

Ketoprofen as the sole initial treatment for nonsevere bovine mastitis: Efficacy and antibiotic reduction

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Abstract

Targeted mastitis treatment concepts are the most scientifically validated approaches to comply with the prudent use of antimicrobials. However, this approach is still a challenge for many farmers, who often lack experience with it. Another option for more prudent use of antibiotics in mastitis therapy could be the initial treatment with a nonsteroidal anti-inflammatory drug (NSAID) for nonsevere mastitis. A randomized, multiherd, noninferiority study was carried out to compare 2 treatments of nonsevere mastitis cases. The animals in the studied group were treated with systemic NSAID (ketoprofen [KE]; KE group; n = 104), followed by antibiotic (AB) treatment only in cases without clinical improvement, and the animals in the reference group (AB group) received an intramammary AB (n = 118). The study included 222 cases of nonsevere clinical mastitis on 3 conventional dairy farms in northern Germany between November 2022 and November 2023. Study outcomes were clinical cure at d 5, bacteriological cure, clinical mastitis recurrence in a period of 60 d, and new infection risk. Mixed logistic regression was used to analyze the effect of treatment on outcomes. The noninferiority assumption was checked with the CI of the treatment difference for clinical cure at d 5, calculating using the LSM and their SD. With regard to the clinical cure on d 5, the treatment in the KE group was inferior. Clinical cure (84.7% [100/118] and 61.5% [64/104]) and bacteriological cure (79.3% [73/92] and 61.2% [41/67]) were significantly higher in the AB group than in the KE group. The risks for recurrent cases (7.3% [8/109] and 15.7% [14/89]) were significantly lower in the AB group than in the KE group. The new infection risk did not differ significantly between the treatment groups (6.8% [8/118] in the AB group and 6.7% [7/104] in the KE group). In 87% of the cases (n = 90) of the KE group, a subsequent antibiotic treatment was not necessary to reach clinical cure on d 5. The use of KE as the sole initial treatment for nonsevere mastitis led to a reduction of antibiotic doses by 86%. Our findings revealed that systemic treatment with KE resulted in lower clinical and bacteriological cure rates, higher recurrence rates of clinical mastitis, and an elevated risk of new infections compared with the antibiotic-treated group. Nevertheless, in more than 85% of cases treated with KE, additional intramammary antibiotic treatment was unnecessary. Thus, this treatment concept represents an alternative

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in mastitis treatment that promoted the deliberate and selective use of antibiotics but has limitations in terms of treatment efficacy in mastitis. A targeted mastitis concept based on the identification of the bacterium involved could help to select cases to be treated with KE alone and therefore overcome this limitation.

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Ketoprofen as the sole initial treatment for nonsevere bovine mastitis: Efficacy and antibiotic reduction

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ABSTRACT

Targeted mastitis treatment concepts are the most scientifically validated approaches to comply with the prudent use of antimicrobials. However, this approach is still a challenge for many farmers, who often lack experience with it. Another option for more prudent use of antibiotics in mastitis therapy could be the initial treatment with a nonsteroidal anti-inflammatory drug (NSAID) for nonsevere mastitis. A randomized, multiherd, noninferiority study was carried out to compare 2 treatments of nonsevere mastitis cases. The animals in the studied group were treated with systemic NSAID (ketoprofen [KE]; KE group; $n = 104$), followed by antibiotic (AB) treatment only in cases without clinical improvement, and the animals in the reference group (AB group) received an intramammary AB ($n = 118$). The study included 222 cases of nonsevere clinical mastitis on 3 conventional dairy farms in northern Germany between November 2022 and November 2023. Study outcomes were clinical cure at d 5, bacteriological cure, clinical mastitis recurrence in a period of 60 d, and new infection risk. Mixed logistic regression was used to analyze the effect of treatment on outcomes. The noninferiority assumption was checked with the CI of the treatment difference for clinical cure at d 5, calculating using the LSM and their SD. With regard to the clinical cure on d 5, the treatment in the KE group was inferior. Clinical cure (84.7% [100/118] and 61.5% [64/104]) and bacteriological cure (79.3% [73/92] and 61.2% [41/67]) were significantly higher in the AB group than in the KE group. The risks for recurrent cases (7.3% [8/109] and 15.7% [14/89]) were significantly lower in the AB group than in the KE group. The new infection risk did not differ significantly between the treatment groups (6.8% [8/118] in the AB group and

6.7% [7/104] in the KE group). In 87% of the cases ($n = 90$) of the KE group, a subsequent antibiotic treatment was not necessary to reach clinical cure on d 5. The use of KE as the sole initial treatment for nonsevere mastitis led to a reduction of antibiotic doses by 86%. Our findings revealed that systemic treatment with KE resulted in lower clinical and bacteriological cure rates, higher recurrence rates of clinical mastitis, and an elevated risk of new infections compared with the antibiotic-treated group. Nevertheless, in more than 85% of cases treated with KE, additional intramammary antibiotic treatment was unnecessary. Thus, this treatment concept represents an alternative in mastitis treatment that promoted the deliberate and selective use of antibiotics but has limitations in terms of treatment efficacy in mastitis. A targeted mastitis concept based on the identification of the bacterium involved could help to select cases to be treated with KE alone and therefore overcome this limitation.

Key words: bovine, mastitis treatment, NSAID, cure, reduction of antibiotic usage

INTRODUCTION

In recent years, evidence has accumulated from clinical trials on the treatment of clinical mastitis (CM) indicating that antibiotic (AB) treatment is not necessary in every case of mastitis (Mansion-de Vries et al., 2016; Schmenger and Krömker, 2020; de Jong et al., 2023). In nonsevere mastitis cases (mild and moderate), AB therapy is only necessary if gram-positive microorganisms are detected (Roberson et al., 2004; Krömker et al., 2010). Therefore, because some cases show no pathogen growth or gram-negative pathogen growth, local AB therapy is not indicated here. The goal of AB treatment is a bacteriological cure (BC), which is to a certain extent influenced by the treatment-independent self-cure rate of the animal. Antibiotic treatment should be carried out if it significantly improves the BC compared with the self-cure rate. This is especially the case in infections

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The list of standard abbreviations for JDS is available at adsa.org/jds-abbreviations-25. Nonstandard abbreviations are available in the Notes.

caused by gram-positive pathogens (Krömker et al., 2010). However, it has been demonstrated that infections with gram-negative pathogens have a high tendency to self-cure and AB treatment does not particularly improve BC (Roberson et al., 2004; Suojala et al., 2010). In addition, AB treatment is not recommended in cases without evidence of pathogens (Roberson, 2003).

