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Evaluating the effectiveness of commercial teat disinfectant products sold in Ireland using the disc diffusion method

S.R. Fitzpatrick[1,2], M. Garvey[2], J. Flynn[1], B. O'Brien[1], D. Gleeson[1†]

[1]Teagasc, Animal & Grassland Research and Innovation Centre, Moorepark, Fermoy, County Cork, Ireland
[2]Department of Life Science, Institute of Technology Sligo, County Sligo, Ireland

Abstract

Evaluation of teat disinfectant products for their effectiveness against the most prevalent mastitis-causing bacteria is important to identify the most effective ingredients against specific bacterial strains. Ninety-six commercially available teat disinfectant products were tested against three bacterial strains associated with mastitis in Ireland (Staphylococcus aureus, Streptococcus uberis and Escherichia coli) using the disc diffusion method. Products were reclassified by active ingredients (n = 9) for analysis. These ingredient groups included: chlorhexidine (n = 25), chlorine dioxide (n = 5), diamine (n = 1), iodine (n = 13), iodine combined with lactic acid (n = 5), lactic acid (n = 15), lactic acid combined with chlorhexidine (n = 21), lactic acid combined with hydrogen peroxide (n = 1) and lactic acid combined with salicylic acid (n = 10). The ingredient group chlorine dioxide resulted in the greatest zones of inhibition for all three bacterial strains. An individual product containing a combination of lactic acid and hydrogen peroxide resulted in the greatest zone of inhibition for Sta. aureus and Str. uberis, whereas a specific product within the chlorine dioxide group resulted in the greatest zones of inhibition for E. coli. High concentrations of active ingredient did not necessarily increase the effectiveness for the majority of teat disinfectant products. It is possible to use the disc diffusion method to evaluate/screen a large number of teat disinfectant products prior to conducting field trials to establish the products' ability to reduce intramammary infections (IMI).

Keywords

Disc diffusion • mastitis • screening • teat disinfectant products

Introduction

Bovine mastitis or intramammary infection (IMI) is an inflammatory reaction which is caused by different microbial pathogens which can gain entry into the mammary gland through the teat canal (Berry & Meaney, 2006; Keane *et al*., 2013). However, despite decades of advances regarding the prevention and treatment of mastitis, it continues to be one of the main causes of impaired milk quality, decreased production, reduced profit and animal morbidity and mortality (Ruegg, 2012). There is a large variation in the pathogens identified within countries, which are associated with mastitis, and these differences may be due to different veterinary and laboratory services and farmer management practises (Zadoks & Fitzpatrick, 2009). In most countries, the most common bacteria associated with mastitis are *Str. agalactiae*, *Str. dysgalactiae, Str. uberis*, *Str. aureus* and *Escherichia coli* (Zadoks & Fitzpatrick, 2009). A study by Keane *et al*. (2013) identified *Sta*. *aureus*, *Str*. *uberis* and *E*. *coli* as the main bacteria associated with clinical mastitis on Irish dairy farms. This study also found that *Str*. *uberis* and *E*. *coli* were more commonly associated with clinical mastitis than *Sta*. *aureus*. Implementation of an effective mastitis control plan can help to prevent and reduce incidence of mastitis and reduce horizontal transmission of bacteria from cow to cow and within the environment. These control measures include hygienic milking and housing conditions, routine milking machine maintenance, teat disinfection pre- and post-milking, dry cow therapy, isolation of infected animals and cow culling (Hillerton & Booth, 2018). Studies have reported that the use of pre-milking teat cleaning regimes, using teat disinfectants, can reduce the bacterial load on the teat skin surface (Gibson *et al*., 2008; Mišeikienė *et al*., 2015; Baumberger *et al*., 2016). Previous studies also showed that pre-milking teat disinfection helped to reduce mastitis caused by environmental bacteria (Pankey, 1989; Oliver *et al*., 1993a,b, 2001) and mastitis caused by *Streptococcus* spp. and Gram-negative bacteria (Oliver *et al*., 1993b). These studies were undertaken when

†Corresponding author: David Gleeson
E-mail: David.Gleeson@teagasc.ie

cows were indoors; however, where cows were grazed on pastures, little to no improvement on mastitis incidence levels was observed (Williamson & Lacy-Hulbert, 2013; Gleeson *et al*., 2018). Post-milking teat disinfection is important for the control of contagious mastitis in herds (Breen, 2019). Contagious bacteria tend to be spread from cow to cow during milking via the milking machine and by the milker's hands. A study by Williamson and Lacy-Hulbert (2013) demonstrated that cows receiving post-milking disinfection had a lower rate of IMIs, caused by *Sta*. *aureus*, *Str*. *uberis*, *Corynebacterium* spp. and coagulase-negative staphylococci, than cows that did not receive post-milking disinfection. The success of teat disinfection in reducing new IMIs may also be influenced by the product active ingredient.

At present, the main knowledge regarding teat disinfectants relates to iodine as it is a broad-spectrum disinfectant and has been proven to be effective against mastitis and new IMIs (Oliver *et al*., 1991; Boddie *et al*., 2004; Böhm *et al*., 2017). However, alternative ingredients to iodine are now desirable due to concerns regarding iodine residues in milk which may be destined for infant milk formula manufacturing. Unfortunately, little knowledge is known regarding the effectiveness of these ingredients and products within an Irish context. There are many test methods available to measure the effectiveness of teat disinfection products. For regulatory purposes, teat disinfection products are required to be evaluated by the BS EN 1656 laboratory test method, known as a European Standards test. The National Mastitis Council (NMC) recommends the use of the experimental challenge and natural exposure protocols as they are useful for demonstrating field efficacy in reducing new IMI and mastitis. However, these tests may be limited to showing efficacy against specific organisms present on individual farms (Lopez-Benavides *et al*., 2012). The disc diffusion laboratory method can be used to screen disinfection products against a broad range of mastitis pathogens (Garvey *et al*., 2017; Fitzpatrick *et al*., 2019a). Screening/testing products using such a method can identify effective products before time-consuming and expensive field tests are undertaken. The objective of this study was to independently screen the effectiveness of 96 commercially available teat disinfection products, against the three main bacteria associated with mastitis in Ireland, *Sta*. *aureus*, *Str*. *uberis* and *E*. *coli*, using the disc diffusion method.

