcus haemolyticus</i> , <i>Staphylococcus chromogenes</i> , <i>Staphylococcus equorum</i> , <i>Staphylococcus sciuri</i> , <i>Staphylococcus xylosum</i> , <i>Aerococcus viridans</i> , <i>Streptococcus uberis</i> , <i>Corynebacterium</i> spp., Other coagulase-negative <i>Staphylococci</i> .	Moderate	Schwenker et al. (2022)
In vivo	Chlorhexidine	MIC of 1.024 µg/mL	Bovine milk/Bovine udder	<i>Streptococcus</i> spp., Coagulase-negative <i>Staphylococcus</i> , <i>Corynebacterium bovis</i>	Moderate	Oliver et al. (1990)
In vitro	Chlorhexidine gluconate solution and sodium hypochlorite	Chlorhexidine 2% and hypochlorite 0.5%	Recycled bedding sand	<i>Mycoplasma</i> spp., <i>Mycoplasma bovis</i>	Very High	Justice-Allen et al. (2010)
In vitro	Chlorhexidine, Quetocromin, Dihydroergocristine, Michellamine B, Toyocamycin, Emodin, Tanshinone IIA, Curcumol	β-lapachone: MIC 32 µM and MBC 32 µM	Bovine udder	<i>Streptococcus uberis</i>	Very High	Ferguson et al. (2019)
In vitro	Chlorhexidine, Quetocromin, Dihydroergocristine, Michellamine B, Toyocamycin, Emodin, Tanshinone IIA, Curcumol	β-lapachone: MIC 32 µM and MBC 32 µM	Bovine udder	<i>Escherichia coli</i>	Low	Ferguson et al. (2019)

Effectiveness of the tested products expressed as percentage of inhibition: < 50% – Low effectiveness; 50 to 75% – Moderate effectiveness; 75 to 90% – High effectiveness; > 90% – Very high effectiveness.

Table 1. Continued...

Type of Study	Evaluated Compounds	Tested Concentrations	Sample Collection Site	Evaluated Microorganisms	Efficacy	Authors
In vitro	Chlorhexidine	0.0002%	Bovine milk	<i>Staphylococcus aureus</i>	Very High	Azizoglu et al. (2013)
In vitro	Iodine	-	Bovine udder	<i>Staphylococcus aureus</i> , <i>Streptococcus agalactiae</i> , <i>Escherichia coli</i> , <i>Nocardia asteroides</i>	Dose-dependent	Larocque et al. (1992)
	Chlorhexidine acetate	Linear dodecyl benzene sulfonic acid (1.94%) and iodine (1%), ethanol-iodine complex: not specified				
In vitro	Chlorhexidine gluconate, Nonyl phenoxy poly (ethyleneoxy) iodine, Ethanol-iodine complex	-	Not specified	<i>Prototheca zopfii</i>	Very High	Sobukawa et al. (2011)
In vivo	Chlorhexidine, Alkyl Dimethyl Glycine	1.94%	Bovine udder	<i>Staphylococcus aureus</i> <i>Streptococcus agalactiae</i>	High	Pankey et al. (1984)
	Hydrochloride, Chlorine dioxide, Povidone-iodine, Sodium hypochlorous acid					
In vivo	Chlorhexidine gluconate, Dodecyl benzene sulfonic acid, Iodine, Sodium dichloro-s-triazinetriene, Povidone-iodine	-	Teats	<i>Enterococcus faecalis</i> , <i>Enterococcus faecium</i> , <i>Staphylococcus intermedius</i> , <i>Streptococcus uberis</i> , <i>Streptococcus dysgalactiae</i>	Low	Santos et al. (2023)
	Neutral detergent, Acid detergent, Chlorinated alkaline detergent, Chlorinated alkaline detergent at 45°C, Distilled water at 45°C					
In vitro	<i>Lactococcus cremoris</i>	-	Bovine milk	<i>Staphylococcus spp.</i> , <i>Streptococcus spp.</i> , <i>Enterococcus spp.</i> , <i>Escherichia coli</i>	High	Addis et al. (2024)
In vitro	<i>Lactococcus cremoris</i>	-	Bovine udder	<i>Staphylococcus aureus</i> , <i>Streptococcus spp.</i>	Very High	Gazzola et al. (2024)
In vivo	Phage cocktail named SPBC-Sj	0>10 ⁷ CFU/mL	Bovine milk/Bovine udder	<i>Escherichia coli</i> , <i>Staphylococcus aureus</i> , <i>Pseudomonas aeruginosa</i>	High	Xue et al. (2024)

Effectiveness of the tested products expressed as percentage of inhibition: < 50% – Low effectiveness; 50 to 75% – Moderate effectiveness; 75 to 90% – High effectiveness; > 90% – Very high effectiveness.

3.6. Final considerations

Disinfectant selection must consider the target pathogen, application method, appropriate concentration, and specific production context. Evidence-based protocols are essential to ensure efficacy, minimize environmental risks, and promote the sustainability of dairy production systems.

4. Conclusion

This review highlighted that the use of disinfectants is a key strategy in the prevention and control of bovine mastitis, contributing significantly to the reduction of microbial load on teats before and after milking. The diversity of compounds evaluated in the analyzed studies—including iodine, chlorhexidine, hypochlorite, lactic acid, peroxides, and biguanides—reflects the ongoing efforts of the scientific community and the dairy industry in the search for effective and safe solutions.

The literature review provides an in-depth scientific analysis of the applications of disinfectants in the control of bovine mastitis. Despite the wide variety of formulations and evaluation methods employed, many studies converge on the efficacy of certain active ingredients, especially iodine and chlorhexidine. However, the results also reveal significant gaps, notably the lack of research on alternative and natural compounds, as well as the need for methodological standardization in efficacy testing.

A predominance of studies conducted in North America and Western Europe was observed, with the United States and Brazil standing out as the main contributors to research in this area. Despite the wide variety of formulations and evaluation methods used, many studies converge on the efficacy of certain active ingredients, especially iodine and chlorhexidine, which are widely recognized for their consistent antimicrobial activity. However, the findings also reveal significant gaps, such as the lack of investigations into alternative and natural compounds, as well as the need for methodological standardization in efficacy testing.

The results emphasize the importance of a global and multidisciplinary approach that integrates microbiological, toxicological, and environmental aspects in the selection and use of disinfectants, also considering the risk of antimicrobial resistance. Therefore, further research is needed to explore sustainable and economically viable formulations that align with the demands of modern dairy production and animal welfare principles.

In conclusion, disinfectants remain an indispensable tool for managing bovine mastitis, but their selection and application should be guided by robust scientific evidence, contextualized to different regional and productive realities.

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Data Availability Statement

The research data analyzed in this study are not publicly available by any means. However, the essential data supporting the results are presented within the article itself, in the form of tables and figures. Additional information may be made available upon reasonable request to the corresponding author, considering potential ethical and institutional limitations.

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