Minimizing and optimizing AB consumption, a declared political goal (EU [VO] 2019/6; European Union, 2019), is particularly important in light of the increasing development of bacterial resistance. To address the challenge of increasing development of bacterial resistance, targeted mastitis therapy has emerged as a promising approach, incorporating on-farm microbiological diagnostics as a core element, significantly reducing AB use without compromising cure rates. Within this therapeutic framework, which also considers the animal's clinical condition and other animal-specific factors, the use of nonsteroidal anti-inflammatory drugs (NSAID) is recommended for all CM cases (Mansion-de Vries et al., 2016; Schmenger et al., 2020). This not only positively affects clinical cure (CC) but also enhances the animal's well-being (McDougall et al., 2009; Mansion-de Vries et al., 2015). As a supportive treatment, NSAID are already used regularly in many European countries. Nonsteroidal anti-inflammatory drugs inhibit cyclooxygenase enzymes, resulting in reduced synthesis of some inflammatory mediators, particularly prostaglandin E₂. There may therefore be a concern that reducing the inflammatory process in mastitis may limit the healing of the infected quarter. However, it has been shown experimentally in mice that ketoprofen (KE) reduces immunosuppression and increases the animal's response to sepsis (Brogliato et al., 2012). In cattle with CM, the addition of parenteral KE to AB treatment for several days increases the cure rate compared with AB treatment alone (Shpigel et al., 1994). Moreover, a recent study conducted in Germany (Krömker et al., 2021) demonstrated that, in cases of nonsevere CM in cows with long-lasting udder issues, AB treatment does not provide advantages over KE, an NSAID treatment.

The life of the animal is typically not endangered in nonsevere CM cases, as defined by the International Dairy Federation (IDF; IDF, 2011). This means that commencing treatment with an NSAID seems to offer a potential substitute for the use of AB in addressing nonsevere CM cases. Some farmers even use NSAID (especially KE and carprofen) as a first-line treatment when on-farm testing identifies a gram-negative pathogen (Preine et al., 2022). Therefore, in cases of nonsevere CM, initial treatment exclusively with an NSAID could be a possible alternative and a first step toward a treatment guided by substantiated evidence.

Overhauling the treatment regimen on dairy farms often presents some challenges, and the extra diagnostic

work demand required when applying a targeted form of therapy frequently necessitates a reorganization of daily work routines. Furthermore, using KE as the sole initial treatment for nonsevere CM cases could be a strategy that maintains the flexibility to transition to AB treatment, which is typically administered when no improvement in clinical presentation is observed.

This study aimed to evaluate the efficacy of a specific therapy concept including sole systemic KE treatment compared with a reference therapy with intramammary AB treatment in terms of CC at d 5 (CC5), BC, CM recurrence in a period of 60 d (R60), and new infection risk (NI) in the treatment of nonsevere CM. In addition, the potential reduction in the use of AB and the likely milk losses avoided by not using AB were examined. We hypothesized that the sole initial treatment of nonsevere CM cases with systemic administration of KE is not inferior to intramammary antimicrobial treatment in terms of CC5.

MATERIALS AND METHODS

Ethical Approval

This study was conducted in accordance with the guidelines on good clinical practice (EMA, 2000). Clinical trial registry number was 7221.3–2-007/22. The study complies with the Reporting Guidelines for Randomized Controlled Trials in Livestock and Food Safety (REFLECT checklist; O'Connor et al., 2010).

Farms and Cows

The study was carried out on 3 conventional dairy farms located in northern Germany (Mecklenburg-Western Pomerania) between August 2022 and November 2023. The farms were selected on the basis of their willingness to participate in this study and their participation in the DHI system. Herd sizes ranged between ~384 and 1,200 lactating German Holstein-Frisian dairy cows. The milk production ranged from 9,700 and 11,800 kg/cow per year. The bulk milk SCC varied between 170,000 and 320,000 cells/mL (annual average). Cows were milked 2 to 3 times per day in herringbone or rotary parlors. All 3 farms implemented reasonable hygiene management methods in housing, milking, and treating cows. All farms use approved postdipping agents based on iodine or chlorine dioxide. The milking clusters were disinfected after each milking with a 1,000 ppm (1,000 ppm = 1 g/kg) peracetic acid solution in a dipping process (draining tub). On all farms, premilking was done manually, and reusable wipes are used for cleaning (one per animal per milking session), a process that took ~10 s per animal. This was followed

by a waiting period of 50 to 80 s before the milking clusters are attached. All farms fed TMR. The farms' hygiene situation was continually monitored, evaluated, and optimized on the basis of dairy herd-specific indicators by the study personnel.

Inclusion criteria for the cows were the spontaneous occurrence of nonsevere (mild or moderate) CM and no AB treatment during the preceding 4 wk. Every cow had to be registered with a unique ear tag to clearly identify each animal. Lactating Holstein-Friesian dairy cows of all parities with mild or moderate CM signs in one udder quarter and only cows free of significant udder, teat, or teat orifice lesions or another additional disease at the same time were eligible for inclusion. The mastitis severity score was defined according to IDF guidelines (IDF, 2011). A CM case was classified as mild if there was only a change in the appearance of milk (color, viscosity, consistency). A moderate CM additionally showed local clinical signs of inflammation of the udder parenchyma (i.e., swelling, heat, pain, redness). In case of general clinical signs (fever, lack of appetite), the CM was classified as a severe mastitis. Before the start of the trial, it was decided that animals that would develop severe mastitis during the course of the trial should be classified as treatment failures.

Commonly used cow-level data, including lactation number, affected quarter location, cow SCC of the 3 most recent DHI recordings before CM, DIM at CM occurrence, and concurrent diseases and treatments for a period of 60 d after enrollment were recorded.

Sample Size Determination

Based on discussions with farmers in our region, the acceptable margin of noninferiority was set at 0.25 for this study. The 95% CI approach was used to calculate the required sample size based on the CC rate. The required number of cases was calculated based on the assumption that the actual difference (CC5) in favor of standard treatment is 8% (83% vs. 75%). This led to a case number of at least 196 CM cases (90% power and 5% significance limit and $d = 25\%$; i.e., the upper limit of a one-sided 95% CI or a 2-sided 90% CI excludes a difference in favor of the standard group of more than 25%). To obtain reliable results despite a premature loss of 10% of the animals, it was calculated that a total of 220 animals with CM had to be included. The calculations were performed with Sealed Envelope (<https://www.sealedenvelope.com>).

Randomization and Blinding

The cows were randomly assigned to one of the 2 groups using a randomization list. Because of the differ-

ences in treatment regimens and product administration, it was not possible to blind the farmers or herds persons to product administration because these people implemented the treatment depending on the treatment group. However, the treatments were carried out by different farm personnel than those who conducted the clinical examination in the milking parlor. The laboratory personnel performing microbiological analysis were unaware of the treatment given to the quarters being sampled and the study personnel were blinded and had no active influence on treatment protocols or the evaluation of results.

Treatment

If a case of CM occurred in an animal that met the inclusion criteria, classification of the severity score, the allocation to a treatment group, and the treatment were performed by instructed farm staff. Two different treatment regimens were compared in the study: animals of the control group (AB) received local antimicrobial treatment (intramammary; 2 applications at 24-h intervals with 200 mg cefalexin and 100,000 IU, kanamycin; Ubrolexin, Boehringer Ingelheim, Ingelheim am Rhein, Germany) according to the label of the product. Animals of the second group (KE) received systemic treatment with the KE intramuscularly (3 applications at 24-h intervals with 3 mg ketoprofen per kilogram of BW; Romefen PR 10%/Ketofen 10%, Ceva Animal Health, Düsseldorf, Germany; withdrawal time for milk was 0 d). In case of aggravation or no recovery of clinical signs during the following 3 d, intramammary antimicrobial treatment was added as in the control group.