Materials and methods

Teat disinfectant information

Ninety-six commercially available teat disinfectant products (Table 1), with different active ingredients of varying concentrations, were tested against mastitis-causing bacteria, using the disc diffusion method. The teat disinfectant products were either ready-to-use (RTU) (*n* = 82), concentrate (conc.) products (*n* = 9) or required activation before use (*n* = 5). Concentrate products were diluted using sterile distilled water according to the manufacturer's recommendation to avoid possible issues with water hardness or contaminated water. Five chlorine dioxide-based products (products 11, 70, 89, 90 and 95) were mixed with an activator before use according to the manufacturer's recommendations. The disinfectant products used were recommended either for both pre-/post-milking teat disinfection (*n* = 49), pre-milking teat disinfection only (*n* = 3) or post-milking disinfection only (*n* = 44). Concentrations of active ingredients are declared where indicated by the manufacturer on the product label.

Bacterial strain identification

The bacteria applied in this study were isolated from the teat skin of cows within the Teagasc Moorepark research herd, by taking skin swab samples from lactating cows' teats using moistened cotton swabs, according to NMC (NMC, 2017) guidelines. The isolates were gram stained and bacterial identification was carried out using biochemical tests including lactose fermentation, motility test medium (Sigma-Aldrich, Dublin, Ireland), catalase and oxidase tests, tube coagulase (Sigma-Aldrich, Dublin, Ireland) and growth/reaction on various types of agars including blood agar (Sigma-Aldrich, Dublin, Ireland), MacConkey (Merck Millipore, Cork, Ireland), Baird Parker (Merck KGaA64271, Darmstadt, Germany) and modified Edwards agar (Oxoid 3M0027, Hampshire, UK), Simmons citrate agar (Sigma-Aldrich, Dublin, Ireland), CAMP esculin agar (Sigma-Aldrich, Dublin, Ireland) and triple sugar iron agar (Sigma-Aldrich, Dublin, Ireland). Analytical Profile Index-Staph (API-Staph Kit, bioMerieux, Marcy-l'Etoile, France) and API 20 tests (API, bioMerieux, Marcy-l'Etoile, France) were also used according to the manufacturer's instructions.

Disc diffusion method

The disc diffusion method was used to determine the ability of the teat disinfectant products to inhibit bacterial growth. This method was chosen based on a previous study that demonstrated that the method can effectively screen/evaluate a number of teat disinfectant products in a short period of time (Fitzpatrick *et al*., 2019a).

The disc diffusion method was performed using the techniques as described by Fitzpatrick *et al.* (2019a). Each bacterial strain isolated was grown on Mueller Hinton (MH) agar (Sigma-Aldrich, Ireland) plates with blank filter paper discs (three per plate; Cruinn, Dublin, Ireland) impregnated with a different teat disinfectant product. The experiment was independently repeated over 3 d, with three plate replications for each product against each bacterial strain tested. A pilot study was performed to determine the inclusion of sterile skimmed milk or sterile bovine serum albumin (BSA) as an interfering

Table 1: Test teat disinfectant product code and active ingredient, as declared by the manufacturer on product label

Product	#	Ingredient (w/w)	Pre or post
Arkshield[1]	7	5% Lactic acid/0.3% chlorhexidine	Pre/post
Arrabawn Udder Guard[1]	40	0.5% Chlorhexidine	Pre/post
Bacto-Lac[1]	31	5% Lactic acid/0.05% chlorhexidine	Pre/post
Barri-max[1]	65	2.4% Lactic acid	Post
Biolac Pre-Post[1]	44	0.25% Lactic acid/0.03% salicylic acid	Pre/post
Biolac Pre-Post[1]	59	0.25% Lactic acid/0.03% salicylic acid	Pre/post
Bisept[2]	70	0.05% Chlorine dioxide	Pre/post
Blue Barrier Spray[1]	49	Lactic acid/0.6% chlorhexidine[3]	Post
Blu-gard N Spray[1]	15	3.46% Lactic acid	Post
C-Dip[1]	61	0.53% Chlorhexidine	Post
Co-op Source Duo-Teat Shield[1]	39	2% Lactic acid/0.3% chlorhexidine	Pre/post
D 4 Iodine[4]	19	0.5% Iodine	Post
Dairy Pro UltraDip[1]	74	3% Lactic acid	Post
DairyLac SA[1]	76	3% Lactic acid	Post
Deosan Mastocide[1]	32	0.5% Chlorhexidine	Post
Deosan Summer Teat Care[1]	33	0.425% Chlorhexidine	Post
Deosan Super Iodip[4]	34	0.5% Iodine	Post
Deosan Teat Foam Advance[1]	13	0.6% Chlorhexidine	Pre/post
Deosan Teatcare Plus[1]	14	0.425% Chlorhexidine	Post
Deosan Triathalon[1]	81	1.76% Lactic acid	Pre/post
Dermalac Emprasan[1]	27	0.25% Lactic acid/salicylic acid[3]	Pre/post
Dual Dip Supreme[1]	47	Lactic acid/0.6% chlorhexidine[3]	Pre/post
Dual Dip[1]	45	2% Lactic acid/0.3% chlorhexidine	Pre/post
Duo-cel[1]	38	2.5% Lactic acid/0.3% chlorhexidine	Pre/post
Duogold[1]	17	2% Lactic acid/0.3% chlorhexidine	Pre/post
Duo-Teat Shield[1]	25	2% Lactic acid/0.3% chlorhexidine	Pre/post
Emprasan dual[1]	53	0.25% Lactic acid/salicylic acid[3]	Pre/post
Flexigard Spray[1]	94	4% Lactic acid	Post
Fortress Protect Film[1]	73	3% Lactic acid/0.2% chlorhexidine	Post
Gold Glycodip XL[1]	62	0.5% Iodine/1% lactic acid	Post
Hamra Red[1]	12	0.42% Chlorhexidine	Post
Hexa-cel RTU[1]	42	0.52% Chlorhexidine	Pre/post
Hexaguard[1]	1	0.74% Chlorhexidine	Pre/post
Hexaklene R[1]	2	0.5% Chlorhexidine	Pre/post
Hexa-Spray[1]	82	0.5% Chlorhexidine	Pre/post
Hypraspray[1]	87	2% Lactic acid/0.03% chlorhexidine	Pre/post
Hypred Quick Spray[1]	20	2% Lactic acid/0.1% salicylic acid	Pre/post
Ioguard[4]	3	0.5% Iodine	Post
Ioklar Multi[1]	92	0.25% Iodine	Pre/post
Io-Shield D[1]	91	1.35% Iodine	Post
Io-Shield Spray[1]	93	0.5% Iodine	Post
Kenocidin Spray and Dip[1]	9	0.5% Chlorhexidine	Post