Therapy was applied following strict hygienic measures by trained farm personnel. Cows with CM in more than one quarter were not included in the trial. During the whole trial, cows were kept under their usual housing conditions. Animals from both treatment groups were not separated during the trial and were kept under the same conditions on the farms.

Study Procedure

During the daily milking routine, CM was detected by the milking personnel, who classified its severity according to the guidelines of the IDF (IDF, 2011). The numbers of milkers varied from farm to farm between 3 and 10 people. The milking personnel on each farm are systematically trained in detecting mastitis and taking quarter milk samples. If a cow with a nonsevere CM case in one quarter was identified, farm staff checked for inclusion criteria using the herd management program. If a cow was included in the study, a foremilk sample of the affected udder quarter was taken in accordance with the guidelines of aseptic milk sampling for microbiological

Table 1. Pathogens isolated from quarter milk samples at the day of admission

Pathogen	Percentage (n) of isolated pathogens	AB group, n	KE group, n [AB treated]
<i>Streptococcus uberis</i>	23.0 (51)	30	21 [7]
NAS	9.9 (22)	16	6 [1]
<i>Escherichia coli</i>	9.5 (21)	11	10
<i>Streptococcus dysgalactiae</i>	7.2 (16)	10	6 [2]
Mixed	6.8 (15)	8	7
<i>Staphylococcus aureus</i>	4.1 (9)	3	6 [1]
<i>Pseudomonas</i> spp.	3.2 (7)	4	3
<i>Serratia</i> spp.	2.7 (6)	2	4
<i>Enterococcus</i> spp.	2.3 (5)	1	4
<i>Trueperella pyogenes</i>	1.8 (4)	1	3
Coliform bacteria	1.4 (3)	1	2
<i>Acinetobacter</i> spp.	0.9 (2)	2	0
<i>Corynebacterium</i> spp.	0.5 (1)	1	0
<i>Raoultella ornithinolytica</i>	0.5 (1)	1	0
Other streptococci	0.5 (1)	1	0
Contaminated	1.8 (4)	0	4
Total isolated pathogens	75.7 (168)	92	76 [11]
No specific growth	24.3 (54)	26	28 [3]
Grand total, % (n) [AB treated]	100.0 (222)	100.0 (118)	100.0 (104) [14]

analysis (GVA, 2009). This included cleaning the teat apex and subsequent disinfection with 70% ethanol. The contact time of the disinfectant was about 30 s. After discarding the first 3 streams of milk, the samples were taken. Following the randomization protocol, the animals received the appropriate treatment depending on the assigned treatment group. Each cow was included in the study with only one CM case according to the aforementioned treatment protocol. Farm personnel assessed the foremilk and mammary glands and documented the results of the examination. This was part of the daily routine on all test farms. In addition, treatment protocols were completed in accordance with the study procedure.

Clinical examination of the cows participating in the study was performed every 8 to 12 h by the study personnel during milking procedure for at least 6 d. Development of clinical signs was documented. Farm staff were required to report any abnormalities in the animals between milkings.

If there was no clinical improvement or if the clinical appearance worsened, the case was recorded as a treatment failure and the cows were additionally treated intramammarily with AB as in the control group. Clinical mastitis cases without clinical symptoms on d 5 (± 1) were assessed as clinically cured. These cured quarters were observed from d 6 to d 60 after the end of treatment for recurrent CM cases and a quarter foremilk sample of the respective udder quarter was taken in case of return of clinical signs. After treatment, quarter foremilk samples were collected from relevant quarters at d 14 (± 3) and 21 (± 3) after the end of treatment by a veterinarian of the study personnel. All samples were frozen and transported frozen to the laboratory.

Laboratory Procedure

All milk samples were aseptically collected and frozen until analysis (-20°C). Ly20, a boric acid-based preserving agent, was used in test tubes (GVA, 2009). Microbiological examinations were performed at the University of Applied Sciences and Arts Hannover (Hannover, Germany) in accordance with the guidelines of the German Veterinary Association (GVA, 2009).

For microbiological analysis, 10 μL of the milk sample were applied. Each milk sample was plated on esculin blood agar (Oxoid, Wesel, Germany) and incubated at 37°C for at least 48 h under aerobic conditions. The grown colonies were Gram stained and the activity of catalase, hemolysis patterns, and esculin hydrolysis were examined. Yeasts and Prototheca were differentiated exclusively by microscopy. Coryneforms showed no hemolysis, produced catalase, and appeared gram-positive and rod-shaped. *Bacillus* spp. were also catalase-positive, and they appeared gram-labile and formed endospores. All other microorganisms underwent a MALDI TOF analysis (Bruker Daltonics, microflex LT/SH smart, MBT Compass Library, V8). A milk sample was considered contaminated if more than 2 pathogens were detected.

Outcome Variables and Definitions

The primary outcome was CC5, and secondary outcomes were BC at d 14 ± 3 and 21 ± 3 , R60, and NI. The absence of clinical symptoms in milk (e.g., without flaky sediments, watery appearance, or discoloration) and on udder quarter (e.g., without swelling, heat, redness or pain) was defined as CC. The absence of the pathogen cultured

Table 2. Distribution of the independent variable categories

Independent variable	AB group n (%)	KE group n (%)	Total ¹
SCC history ($P = 0.42$)			
1 ($3 \times < 101,000$ cells/mL)	11 (13%)	7 (9%)	18
2 (not in 1 or 3)	68 (78%)	63 (77%)	131
3 ($3 \times > 700,000$ cells/mL)	8 (9%)	12 (15%)	20
Lactation number ($P = 0.887$)			
1	13 (11%)	10 (10%)	23
2	30 (25%)	29 (28%)	59
>2	75 (64%)	65 (63%)	140
DIM ($P = 0.33$)			
1 (<101)	58 (49%)	42 (40%)	100
2 (101–200)	28 (24%)	25 (24%)	53
3 (>201)	32 (27%)	37 (36%)	69
Pathogen group ($P = 0.100$)			
No growth or contaminated	26	33	59
Gram-negative	23	16	39
Minor	16	7	23
Mix	8	7	15
Other	2	7	9
<i>Staphylococcus aureus</i>	3	6	9
<i>Streptococcus</i> spp.	40	28	68

¹No data available for all animals – first lactation, lactational change. For some animals, data such as SCC history were not available. These were, for example, heifers that had not yet taken part in 3 milk controls.

pretreatment in both post-treatment samples at d 14 and 21 defined BC. A case was still defined as bacteriologically cured if a bacterial species other than the pathogen cultured pretreatment was isolated in the post-treatment samples. If one post-treatment sample was contaminated, the outcome of the other post-treatment sample was used to ascertain the BC. In cases where 2 pathogens were isolated in the pretreatment sample, the case was defined as a mixed infection. In these cases, a BC occurred if neither of the 2 pathogens were cultured in both post-treatment samples.

Quarters with clinically cured cases were monitored over a period from d 6 to d 60 after the end of treatment. They were defined as recurrent quarters when one or more CM cases were detected during this period (R60). If a quarter was free of CM within the observed time frame, it showed no R60. For each CM case, the amount of milk that could not be delivered was determined on the basis of the amount of milk before the clinical case, the time it took to CC, and the withdrawal time to be taken into account. The NI was defined as the presence of a microorganism in a quarter milk sample on d 14 and d 21 that was not present in the sample taken before treatment.

Statistical Analysis

The data were collected and analyzed using Excel, Office 2010 (Microsoft Corporation) and SPSS (IBM SPSS 28.0.0.0; IBM, Armonk, NY). The CM case of an udder quarter represented the statistical unit. For every CM case, CC5 or no CC5, R60 or no R60 and BC or no BC (encoded as 1 or 0, respectively) were determined

according to the aforementioned definitions, constituting the binary dichotomous-dependent variables. Univariable significant risk factors ($P < 0.2$) were then analyzed using generalized linear mixed models.

The general mixed model structure was as follows:

$$f(Y_{ijk}) = \text{intcpt} + (\beta_1 \times \text{treatment}) + (\beta_k \times \text{covariates}_k) + \text{herd}_j(\text{random}) + \text{error}_{ij},$$

where $f(Y)$ is the logit link function, Y_{ijk} is the outcome (yes/no) in quarter i , intcpt is the intercept, treatment is the variable indicating whether a quarter is in the AB or KE group, and β_1 refers to the regression coefficient for treatment. Other covariates (β_k) investigated stage of lactation (grouped: early, <101 DIM; mid, 101–200 DIM; late, >201 DIM), parity (grouped: 1, 2, >2), SCC history (DHI; grouped: 1, last 3 milk controls low, all <101,000; 2, medium, not in 1 or 3; 3, high, all >700,000 cells/mL), and pathogen (grouped: no growth; minor, including NAS; *Staphylococcus aureus*, streptococci, and gram-negative bacteria; mixed infection; other; or contaminated samples) cultured pretreatment as important covariates. At the herd level, the treatment groups were balanced with regard to the covariates ($P > 0.05$). Herd was selected as a random effect but had no relevant influence. The variable was retained as a design variable in the models. Backward stepwise procedures were used to select the final multivariable regression models. Potential risk factors were excluded if $P > 0.05$. Meaningful biological interactions between the fixed effects were also used in the final model if significant ($P < 0.05$) and if they

Table 3. Descriptive statistics for study outcomes by experimental group

Variable	Group	
	KE ¹	AB ²
CC5, % (n)	61.5 (64/104)	84.7 (100/118)
BC, % (n)	61.2 (41/67)	79.3 (73/92)
R60, % (n)	15.7 (14/89)	7.3 (8/109)
NI, % (n)	6.7 (7/104)	6.8 (8/118)

¹KE = test treatment group (ketoprofen group), the cows were treated with 3 applications of 3 mg ketoprofen per kilogram of BW (Romefen PR 10%/Ketofen 10%; Ceva Animal Health, Düsseldorf, Germany) at 24-h intervals.

²AB = reference treatment group (antibiotic group), cows were treated with 2 intramammary applications of 200 mg cefalexin and 100,000 IU, kanamycin (Ubrolexin, Boehringer Ingelheim, Ingelheim am Rhein, Germany) at 24-h intervals.

did not increase the Akaike information criterion (AIC). Nonsignificant effects or interactions that increased the AIC were not included in the final models. Model fit was evaluated by checking the normality of the residuals.

In all calculated models, only the treatment variable reached the respective final model. Statistical significance was assumed at $\alpha = 0.05$.

The model was used to calculate LSM of the treatment groups for CC5, BC, R60, and NI. The differences between treatments were estimated in this way. The non-inferiority assumption was checked with the CI of the treatment difference (primary outcome: CC5), calculating using the LSM and their SD (Schukken et al., 2013).

RESULTS

In total, 222 dairy cows with CM at the day of admission were included in the study on the 3 farms. A total of 222 quarters were microbiologically analyzed. Four samples were contaminated and therefore removed from calculations. Finally, 218 mastitis cases were assessed. In 148 cases, the severity was classified as mild (73 in the AB group, 71 in the KE group), whereas 74 cases were classified as moderate (45 in the AB group and 33 in the KE group). No animal had to be registered as a therapy failure because of developing severe mastitis during the course of the trial. In 2 cases in the AB group that started as mild mastitis, moderate mastitis was diagnosed on d 3.

In 75.2% (n = 164) of the quarters, pathogens were detected (Table 1). Mostly isolated pathogens were *Streptococcus uberis* with 23.4% (n = 51), NAS with 10.1% (n = 22), *Escherichia coli* with 9.6% (n = 21), and *Streptococcus dysgalactiae* with 7.3% (n = 16). The percentages of quarters with no detected pathogen were 24.8% (n = 54). The mastitis cases in the KE group that had to be treated with AB (Table 1, in brackets) had significantly more streptococcal and staphylococcal infections than the comparison groups (chi-squared test, $P = 0.03$).

The treatment groups did not differ significantly from each other in terms of lactation number ($P = 0.887$), stage of lactation ($P = 0.330$), SCC history of the animals ($P = 0.420$), and the distribution of pathogen groups ($P = 0.100$; Table 2). The milking frequency varied between the farms. However, this had no effect on milking routine or on mastitis detection. Any farm-specific effects were taken into account as random variables in the calculated models.

Treatment

A total of 118 CM cases belonged to the AB group, whereas 104 cases received KE. In accordance with the experimental rules, AB were used to retreat 14 animals belonging to the KE group due to lack of improvement of clinical signs on d 3.

Effect of Treatment on CC5

The overall crude CC5 was 84.7% and 61.5% for the AB and KE groups, respectively (Table 3). The predominant organisms recovered from affected quarters were *Strep. uberis*, NAS, and *E. coli* (Table 1). The final logistic regression model showed that the overall adjusted proportion of quarters experiencing a CC5 was different for the KE group (67.8%) compared with the AB group (referent [Ref.]; 91.2%; odds ratio [OR]: 4.934 [95% CI: 2.412–10.092]; $P < 0.001$; Table 4).

The CC5 of the KE-treated group was on average 23.9% (point estimate) worse than that of the AB-treated group (95% CI: –59.4% to 35.5%). The KE group was thus inferior to the AB group with regard to the target variable. When regarding the CC5 depending on the group of the causative pathogen, the univariable analysis showed that in the cases caused by streptococci CC was significantly higher when treated with AB (85%) compared with KE (37%, $P < 0.001$).

Bacteriological Cure

The determination of BC was conducted for 159 CM cases. The remaining cases were excluded from the study because no microorganisms were cultured (54 cases) or because the pretreatment or post-treatment samples were contaminated (9 cases). The overall BC rate was 71.7% (114/159). The probability of BC in the AB group was 79.3% (73/92) and in the KE group 61.2% (41/67), respectively (Table 3).