Table 1: Continued

Product	#	Ingredient (w/w)	Pre or post
Kenolac SD[1]	80	3.6% Lactic acid	Post
Kenolac[1]	10	3.6% Lactic acid	Post
Kenomint SD[1]	78	0.5% Chlorhexidine	Post
Kenomint[1]	77	0.5% Chlorhexidine	Post
Kenomix SD[2]	89	0.0157% Chlorine dioxide	Post
Kenomix[2]	11	0.0157% Chlorine dioxide	Post
Kenopure[1]	79	3.2% Lactic acid	Pre
Lactic Lather[1]	46	1.6% Lactic acid/hydrogen peroxide[3]	Pre
Lacto dual[1]	36	2.5% Lactic acid/1.5% chlorhexidine	Pre/post
Lacto-cel[1]	35	2.4% Lactic acid	Pre/post
Lacto-Mil[1]	96	5% Lactic acid	Pre/post
Lactospray[1]	4	2.4% Lactic acid	Pre/post
Lanodip 4 XL[1]	30	0.5% Iodine/0.5% lactic acid	Post
Lanodip Pre-Post[1]	55	0.29% Iodine/0.8% lactic acid	Pre/post
Lely Quaress-Cura[1]	43	3% Lactic acid/salicylic acid[3]	Post
Luxdip 50B[1]	69	0.5% Iodine	Post
Masocare Platinum[1]	85	0.54% Iodine	Pre/post
Masodine Concentrate[4]	83	0.5% Iodine	Pre/post
Masodip Platinum[1]	84	0.436% Chlorhexidine	Pre/post
Maxadine C[4]	8	0.5% Iodine	Post
Maxidine RTU[1]	37	0.5% Iodine	Post
Nano Dual[1]	28	1.93% Lactic acid/0.2% chlorhexidine	Pre/post
Novo Dual[1]	29	4% Lactic acid/0.27% chlorhexidine	Pre/post
Novodip[1]	60	4.9% Lactic acid/1.28% chlorhexidine	Post
Novospray[1]	54	4.9% Lactic acid/0.3% chlorhexidine	Post
Prefoam+[1]	21	2% Lactic acid/0.1% salicylic acid	Pre
Protect Pre Post[1]	75	3% Lactic acid/0.25% chlorhexidine	Pre/post
PureChem Chlorhexidine Summer Grade[1]	52	1.49% Chlorhexidine	Pre/post
PureChem Chlorhexidine[1]	48	1.49% Chlorhexidine	Post
PureChem Dual Dip[1]	51	1% Lactic acid/1.49% chlorhexidine	Pre/post
PureChem Iodophor[4]	50	0.5% Iodine	Post
Quatro[1]	41	0.5% Chlorhexidine	Pre/post
SalvoDip B[1]	71	2.4% Lactic acid	Post
Salvohex[1]	67	2% Lactic acid/0.3% chlorhexidine	Post
Salvospray[1]	68	2.4% Lactic acid	Pre/post
SensoDip 50[1]	16	0.5% Chlorhexidine	Post
SensoDip[1]	72	0.5% Chlorhexidine	Pre/post
Sensospray[1]	66	0.5% Chlorhexidine	Post
Silkdip[4]	24	0.5% Iodine	Post
Summer C-Dip[1]	58	0.5% Chlorhexidine	Post
Super Cow Teat Foam[1]	26	0.6% Diamine	Pre/post

Table 1: Continued

Product	#	Ingredient (w/w)	Pre or post
Supergold[1]	18	0.5% Chlorhexidine	Pre/post
Supreme[1]	64	2.5% Lactic acid/0.37% chlorhexidine	Pre/post
Sure Spray Duo[1]	6	2% Lactic acid/0.3% chlorhexidine	Pre/post
Surespray[1]	5	0.5% Chlorhexidine	Pre/post
Synofilm[1]	88	8% Lactic acid	Post
Teat Gard C[1]	63	0.5% Chlorhexidine	Pre/post
TriCide Gold[1]	57	0.15% Iodine/1% lactic acid	Pre/post
TriCide[1]	56	0.15% Iodine/1% lactic acid	Post
Uddergold[2]	90	0.32% Acidified sodium chlorite	Post
Valiant[2]	95	0.038% Sodium chloride	Post
Virolac Film[1]	23	2% Lactic acid/0.1% salicylic acid	Post
Virolac Spray[1]	86	2% Lactic acid/0.1% salicylic acid	Pre/post
Virolac Concentrate[4]	22	2% Lactic acid/0.1% salicylic acid	Pre/post

[1]Ready to use (RTU).
[2]Requires activation before use.
[3]The concentration of some active ingredients for combination products was not declared by the manufacturer.
[4]Concentrate.
= product number.

substance to imitate the environment that the teat disinfectants would be used in. In this pilot study, the disc diffusion method was modified to allow for the addition of the aforementioned interfering substances. This modification involved adding the adjusted bacterial suspension to the interfering substance/organic matter suspension. The method was then carried out as described above. The zone of no growth (zone of inhibition) (measured in millimetres [mm], using a digital calliper [RS digital calliper 600/880, Mitutoyo Digimatic, Hampshire, UK]) around the disc is a measure of the ability of the teat disinfectant product to inhibit the growth of the test bacterial strain.

Statistical analysis

Statistical analysis was carried out using SAS for Windows, version 9.4 (SAS Institute Inc., Cary, NC, USA; SAS, 2014). The results were analysed using the PROC GLIMMIX procedure. Pair-wise comparisons were adjusted for multiplicity effect using simulation procedures to adjust *P*-values. Residual checks were made to ensure assumptions of analysis were met. This was used to determine the difference in susceptibility or resistance of bacterial species and differences in zones of inhibition between teat disinfectant products within ingredient groups. Products used within the study were reclassified by active ingredients (n = 9) to minimise/control the occurrence of type II errors during analysis. Comparisons between ingredient groups and between products within each ingredient group were compared using LSMEANS in the PROC GLIMMIX procedure. These ingredient groups included: chlorhexidine (n = 25), chlorine dioxide (n = 5), diamine (n = 1), iodine (n = 13), iodine combined with lactic acid (n = 5), lactic acid (n = 15), lactic acid combined with chlorhexidine (n = 21), lactic acid combined with hydrogen peroxide (n = 1) and lactic acid combined with salicylic acid (n = 10).