Results of the generalized linear mixed model showed the LSM of 78.6% for the AB group (Ref.) and 59.6% for the KE group (OR: 2.236; 95% CI: 1.103–4.532).

The model demonstrated that significant differences in BC were found between the reference treatment AB and the test treatment KE ($P = 0.026$; Table 4).

The treatment groups did not differ significantly from each other in terms of BC at pathogen-group level. The largest numerical difference between the treatment groups was found for streptococci (AB: 36/40 = 90.0% BC; KE: 21/28 = 75% BC). Excluding the follow-up treatments in the KE group on d 3, the BC here was only 14/28 = 52%.

R60

Only CM cases of cows that reached a CC and were still in milk 60 d after the end of treatment were included in this analysis (Pinzón-Sánchez and Ruegg, 2011). A total of 198 clinically cured quarters were included in the analysis. The overall R60 rate was 11.1% (22/198). The probability of achieving CM recurrence within 60 d after the end of treatment in the AB group was 7.3% (8/109) and in the KE group 15.7% (14/89), respectively (Table 3).

Results of the generalized linear mixed model showed different LSM of 5.8% for the AB (Ref.) group and 14.0% for the KE group (OR: 0.382; 95% CI: 0.149–0.982; $P = 0.046$; Table 4).

NI

The overall NI rate was 6.8% (15/222). It did not differ significantly between the treatment groups ($P = 0.985$; Table 3). Results of the generalized linear mixed model showed LSM of 7.2% for the AB group (Ref.) and 7.1% for the KE group (OR: 1.011; 95% CI: 0.345–2.959; $P = 0.985$; Table 4).

Effect of Treatment on AB Reduction and Milk Losses

The design of the study led to a planned reduction in AB consumption. Compared with the AB group, 86% fewer AB doses were used in the KE group ($P < 0.001$). Based on the treatment protocols, 91 kg (37.4%) less waste milk per mean case was determined for mastitis cases in the KE group compared with cases in the AB group until CC (AB: 243 kg; KE: 152 kg; $P < 0.001$; crude data). Less waste milk also means less effort in milking, less documentation, and therefore lower costs. The multivariable analysis using a linear mixed model showed that the milk loss in the AB group up to CC was 231 kg and in the KE group it was 144 kg. The difference was 87 kg.

DISCUSSION

This study addresses a critical challenge in dairy farming—reducing AB usage without compromising

the effectiveness of mastitis treatment. Although current studies showed that a targeted therapy model is considered as the best method, its implementation in practice is challenging. Therefore, we tested a simpler, more practical approach that starts with KE treatment and moves to AB only if clinical signs persist, addressing the growing threat of antimicrobial resistance. It aligns with global efforts to promote prudent AB use. The key question remains: Could KE alone suffice as a first-line treatment for certain cases, balancing efficacy with reduced AB dependency?

The primary outcome in this trial was CC5, because farmers and veterinarians usually assess the treatment success based on CC (Falkenberg et al., 2019). Clinical cure risk on d 5 differed significantly between the study groups. The CC5 rate in the KE group was 23.4% less than that in the AB group (LSM; mixed model). The univariate analysis showed that there was a significant difference for CC5 between the AB (85%) and KE (37%) groups in the cases caused by streptococci (and mixed infections) only ($P < 0.001$). This indicates that a targeted mastitis concept, aiming at the selective AB therapy of gram-positive cocci may be the better option for mastitis treatment in terms of CC.

The CC is very important in the decision-making process in mastitis therapy (Falkenberg et al., 2019). Many farmers measure the success of a treatment by the absence of clinical symptoms after the end of therapy. This is done for understandable reasons, because clinical symptoms are visible and easy to recognize. However, CC only correlates to a certain extent with BC, which is the real measure for assessing successful treatment (Krömker et al., 2020).

Other studies found a lower probability of CC of around 60% for CM cases treated with AB compared with our results (Schukken et al., 2013; Swinkels et al., 2014; Krömker et al., 2021). The studies by Swinkels et al. (2014) and Krömker et al. (2021) both examined animals with chronic udder diseases (based on persistently high SCC and cows with previous CM cases in current lactation). It is possible that CC risk worsens with increasing chronicity of mastitis.

Clinical cure in KE is particularly delayed for those cases that are re-treated on d 3 due to a lack of cure. From the farmer's point of view, it must now be weighed up whether the reduction in AB use and milk loss compensates for a worsening of the cure variables. In addition, it must be considered that the application of a targeted mastitis therapy (TMT) concept can generally reduce AB doses and discarded milk without reduced cure rates (CC, BC, and RC; Mansion-de Vries et al., 2016; Kock et al., 2018; Schmenger and Krömker, 2020; de Jong et al., 2023). However, as mentioned previously, in these studies this depends on the incidence of CM with

Table 4. Final generalized linear mixed model results for the outcome variables¹

Item	Coefficient	SEM	t-value	OR	95% CI for OR	P-value
CC5						
KE	1.596	0.3631	4.396	4.934	2.412–10.092	<0.001
AB	0*					
BC						
KE	0.805	0.3577	2.250	2.236	1.103–4.532	0.026
AB	0*					
R60						
KE	−0.961	0.4781	−2.011	0.382	0.149–0.982	0.046
AB	0*					
NI						
KE	0.010	0.5451	0.019	1.011	0.345–2.959	0.985
AB	0*					

¹CC5 = clinical cure at d 5; KE = test treatment group (ketoprofen group); AB = reference treatment group (antibiotic group); BC = bacteriological cure; R60 = CM recurrence in a period of 60 d; NI = new infection risk.

*Redundant coefficient set to zero.

gram-positive pathogens and the number of cases that occur in animals with a low probability of cure (cows unworthy of treatment).

Treatment based solely on KE was not as effective as AB use when evaluating BC, one of the secondary outcome variables.

The result must be viewed in consideration of the pathogen population in the test farms with a focus on *Strep. uberis* mastitis. Streptococcal mastitis benefits more from AB therapy than mastitis caused by other microorganisms (Guterbock et al., 1993; Morin et al., 1998; Roberson et al., 2004; Keller and Sundrum, 2018). If no BC can be achieved, recurrence must be expected (Grieger et al., 2014). Interestingly, only cases caused by gram-positive pathogens (*Strep. uberis*, NAS, *Strep. dysgalactiae*, *Staph. aureus*) from the KE group required additional AB treatment because they showed no improvement on d 3. A possible explanation for this could be that these pathogens are categorized as worthy of treatment with an AB but were initially only treated with an NSAID in the KE group.

The overall BC risk was high in this study with 71.7%, as was the BC risk for CM cases treated with AB (78.6%). Similar results for BC risks of ~70% were found in previous studies (Schukken et al., 2013; Swinkels et al., 2014; Schmenger and Krömker, 2020). In comparison, the results for BC in this study were significantly better than in other studies that only included cows with long-lasting udder diseases (Sol et al., 2000; Linder et al., 2013; Schukken et al., 2013; Krömker et al., 2019; Krömker et al., 2021).

Another secondary outcome variable in this study was R60. The probability of achieving CM recurrence within 60 d after the end of treatment was higher for animals of the KE group (14.0%) compared with animals of the AB group (5.8%). Different recurrence rates were described in previous trials dealing with cows suffering from longer-

lasting udder diseases (Ziesch et al., 2018; Krömker et al., 2021). Even though the recurrence rates were relatively low compared with similar studies, the large difference in the recurrence rate between the groups is an indication that the protocol tested should be dispensed with.

Clinical mastitis is a disease of recurrent nature (Schukken et al., 2010). Considering that high recurrence rates are described in various studies (Picker, 2012; Zoche-Golob and Spilke, 2013) and that recurrent CM cases have a negative effect on the lifespan of a dairy cows (Bar et al., 2008), the aim should be to minimize the recurrence rate on the farm. Recurrent mastitis includes persistent infections that are caused, for example, by unsuccessful therapies and new infections. Cases of CM requiring treatment, for example due to a gram-positive pathogen, should be treated with AB. Such cases in the KE group, which were treated accordingly only with NSAID, could represent cases with unsuccessful therapy and therefore cases with ongoing infections.

The AB-reduced treatment concept (KE) decreased the use of AB doses by 86% and the amount of waste milk by 35.9% per case compared with the reference treatment group (AB). Similar savings are also reported with the use of TMT (Lago et al., 2011; Vasquez et al., 2017; Schmenger and Krömker, 2020). However, the savings in AB doses in these studies on TMT depend on the incidence of CM with gram-positive infections and the number of cases that occur in animals with a low probability of cure (therapy-unworthy cows).

The use of AB in food-producing animals is generally declining (ECDC et al., 2024). Nevertheless, there is a need to reduce the use of AB further or use them more prudently to limit AB resistance development (Guo et al., 2021). In dairy farming, the AB treatment of mastitis accounts for more than 70% of the total use of AB (van Werven, 2018). However, a reduction in AB use should not be the decisive factor in treatment, which may be

at the expense of the animals in the form of lower cure rates. The primary goal should always be to achieve the highest possible BC rate. Studies investigating TMT concepts have shown that similar cure rates can be achieved using such evidence-based treatments while at the same time reducing the use of AB (Schmenger and Krömker, 2020; de Jong et al., 2023).

Waste milk, meaning milk from cows that have been treated with AB substances and must therefore be discarded during the prescribed withdrawal periods as well as milk with significant alterations, cannot be sold for human consumption. There is an ongoing discussion about what should be done with this waste milk (e.g., fed to calves, channeled into manure, and so on). In any case, wasted milk is not in line with a resource-saving milk production desired by consumers and also leads to monetary losses for the farmer. The longer the treatment periods, the higher the financial losses due to more milk from animals that are under withdrawal period (Pinzón-Sánchez and Ruegg, 2011). The active substance KE has no withdrawal time for milk. For this reason, there is no milk waste in this group due to withdrawal times. The reduction in discarded milk not only has a positive financial impact but also contributes to more sustainable cattle farming. Fewer cows will be needed in the long run to produce the same amount of milk for human consumption, leading to lower amounts of greenhouse gases. This makes the process more environmentally friendly.

The study has some limitations that need to be considered. First of all, it should be noted that due to the lack of a negative control group, which would have remained completely untreated, it is not possible to determine whether untreated cases would have led to similar results with regard to the reduction of AB consumption. Furthermore, although laboratory personnel and study evaluators were blinded to treatment groups, farm staff implementing treatments were not blinded due to the differences in administration methods (intramammary AB vs. systemic NSAID). However, all 3 farms were contract-labor farms, and the clinical evaluations were carried out in the milking parlor by the milking staff, while the treatments were administered by different farm personnel. This separation of tasks in this study reduces the risk of observer bias.

Furthermore, the study was conducted on 3 farms in northern Germany, which may limit the generalizability of the findings to other farming systems or regions with different management practices, climates, or herd demographics. However, northern Germany is a key region for the country's dairy production, making selected farms relevant for exploring practices representative of this sector. Similar constraints were observed in studies such as the study by Latosinski et al. (2020), which also

included a limited number of farms, suggesting that this is a common challenge in on-farm clinical trials.

One notable strength of this study compared with other studies investigating the efficacy of NSAID in the treatment of CM (e.g., McDougall et al., 2016; Latosinski et al., 2020) is the detailed reporting of case severity distribution (mild or moderate). This differentiation enhances the applicability of findings, indicating that our results are most relevant for mild and moderate cases of CM. This specificity allows for a more nuanced interpretation and application of the treatment protocols explored in the study.

CONCLUSIONS

Our findings indicated that systemic treatment with KE is inferior to AB treatment regarding CC rates, especially in cases caused by streptococci. Ketoprofen treatment alone also resulted in lower BC rates, along with higher rates of CM recurrence. The risk of new infections did not vary significantly between the groups. However, in more than 85% of cases treated with KE, subsequent intramammary AB treatment was not required, leading to significantly fewer AB doses being used and a reduced quantity of milk being discarded. It should be noted, however, that this reduction was a direct consequence of the study design, because intramammary AB were only administered to cases with aggravation or no recovery of clinical signs. Using KE as the sole initial treatment, when guided by pathogen identification, can serve as a TMT protocol aimed at the assertive and selective use of AB.

NOTES

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Nonstandard abbreviations used: AB = antibiotic; AIC = Akaike information criterion; BC = bacteriological cure; CC = clinical cure; CC5 = clinical cure at d 5;

CM = clinical mastitis; IDF = International Dairy Federation; KE = ketoprofen; NI = new infection risk; NSAID = nonsteroidal anti-inflammatory drug; OR = odds ratio; R60 = CM recurrence in a period of 60 d; Ref. = referent group; TMT = targeted mastitis therapy.