Results

The bacteria isolated from teat skin swab samples and identified in this study were found to be *Sta. aureus*, *Str. uberis* and *E. coli*. Furthermore, the pilot study showed that the use of either of the two different interfering substances did not have a negative impact on the effectiveness of the teat disinfectants used. Therefore, no interfering substance was included in the current study.

The average zone of inhibition for each product group can be seen in Figure 1. Lactic acid combined with hydrogen peroxide achieved the largest zones of inhibition for all three bacterial strains (*Str. uberis* [27.9 mm], *Sta. aureus* [25.1 mm] and *E. coli* [19.3 mm]). This was followed by the ingredient group chlorine dioxide (*Str. uberis* [21.3 mm], *Sta. aureus* [20.0 mm] and *E. coli* [18.1 mm]). Chlorhexidine group and diamine resulted in the smallest bacterial inhibitions for *Str. uberis* (17.9 mm and 16.1 mm, respectively), with a combination of lactic acid and salicylic acid achieving

Figure 1. Least squares means of the zones of inhibition (mm) of (A) *Str. uberis*, (B) *Sta. aureus* and (C) *E. coli* across all teat disinfectant active ingredient groups tested by the disc diffusion method. Error bars indicate SEM. [a,b,c,d,e,f]Inhibitions not sharing the same superscript for each bacterial strain were significantly different ($P < 0.05$).

significantly smaller zones of inhibition against *Sta. aureus* (13.2 mm) compared to chlorine dioxide ($P < 0.05$). For *E. coli*, the ingredient iodine and lactic acid resulted in a smaller level of bacterial inhibition (10.9 mm) compared to a combination of lactic acid and hydrogen peroxide and chlorine dioxide ($P < 0.05$).

Within the study, two ingredient groups (diamine and lactic acid combined with hydrogen peroxide) contained only one

product each. The ingredient group, diamine, demonstrated a smaller level of bacterial inhibition, with an overall average of 16.1 mm, 14.5 mm and 13.6 mm for *Str. uberis*, *Sta. aureus* and *E. coli*, respectively. The ingredient group which included a combination of lactic acid and hydrogen peroxide achieved a large bacterial inhibition against *Str. uberis* (27.9 mm), *Sta. aureus* (25.1 mm) and *E. coli* (19.3 mm). Bacterial inhibition of teat disinfectant products against different bacterial strains within each ingredient group can be observed in Table 2.

Chlorhexidine

Twenty-five products belonged to the chlorhexidine group. These products ranged in chlorhexidine concentrations from 0.42% to 1.49% w/w chlorhexidine. For *Str. uberis*, product 1 (0.74% w/w chlorhexidine) resulted in the largest zone of inhibition of 21.4 mm, which was significantly larger than product 48 (1.49% w/w chlorhexidine), resulting in an inhibition of 15.6 mm ($P < 0.05$). For *Sta. aureus*, product 5 (0.5% w/w chlorhexidine) resulted in the largest inhibition of 17.1 mm, which differed significantly from product 82 (0.5% w/w chlorhexidine; 14.0 mm) ($P < 0.05$). For *E. coli*, product 2 (0.5% w/w chlorhexidine) resulted in the largest inhibition of 20.6 mm. This differed significantly from product 82, which resulted in a smaller inhibition of 12.3 mm ($P < 0.05$). Within this ingredient group, a trend was observed for the effectiveness of teat disinfectant products against the three bacterial strains. Product 1 observed the largest inhibition for *Str. uberis* (21.4 mm) and resulted in the second and third largest inhibitions for *Sta. aureus* (17.7 mm) and *E. coli* (18.3 mm), respectively.

Chlorine dioxide

The chlorine dioxide ingredient group consisted of five different teat disinfectant products. These products ranged in chlorine dioxide concentrations from 0.0157% to 0.038% w/w. For *Str. uberis*, *Sta. aureus* and *E. coli*, product 95 (0.038% w/w chlorine dioxide) resulted in the largest zones of inhibition of 22.8 mm, 22.4 mm and 21.5 mm, respectively. Furthermore, product 70 (0.05% w/w chlorine dioxide) resulted in a smaller inhibition of 19.4 mm, 18.2 mm and 12.2 mm for *Str. uberis*, *Sta. aureus* and *E. coli*, respectively, compared to product 95 ($P < 0.05$).

Diamine

Only one teat disinfectant product contained the ingredient diamine (0.6% w/w diamine; product 26). This product resulted in zones of inhibition of 16.1 mm, 14.5 mm and 13.6 mm for *Str. uberis*, *Sta. aureus* and *E. coli*, respectively.

Iodine

There were 13 iodine products tested within this study. These products ranged from a concentration of 0.25% w/w to 1.35% w/w iodine. For *Str. uberis*, product 8 (0.5% w/w iodine) resulted in the largest zone of inhibition of 23.0 mm. Product 92 (0.25% w/w iodine) resulted in a smaller inhibition of 12.1 mm, compared to product 9 ($P < 0.05$). For both *Sta. aureus* and *E. coli*, product 91 (1.35% w/w iodine) resulted in the largest inhibition of 18.2 mm and 16.3 mm, respectively. Similar to *Str. uberis*, product 92 resulted in a smaller inhibition for *Sta. aureus* (9.2 mm) and *E. coli* (7.5 mm), which differed significantly from product 91 ($P < 0.05$). Product 24 (0.5% w/w iodine) resulted in the second largest inhibition for *Str. uberis* (21.6 mm) and *Sta. aureus* (17.1 mm) and the fourth largest zone of inhibition for *E. coli* (12.0 mm).

Iodine and lactic acid

A total of five products which contained iodine combined with lactic acid were evaluated. These products ranged in concentrations from 0.15% w/w to 0.5% w/w iodine combined with 0.8% w/w to 1% w/w lactic acid. No significant difference was observed for *Str. uberis* among the iodine and lactic acid products. However, product 30 (0.5% w/w iodine combined with 0.5% w/w lactic acid) resulted in the numerically largest inhibition of 21.9 mm. Furthermore, product 57 (0.15% w/w iodine combined with 1% w/w lactic acid) resulted in the smallest inhibition (20.3 mm) for *Str. uberis*. Product 30 resulted in the largest inhibition of 16.5 mm and 12.1 mm against *Sta. aureus* and *E. coli*, respectively. Product 57 (0.15% w/w iodine combined with 1% w/w lactic acid) resulted in the smallest inhibition of 13.0 mm and 10.0 mm for *Sta. aureus* and *E. coli*, respectively, compared to product 30 ($P < 0.05$).

Lactic acid

Within the study, 15 products which contained various concentrations of lactic acid were tested. These products ranged in lactic acid concentration from 1.76% w/w to 8% w/w lactic acid. For *Str. uberis* and *E. coli*, product 94 (4% w/w lactic acid) resulted in the numerically largest inhibition of 22.4 mm and 18.2 mm, respectively. Product 88 (8% w/w lactic acid) showed a significantly smaller inhibition of 16.6 mm for *Str. uberis* (compared to product 94 [22.4 mm] [$P < 0.05$]). For *E. coli*, product 10 (3.6% w/w lactic acid) showed a significantly smaller inhibition of 10.0 mm compared to product 94 (18.2 mm) ($P < 0.05$). Products 10 and 88 both had the numerically largest inhibition of 19.3 mm for *Sta. aureus*, which for both products was significantly larger than that for product 96 (5% w/w lactic acid [13.7 mm]) ($P < 0.05$). Across the lactic acid teat disinfectant products, product 94 resulted in the numerically largest inhibitions against both *Str. uberis* and *E. coli*, and the third largest inhibition for *Sta. aureus*.

Lactic acid and chlorhexidine

Of the 96 products tested, 21 of these products contained a combination of lactic acid and chlorhexidine. These products

Table 2: Least squares means of the zones of inhibition (mm) for 96 teat disinfectant products, categorised by ingredient group, against *Str. uberis*, *Sta. aureus* and *E. coli*

Product	#	Ingredient (w/w)	*Str. uberis*	*Sta. aureus*	*E. coli*
Chlorhexidine products					
Arrabawn Udder Guard[1]	40	0.5% Chlorhexidine	$16.6^{d,e,f,g}$	$15.3^{b,c,d}$	$16.2^{b,c,d}$
C-Dip[1]	61	0.53% Chlorhexidine	$17.6^{d,e,f,g}$	$15.7^{b,c,d}$	$15.6^{b,c,d,e}$
Deosan Mastocide[1]	32	0.5% Chlorhexidine	$19.1^{a,b,c,d,e,f}$	$17.4^{a,b,c}$	$16.7^{b,c,d}$
Deosan Summer Teat Care[1]	33	0.425% Chlorhexidine	$17.4^{d,e,f,g}$	$16.7^{a,b,c,d}$	$17.9^{a,b,c,d}$
Deosan Teat Foam Advance[1]	13	0.6% Chlorhexidine	$19.5^{a,b,c,d,e}$	$16.1^{a,b,c,d}$	$16.3^{b,c,d}$
Deosan Teatcare Plus[1]	14	0.425% Chlorhexidine	15.8^{g}	$16.9^{a,b,c,d}$	$17.6^{a,b,c,d}$
Hamra Red[1]	12	0.42% Chlorhexidine	$17.8^{b,c,d,e,f,g}$	$16.0^{b,c,d}$	$15.9^{b,c,d,e}$
Hexa-cel RTU[1]	42	0.52% Chlorhexidine	$15.9^{f,g}$	$16.1^{a,b,c,d}$	$15.1^{b,c,d,e}$
Hexaguard[1]	1	0.74% Chlorhexidine	21.4^{a}	$17.7^{a,b}$	$18.3^{a,b,c}$
Hexaklene R[1]	2	0.5% Chlorhexidine	$20.9^{a,b,c}$	$15.9^{b,c,d}$	20.6^{a}
Hexa-Spray[1]	82	0.5% Chlorhexidine	$17.3^{d,e,f,g}$	14.0^{d}	12.3^{e}
Kenocidin Spray/Dip[1]	9	0.5% Chlorhexidine	$17.0^{d,e,f,g}$	$16.2^{a,b,c,d}$	$14.2^{d,e}$
Kenomint[1]	77	0.5% Chlorhexidine	$17.7^{c,d,e,f,g}$	$16.0^{b,c,d}$	$16.3^{b,c,d}$
Kenomint SD[1]	78	0.5% Chlorhexidine	$19.1^{a,b,c,d,e,f}$	$14.6^{c,d}$	$14.5^{c,d,e}$
Masodip Platinum[1]	84	0.436% Chlorhexidine	$16.4^{e,f,g}$	$17.9^{a,b}$	$14.4^{d,e}$
PureChem Chlorhexidine[1]	48	1.49% Chlorhexidine	15.6^{g}	$15.0^{b,c,d}$	$14.8^{b,c,d,e}$
PureChem Chlorhexidine Summer Grade[1]	52	1.49% Chlorhexidine	$17.4^{d,e,f,g}$	$16.3^{a,b,c,d}$	$16.3^{b,c,d}$
Quatro[1]	41	0.5% Chlorhexidine	$16.1^{f,g}$	$16.6^{a,b,c,d}$	$16.5^{b,c,d}$
SensoDip[1]	72	0.5% Chlorhexidine	$17.0^{d,e,f,g}$	$15.9^{b,c,d}$	$15.9^{b,c,d,e}$
SensoDip 50[1]	16	0.5% Chlorhexidine	$18.4^{a,b,c,d,e,f,g}$	$17.4^{a,b,c}$	$17.3^{a,b,c,d}$
Sensospray[1]	66	0.5% Chlorhexidine	$16.9^{d,e,f,g}$	$17.1^{a,b,c,d}$	$14.9^{b,c,d,e}$
Summer C-Dip[1]	58	0.5% Chlorhexidine	$19.4^{a,b,c,d,e,}$	$15.1^{b,c,d}$	$14.5^{c,d,e}$
Supergold[1]	18	0.5% Chlorhexidine	$21.0^{a,b}$	$16.5^{a,b,c,d}$	$18.5^{a,b}$
Surespray[1]	5	0.5% Chlorhexidine	$19.8^{a,b,c,d}$	19.2^{a}	$18.2^{a,b,c}$
Teat Gard C[1]	63	0.5% Chlorhexidine	$16.0^{f,g}$	$15.2^{b,c,d}$	$15.4^{b,c,d,e}$
Chlorine dioxide products					
Bisept[2]	70	0.05% Chlorine dioxide	19.4^{c}	18.5^{b}	12.2^{c}
Kenomix[2]	11	0.0157% Chlorine dioxide	22.6^{a}	18.2^{b}	$19.3^{a,b}$
Kenomix SD[2]	89	0.0157% Chlorine dioxide	21.0^{b}	$21.4^{a,b}$	$20.5^{a,b}$
Uddergold[2]	90	0.32% Acidified sodium chlorite	$20.9^{b,c}$	$19.5^{a,b}$	17.3^{b}
Valiant[2]	95	0.038% Sodium chloride	22.8^{a}	22.4^{a}	21.5^{a}
Diamine products					
Super Cow Teat Foam[1]	26	0.6% Diamine	16.1	14.5	13.6
Iodine products					
D 4 Iodine[3]	19	0.5% Iodine	$21.4^{a,b,c}$	$16.1^{a,b,c}$	10.9^{b}
Deosan Super Iodip[3]	34	0.5% Iodine	$20.0^{b,c,d,e}$	$15.2^{b,c,d,e}$	11.7^{b}
Ioguard[3]	3	0.5% Iodine	$21.0^{a,b,c,d}$	$13.5^{c,d,e}$	$10.2^{b,c}$
Ioklar Multi[1]	92	0.25% Iodine	12.1^{g}	9.2^{f}	7.5^{c}
Io-Shield D[1]	91	1.35% Iodine	$19.0^{d,e,f}$	18.2^{a}	16.3^{a}
Io-Shield Spray[1]	93	0.5% Iodine	17.3^{f}	$14.5^{b,c,d,e}$	12.8^{b}