REFERENCES

- Bar, D., Y. T. Gröhn, G. Bennett, R. N. González, J. A. Hertl, H. F. Schulte, L. W. Tauer, F. L. Welcome, and Y. H. Schukken. 2008. Effects of repeated episodes of generic clinical mastitis on mortality and culling in dairy cows. *J. Dairy Sci.* 91:2196–2204. <https://doi.org/10.3168/jds.2007-0460>.
- Brogliato, A. R., C. A. Antunes, R. S. Carvalho, A. P. T. Monteiro, R. F. Tinoco, M. T. Bozza, C. Canetti, M. Peters-Golden, S. L. Kunkel, R. Vianna-Jorge, and C. F. Benjamim. 2012. Ketoprofen impairs immunosuppression induced by severe sepsis and reveals an important role for prostaglandin E2. *Shock* 38:620–629. <https://doi.org/10.1097/SHK.0b013e318272ff8a>.
- de Jong, E., L. Creyten, S. De Vliegher, K. D. McCubbin, M. Baptiste, A. A. Leung, D. Speksnijder, S. Dufour, J. R. Middleton, P. L. Ruegg, T. J. G. M. Lam, D. F. Kelton, S. McDougall, S. M. Godden, A. Lago, P. J. Rajala-Schultz, K. Orsel, V. Krömker, J. P. Kastelic, and H. W. Barkema. 2023. Selective treatment of nonsevere clinical mastitis does not adversely affect cure, somatic cell count, milk yield, recurrence, or culling: A systematic review and meta-analysis. *J. Dairy Sci.* 106:1267–1286. <https://doi.org/10.3168/jds.2022-22271>.
- ECDC (European Centre for Disease Prevention and Control), EFSA (European Food Safety Authority), and EMA (European Medicines Agency). 2024. Antimicrobial consumption and resistance in bacteria from humans and food-producing animals. *EFSA J.* 22:e8589. <https://doi.org/10.2903/j.efsa.2024.8589>.
- EMA (European Agency for the Evaluation of Medicinal Products). 2000. VICH Topic GL9 (GCP): Guideline on Good Clinical Practices. EMA, London, UK.
- European Union. 2019. Regulation (EU) 2019/6 of the European Parliament and of the Council of 11 December 2018 on veterinary medicinal products and repealing Directive 2001/82/EC. *Official Journal L4:43–167*.
- Falkenberg, U., V. Krömker, W. Heuwieser, and C. Fischer-Tenhagen. 2019. Survey on routines in udder health management and therapy of mastitis on German dairy farms. *Milchwissenschaft* 72:11–15. <https://doi.org/10.25968/MSI.2019.2>.
- Grieger, A. S., V. Zoche-Golob, J. H. Paduch, M. Hoedemaker, and V. Krömker. 2014. Rezidivierende klinische Mastitiden bei Milchkühen—Bedeutung und Ursachen. *Tierarztl. Prax. Ausg. G Grosstiere Nutztiere* 42:156–162. <https://doi.org/10.1055/s-0038-1623218>.
- Guo, K., Y. Zhao, L. Cui, Z. Cao, F. Zhang, X. Wang, J. Feng, and M. Dai. 2021. The influencing factors of bacterial resistance related to livestock farm: Sources and mechanisms. *Front. Anim. Sci.* 2:650347. <https://doi.org/10.3389/fanim.2021.650347>.
- Guterbock, W. M., A. L. Van Eenennaam, R. J. Anderson, I. A. Gardner, J. S. Cullor, and C. A. Holmberg. 1993. Efficacy of intramammary antibiotic therapy for treatment of clinical mastitis caused by environmental pathogens. *J. Dairy Sci.* 76:3437–3444. [https://doi.org/10.3168/jds.S0022-0302\(93\)77682-1](https://doi.org/10.3168/jds.S0022-0302(93)77682-1).
- GVA (German Veterinary Association). 2009. Guidelines for Aseptic Milk Sampling and Guidelines to Isolate and Identify Mastitis Pathogens. 2nd ed. German Veterinary Association, Gießen.
- IDF (International Dairy Federation). 2011. Suggested interpretation of mastitis terminology. No. 448/2011, Bulletin of the International Dairy Federation, Brussels, Belgium. Accessed Sep. 2, 2024. https://fil-idf.org/wp-content/uploads/woocommerce_uploads/2011/03/Bulletin-of-the-IDF-No.-448_2011-Suggested-Interpretation-of-Mastitis-Terminology-revision-of-Bulletin-of-IDF-N%C2%B0-338_1999-1-fdv1h1.pdf.
- Keller, D., and A. Sundrum. 2018. Comparative effectiveness of individualised homeopathy and antibiotics in the treatment of bovine clinical mastitis: Randomised controlled trial. *Vet. Rec.* 182:407. <https://doi.org/10.1136/vr.104555>.
- Kock, J., N. Wente, Y. Zhang, J. H. Paduch, S. Leimbach, D. Klocke, C. C. Gelfert, and V. Krömker. 2018. Udder health effects of an evidence-based mastitis therapy concept in Northwestern Germany. *Milchwissenschaft* 71:14–20. <https://doi.org/10.25968/MSI.2018.4>.
- Krömker, V., J. H. Paduch, D. Klocke, J. Friedrich, and C. Zinke. 2010. Efficacy of extended intramammary therapy to treat moderate and severe clinical mastitis in lactating dairy cows. *Berl. Munch. Tierarztl. Wochenschr.* 123:147–152.
- Krömker, V., A. Schmenger, D. Klocke, E. Mansion-de Vries, N. Wente, Y. Zhang, and S. Leimbach. 2021. Non-inferiority trial investigating the efficacy of non-steroidal anti-inflammatory drugs and antimicrobial treatment of mild to moderate clinical mastitis in dairy cows with long-lasting udder diseases. *Front. Vet. Sci.* 8:660804. <https://doi.org/10.3389/fvets.2021.660804>.
- Krömker, V., A. Schmenger, S. Leimbach, M. tho Seeth, and D. Klocke. 2020. Prognose der Entwicklung von Mastitiden auf der Basis der Befunde am Tag des Auftretens von Mastitiden. [Prediction of the development of mastitis based on the findings on the day of its onset.]. Page 5.3 in Proc. BPT Kongress. Hanover.
- Krömker, V., N. Wente, Y. Zhang, J. Bolte, R. Renner, A. Schmenger, I. Titze, J. Wallis, P. Mayer, and D. Klocke. 2019. Comparison of a non-antibiotic treatment with an antibiotic treatment of chronic mastitis. *Milchwissenschaft* 72:34–38. <https://doi.org/10.25968/MSI.2019.6>.
- Lago, A., S. M. Godden, R. Bey, P. L. Ruegg, and K. Leslie. 2011. The selective treatment of clinical mastitis based on on-farm culture results: I. Effects on antibiotic use, milk withholding time, and short-term clinical and bacteriological outcomes. *J. Dairy Sci.* 94:4441–4456. <https://doi.org/10.3168/jds.2010-4046>.
- Latosinski, G. S., M. J. Amzalak, and J. C. F. Pantoja. 2020. Efficacy of ketoprofen for treatment of spontaneous, culture-negative, mild cases of clinical mastitis: A randomized, controlled superiority trial. *J. Dairy Sci.* 103:2624–2635. <https://doi.org/10.3168/jds.2019-17504>.
- Linder, M., J. H. Paduch, S. A. Grieger, E. M. Mansion-de Vries, N. Knorr, C. Zinke, K. Teich, and V. Krömker. 2013. Cure rates of chronic subclinical *Staphylococcus aureus* mastitis in lactating dairy cows after antibiotic therapy. *Berl. Munch. Tierarztl. Wochenschr.* 126:291–296.
- Mansion-de Vries, E. M., M. Hoedemaker, and V. Krömker. 2015. Aspekte einer Evidenzbasierten Therapie Klinischer Mastitiden. *Tierarztl. Prax. Ausg. G Grosstiere Nutztiere* 43:287–295. <https://doi.org/10.15653/TPG-150227>.
- Mansion-de Vries, E. M., J. Lücking, N. Wente, C. Zinke, M. Hoedemaker, and V. Krömker. 2016. Comparison of an evidence-based and a conventional mastitis therapy concept with regard to cure rates and antibiotic usage. *Milchwissenschaft* 69:27–32. <https://doi.org/10.25968/MSI.2016.6>.
- McDougall, S., E. Abbeloos, S. Piepers, A. S. Rao, S. Astiz, T. Van Werven, J. Statham, and N. Pérez-Villalobos. 2016. Addition of meloxicam to the treatment of clinical mastitis improves subsequent reproductive performance. *J. Dairy Sci.* 99:2026–2042. <https://doi.org/10.3168/jds.2015-9615>.
- McDougall, S., M. A. Bryan, and R. M. Tiddy. 2009. Effect of treatment with the nonsteroidal anti-inflammatory meloxicam on milk production, somatic cell count, probability of re-treatment, and culling of dairy cows with mild clinical mastitis. *J. Dairy Sci.* 92:4421–4431. <https://doi.org/10.3168/jds.2009-2284>.
- Morin, D. E., R. D. Shanks, and G. C. McCoy. 1998. Comparison of antibiotic administration in conjunction with supportive measures versus supportive measures alone for treatment of dairy cows with clinical mastitis. *J. Am. Vet. Med. Assoc.* 213:676–684. <https://doi.org/10.2460/javma.1998.213.05.676>.
- O'Connor, A. M., J. M. Sargeant, I. A. Gardner, J. S. Dickson, M. E. Torrence, C. E. Dewey, R. B. Evans, J. T. Gray, M. Greiner, G. Keefe, S. L. Lefebvre, P. S. Morley, A. Ramirez, W. Sischo, D. R. Smith, K. Snedeker, J. Sofos, M. P. Ward, and R. Wills. 2010. The REFLECT statement: Methods and processes of creating reporting guidelines for randomized controlled trials for livestock and food safety. *J. Vet.*