Table 2: Continued

Product	#	Ingredient (w/w)	*Str. uberis*	*Sta. aureus*	*E. coli*
Luxdip 50B[1]	69	0.5% Iodine	19.2^{c,d,e,f}	13.7^{c,d,e}	11.4^{b}
Masocare Platinum[1]	85	0.54% Iodine	19.3^{c,d,e,f}	14.5^{b,c,d,e}	12.4^{b}
Masodine Concentrate[3]	83	0.5% Iodine	20.7^{a,b,c,d,e}	15.5^{b,c,d}	10.8^{b}
Maxadine C[3]	8	0.5% Iodine	23.0^{a}	15.8^{a,b,c,d}	11.9^{b}
Maxidine RTU[1]	37	0.5% Iodine	18.9^{d,e,f}	13.3^{d,e}	11.4^{b}
PureChem Iodophor[3]	50	0.5% Iodine	18.6^{e,f}	12.6^{f}	10.5^{b}
Silkdip[3]	24	0.5% Iodine	21.6^{a,b}	17.1^{a,b}	12.0^{b}
Iodine and lactic acid products					
Gold Glycodip XL[1]	62	0.5% Iodine/1% lactic acid	21.6^{a}	15.3^{a,b}	12.0^{a}
Lanodip 4 XL[1]	30	0.5% Iodine/0.5% lactic acid	21.9^{a}	16.5^{a}	12.1^{a}
Lanodip Pre-Post[1]	55	0.29% Iodine/0.8% lactic acid	21.1^{a}	13.3^{b,c}	10.1^{b}
TriCide[1]	56	0.15% Iodine/1% lactic acid	21.1^{a}	13.0^{c}	10.0^{b}
TriCide Gold[1]	57	0.15% Iodine/1% lactic acid	20.3^{a}	13.2^{c}	10.1^{b}
Lactic acid products					
Barri-max[1]	65	2.4% Lactic acid	19.7^{b,c,d}	16.3^{a,b,c}	13.6^{b}
Blu-gard N Spray[1]	15	3.46% Lactic acid	19.9^{b,c,d}	15.6^{b,c}	11.4^{b,c,d}
Dairy Pro UltraDip[1]	74	3% Lactic acid	17.1^{e,f}	16.1^{a,b,c}	12.1^{b,c,d}
DairyLac SA[1]	76	3% Lactic acid	18.8^{d,e,f}	14.5^{c}	11.8^{b,c,d}
Deosan Triathalon[1]	81	1.76% Lactic acid	19.7^{b,c,d}	14.2^{c}	11.4^{b,c,d}
Flexigard Spray[1]	94	4% Lactic acid	22.4^{a}	19.2^{a}	18.2^{a}
Kenolac[1]	10	3.6% Lactic acid	22.3^{a}	19.3^{a}	10.0^{d}
Kenolac SD[1]	80	3.6% Lactic acid	19.a^{c,d,e}	15.0^{b,c}	13.2^{b,c}
Kenopure[1]	79	3.2% Lactic acid	21.6^{a,b}	16.4^{a,b,c}	11.2^{b,c,d}
Lacto-cel[1]	35	2.4% Lactic acid	19.9^{b,c,d}	15.9^{b,c}	12.1^{b,c,d}
Lacto-Mil[1]	96	5% Lactic acid	19.1^{c,d,e}	13.7^{c}	10.6^{c,d}
Lactospray[1]	4	2.4% Lactic acid	21.3^{a,b,c}	19.2^{a}	10.2^{d}
SalvoDip B[1]	71	2.4% Lactic acid	19.6^{b,c,d}	16.6^{a,b,c}	11.5^{b,c,d}
Salvospray[1]	68	2.4% Lactic acid	18.8^{d,e,f}	17.8^{a,b}	11.5^{b,c,d}
Synofilm[1]	88	8% Lactic acid	16.6^{f}	19.3^{a}	17.3^{a}
Lactic acid and chlorhexidine products					
Arkshield[1]	7	5% Lactic acid/0.3% chlorhexidine	21.6^{a,b,c}	18.9^{a,b,c}	15.4^{b,c,d}
Bacto-Lac[1]	31	5% Lactic acid/0.05% chlorhexidine	18.8^{c,d,e,f}	15.9^{d,e}	14.1^{c,d}
Blue Barrier Spray[1]	49	Lactic acid/0.6% chlorhexidine[4]	22.3^{a}	21.7^{a}	20.3^{a}
Co-op Source Duo-Teat Shield[1]	39	2% Lactic acid/0.3% chlorhexidine	18.1^{d,e,f}	16.5^{c,d,e}	15.4^{b,c,d}
Dual Dip[1]	45	2% Lactic acid/0.3% chlorhexidine	18.2^{d,e,f}	16.0^{d,e}	15.0^{b,c,d}
Dual Dip Supreme[1]	47	Lactic acid/0.6% Chlorhexidine[4]	21.8^{a,b}	21.3^{a,b}	18.5^{a,b}
Duo-cel[1]	38	2.5% Lactic acid/0.3% chlorhexidine	18.5^{d,e,f}	16.4^{c,d,e}	16.2^{b,c}
Duogold[1]	17	2% Lactic acid/0.3% chlorhexidine	19.7^{a,b,c,d,e}	16.8^{c,d,e}	17.3^{a,b,c}
Duo-Teat Shield[1]	25	2% Lactic acid/0.3% chlorhexidine	17.9^{d,e,f}	16.9^{c,d,e}	16.8^{a,b,c}
Fortress Protect Film[1]	73	3% Lactic acid/0.2% chlorhexidine	20.3^{a,b,c,d,e}	17.6^{c,d,e}	13.9^{c,d}
Hypraspray[1]	87	2% Lactic acid/0.03% chlorhexidine	19.1^{b,c,d,e}	16.7^{c,d,e}	12.2^{d}

Table 2: Continued

Product	#	Ingredient (w/w)	*Str. uberis*	*Sta. aureus*	*E. coli*
Lacto dual[1]	36	2.5% Lactic acid/1.5% chlorhexidine	18.2^{d,e,f}	15.2^{e}	16.6^{b,c}
Nano Dual[1]	28	1.93% Lactic acid/0.2% chlorhexidine	21.6^{a,b,c}	16.9^{c,d,e}	14.8^{c,d}
Novo Dual[1]	29	4% Lactic acid/0.27% chlorhexidine	20.8^{a,b,c,d}	17.5^{c,d,e}	15.6^{b,c,d}
Novodip[1]	60	4.9% Lactic acid/1.28% chlorhexidine	19.3^{b,c,d,e}	19.1^{a,b,c}	16.0^{b,c}
Novospray[1]	54	4.9% Lactic acid/0.3% chlorhexidine	20.0^{a,b,c,d,e}	18.4^{b,c,d}	16.3^{b,c}
Protect Pre Post[1]	75	3% Lactic acid/0.25% chlorhexidine	21.7^{a,b}	18.2^{c,d}	14.0^{c,d}
PureChem Dual Dip[1]	51	1% Lactic acid/1.49% chlorhexidine	16.2^{f}	16.3^{c,d,e}	14.5^{a}
Salvohex[1]	67	2% Lactic acid/ 0.3% chlorhexidine	17.8^{e,f}	16.2^{c,d,e}	16. 8^{a,b,c}
Supreme[1]	64	2.5% Lactic acid/0.375% chlorhexidine	19.2^{b,c,d,e}	16.8^{c,d,e}	15.4^{b,c,d}
Sure Spray Duo[1]	6	2% Lactic acid/0.3% chlorhexidine	18.6^{d,e,f}	17.2^{c,d,e}	15.4^{b,c,d}
Lactic acid and hydrogen peroxide products					
Lactic Lather[1]	46	1.6% Lactic acid/ hydrogen peroxide[4]	27.9	25.1	19.3
Lactic acid and salicylic acid products					
Biolac Pre-Post[1]	44	0.25% Lactic acid/0.03% salicylic acid	19.7^{a,b}	16.0^{a}	11.4^{b}
Biolac Pre-Post[1]	59	0.25% Lactic acid/0.03% salicylic acid	21.0^{a}	15.1^{a}	11.8^{b,c}
Dermalac Emprasan[1]	27	0.25% Lactic acid/salicylic acid[4]	21.1^{a}	14.3^{a,b,c}	11.0^{b,c}
Emprasan dual[1]	53	0.25% Lactic acid/salicylic acid[4]	19.6^{a,b}	14.8^{a,b}	16.0^{a}
Hypred Quick Spray[1]	20	2% Lactic acid/0.1% salicylic acid	17.0^{c,d}	9.7^{d}	9.7^{c}
Lely Quaress-Cura[1]	43	3% Lactic acid/salicylic acid[4]	18.9^{a,b,c}	15.9^{a}	12.4^{b}
Prefoam+[1]	21	2% Lactic acid/0.1% salicylic acid	18.1^{b,c,d}	11.7^{b,c,d}	11.1^{b,c}
Virolac Concentrate[3]	22	2% Lactic acid/0.1% salicylic acid	16.8^{c,d}	11.2^{c,d}	10.8^{b,c}
Virolac Film[1]	23	2% Lactic acid/0.1% Salicylic acid	17.2^{c,d}	11.7^{b,c,d}	11.0^{b,c}
Virolac Spray[1]	86	2% Lactic acid/0.1% salicylic acid	16.0^{d}	11.3^{c,d}	10.3^{b,c}

[1]Ready to use (RTU).
[2]Requires activation before use.
[3]Concentrate.
[4]The concentration of some active ingredients for combination products was not declared by the manufacturer.
^{a,b,c,d,e,f,g}Inhibitions not sharing the same superscript in a column within an ingredient group were significantly different ($P < 0.05$).
= product number.

ranged from 1% w/w to 5% w/w lactic acid combined with 0.03% w/w to 1.5% w/w chlorhexidine. Within the lactic acid and chlorhexidine group, product 49 (lactic acid combined with 0.6% w/w chlorhexidine) resulted in the largest inhibitions against *Str*. *uberis*, *Sta*. *aureus* and *E*. *coli* of 22.3 mm, 21.7 mm and 20.3 mm, respectively. Furthermore, product 47 (lactic acid combined with 0.6% w/w chlorhexidine) resulted in the second largest inhibitions for *Str*. *uberis* (21.8 mm), *Sta*. *aureus* (21.3 mm) and *E*. *coli* (18.5 mm) which did not differ significantly from product 49. In comparison to products 49 and 47, product 51 (1% w/w lactic acid combined with 1.49% w/w chlorhexidine) resulted in a smaller inhibition of 16.2 mm for *Str*. *uberis*. Alternatively, product 36 (2.5% w/w lactic acid combined with 1.5% w/w chlorhexidine) showed the smallest inhibition for *Sta*. *aureus* (15.2 mm) and product 87 (2% w/w lactic acid combined with 0.03% w/w chlorhexidine) showed the smallest inhibition for *E*. *coli* (12.2 mm), both of which were significantly different from products 49 and 45 ($P < 0.05$).