- Intern. Med. 24:57–64. <https://doi.org/10.1111/j.1939-1676.2009.0441.x>.
- Picker, J. C. 2012. Aspects of recurrent mastitis. MS thesis. Department of Agriculture, University Göttingen, Göttingen, Germany.
- Pinzón-Sánchez, C., and P. L. Ruegg. 2011. Risk factors associated with short-term post-treatment outcomes of clinical mastitis. *J. Dairy Sci.* 94:3397–3410. <https://doi.org/10.3168/jds.2010-3925>.
- Preine, F., D. Herrera, C. Scherpenzeel, P. Kalmus, F. McCoy, S. Smulski, P. Rajala-Schultz, A. Schmenger, P. Moroni, and V. Krömker. 2022. Different European perspectives on the treatment of clinical mastitis in lactation. *Antibiotics (Basel)* 11:1107. <https://doi.org/10.3390/antibiotics11081107>.
- Roberson, J. R. 2003. Establishing treatment protocols for clinical mastitis. *Vet. Clin. North Am. Food Anim. Pract.* 19:223–234. [https://doi.org/10.1016/S0749-0720\(02\)00071-3](https://doi.org/10.1016/S0749-0720(02)00071-3).
- Roberson, J. R., L. D. Warnick, and G. Moore. 2004. Mild to moderate clinical mastitis: Efficacy of intramammary amoxicillin, frequent milk-out, a combined intramammary amoxicillin, and frequent milk-out treatment versus no treatment. *J. Dairy Sci.* 87:583–592. [https://doi.org/10.3168/jds.S0022-0302\(04\)73200-2](https://doi.org/10.3168/jds.S0022-0302(04)73200-2).
- Schmenger, A., and V. Krömker. 2020. Characterization, cure rates and associated risks of clinical mastitis in northern Germany. *Vet. Sci.* 7:170. <https://doi.org/10.3390/vetsci7040170>.
- Schmenger, A., S. Leimbach, N. Wente, Y. Zhang, A. M. Biggs, and V. Krömker. 2020. Implementation of a targeted mastitis therapy concept using an on-farm rapid test: Antimicrobial consumption, cure rates and compliance. *Vet. Rec.* 187:401.
- Schukken, Y. H., D. Bar, J. Hertl, and Y. T. Gröhn. 2010. Correlated time to event data: Modeling repeated clinical mastitis data from dairy cattle in New York State. *Prev. Vet. Med.* 97:150–156. <https://doi.org/10.1016/j.prevetmed.2010.09.012>.
- Schukken, Y. H., M. J. Zurakowski, B. J. Rauch, B. Gross, L. L. Tikofsky, and F. L. Welcome. 2013. Noninferiority trial comparing a first-generation cephalosporin with a third-generation cephalosporin in the treatment of nonsevere clinical mastitis in dairy cows. *J. Dairy Sci.* 96:6763–6774. <https://doi.org/10.3168/jds.2013-6713>.
- Shpigel, N. Y., R. Chen, M. Winkler, A. Saran, G. Ziv, and F. Longo. 1994. Anti-inflammatory ketoprofen in the treatment of field cases of bovine mastitis. *Res. Vet. Sci.* 56:62–68. [https://doi.org/10.1016/0034-5288\(94\)90197-X](https://doi.org/10.1016/0034-5288(94)90197-X).
- Sol, J., O. C. Sampimon, H. W. Barkema, and Y. H. Schukken. 2000. Factors associated with cure after therapy of clinical mastitis caused by *Staphylococcus aureus*. *J. Dairy Sci.* 83:278–284. [https://doi.org/10.3168/jds.S0022-0302\(00\)74875-2](https://doi.org/10.3168/jds.S0022-0302(00)74875-2).
- Suojala, L., H. Simojoki, K. Mustonen, L. Kaartinen, and S. Pyörälä. 2010. Efficacy of enrofloxacin in the treatment of naturally occurring acute clinical *Escherichia coli* mastitis. *J. Dairy Sci.* 93:1960–1969. <https://doi.org/10.3168/jds.2009-2462>.
- Swinkels, J. M., V. Krömker, and T. J. G. M. Lam. 2014. Efficacy of standard vs. extended intramammary cefquinome treatment of clinical mastitis in cows with persistent high somatic cell counts. *J. Dairy Res.* 81:424–433. <https://doi.org/10.1017/S0022029914000442>.
- van Werven, T. 2018. Managing dairy cows with less antibiotics! *WCDS Adv. Dairy Technol.* 30:37–47.
- Vasquez, A. K., D. V. Nydam, M. B. Capel, S. Eicker, and P. D. Virkler. 2017. Clinical outcome comparison of immediate blanket treatment versus a delayed pathogen-based treatment protocol for clinical mastitis in a New York dairy herd. *J. Dairy Sci.* 100:2992–3003. <https://doi.org/10.3168/jds.2016-11614>.
- Ziesch, M., N. Wente, Y. Zhang, W. Zaremba, S. Engl, and V. Krömker. 2018. Noninferiority trial investigating the efficacy of a nonantibiotic intramammary therapy in the treatment of mild-to-moderate clinical mastitis in dairy cows with longer lasting udder diseases. *J. Vet. Pharmacol. Ther.* 41:11–21. <https://doi.org/10.1111/jvp.12415>.
- Zoche-Golob, V., and J. Spilke. 2013. Herd-specific estimation of milk yield reduction due to recurrent clinical mastitis. *Berl. Munch. Tierarztl. Wochenschr.* 126:269–276.

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