Lactic acid and hydrogen peroxide

Only one product contained a combination of the ingredients lactic acid and hydrogen peroxide (product 46; 1.6% w/w lactic acid combined with hydrogen peroxide). This product resulted in inhibitions of 27.9 mm, 25.1 mm and 19.3 mm for *Str*. *uberis*, *Sta*. *aureus* and *E*. *coli*, respectively.

Lactic acid and salicylic acid

Within the trial, 10 products contained a combination of lactic acid and salicylic acid. These products ranged from concentrations of 0.25% w/w to 3% w/w lactic acid combined

with 0.03% w/w to 0.1% w/w salicylic acid. For *Str. uberis*, product 27 (0.25% w/w lactic acid combined with salicylic acid) resulted in a significantly greater zone of inhibition of 21.1 mm compared to product 86 (2% w/w lactic acid combined with 0.1% w/w salicylic acid [16.0 mm]) ($P < 0.05$). For *Sta. aureus*, product 44 (0.25% w/w lactic acid combined with 0.03% w/w salicylic acid) resulted in the largest zone of inhibition of 16.0 mm. This was significantly different to product 20 (2% w/w lactic acid combined with 0.1% w/w salicylic acid) which had the smallest zone of inhibition of 9.7 mm ($P < 0.05$). For *E. coli*, product 53 (0.25% w/w lactic acid combined with salicylic acid) showed the largest zone of inhibition of 16.0 mm, which differed significantly from product 20 with the smallest zone of inhibition of 9.7 mm ($P < 0.05$).

While all individual products were not statistically compared, some products were observed to have numerically higher inhibitions against each bacterial strain tested than others within the study. Product 46 was found to result in the largest zones of inhibition for both *Str. uberis* (27.9 mm) and *Sta. aureus* (25.1 mm), with product 95 (0.038% w/w chlorine dioxide) achieving the largest zone of inhibition for *E. coli* (21.5 mm).

Discussion

Mastitis control programmes recommend the use of teat disinfection, with some recommending both pre- and post-milking disinfection. By testing teat disinfectant products against bacteria that have previously been identified as the most prevalent mastitis-causing bacteria in Ireland (Keane *et al*., 2013), the effectiveness of these products can be estimated for use in Ireland. The results of this study show that the range of teat disinfection products showed variation in bacterial inhibition against *Str. uberis*, *Sta. aureus* and *E. coli*, with some individual products and ingredient groups resulting in greater bacterial inhibitions than others.

The chlorine dioxide ingredient group showed the greatest zones of inhibition for *Str. uberis*, *Sta. aureus* and *E. coli*, which was significantly different to the iodine group for all three bacterial strains. Chlorine dioxide (1%) was previously shown to have large log percentage reductions against *Sta. aureus*, *E. coli* and *Str. uberis* when tested using the excised teat method (Enger *et al*., 2015). Furthermore, Santos *et al*. (2016) demonstrated, *in vitro*, that a 2.5% chlorine dioxide product resulted in reduction levels comparable to a 0.6% iodine product at four different exposure times (15 s, 30 s, 60 s and 300 s) against 50 *Sta. aureus* strains. However, it has also been stated that chlorine dioxide may be less effective when applied to the teat skin as it can be highly reactive towards organic matter which may be present on the skin surface (Lopes *et al*., 2012).

Two ingredient groups within the study contained just one product each; the variation in product numbers within product ingredient groups may represent a limitation within the study. These ingredient groups include diamine (product 26) and lactic acid combined with hydrogen peroxide (product 46). The product containing diamine resulted in small zones of inhibition for all three bacterial strains. This was similar to a previous study where the same diamine product resulted in some of the smallest zones of inhibition against three *Sta. aureus* isolates, a *Str. uberis* isolate and an *E. coli* isolate (Fitzpatrick *et al*., 2019a). However, when this product was applied to teat skin, it resulted in some of the highest reductions of staphylococcal and streptococcal isolates naturally present on the teat skin (Fitzpatrick *et al*., 2019b). This could be due to the ingredient diamine being less affected by the presence of organic matter than other ingredients and it is also stable at a wide range of pH (Mondin *et al*., 2014). This may allow products having this ingredient to be less affected by organic matter on teat skin. In this study, product 46 containing lactic acid combined with hydrogen peroxide had the greatest zones of inhibition for *Str. uberis* and *Sta. aureus*. Previously, two hydrogen peroxide products (0.5%) have been shown to achieve a >5 log reduction against *Sta. aureus*, *Str. uberis* and *E. coli*, with these products also being comparable to two chlorine dioxide products (Lopez-Benavides *et al*., 2012). Furthermore, the use of a hydrogen peroxide product for both pre- and post-milking was shown to reduce the bacterial contamination on teat skin by 65% (Miseikiene *et al*., 2019). The use of hydrogen peroxide within teat disinfectant products proves to be effective at reducing bacterial load on teat skin but its impact on teat skin condition must be evaluated.

Additionally, a product containing 0.038% w/w chlorine dioxide (product 95) was found to achieve the greatest zones of inhibition (21.5 mm) against *E. coli*. Also, a 1% chlorine dioxide product resulted in a large log reduction against an *E. coli* strain, which was comparable to both a 0.5% and 1% iodophor teat disinfectant, when using the excised teat method (Enger *et al*., 2015). Furthermore, the use of a chlorine dioxide (0.0157%) product reduced naturally present coliforms on the teat skin by 87.9% (Fitzpatrick *et al*., 2019b). However, there may be some negative aspects associated with the use of chlorine dioxide products. All chlorine dioxide products used in this study had to be activated/mixed prior to use; also, depending on the product, the time limit for recommended usage after activation ranged from 24 h up to 26 d.

In the current study, only two ingredient groups showed a trend in effectiveness with increasing concentrations of the active ingredients. These groups included iodine and a combination of iodine and lactic acid. Within the iodine ingredient group, the highest concentration of iodine (1.35% w/w) resulted in the largest zones of inhibition for both *Sta. aureus* and *E. coli*, which was significantly different to the

lowest concentration of 0.25% w/w iodine, which was also effective for *Str. uberis*. In the iodine combined with lactic acid group, the concentrations of 0.5% w/w iodine with 0.5% w/w lactic acid and 0.5% w/w iodine with 1% w/w lactic acid resulted in some of the numerically largest inhibitions for all three bacterial strains. A decrease in effectiveness could be observed within these products as iodine levels were reduced. However, products which contained higher concentrations of ingredients, within the other ingredient groups, for example, product 48 (1.49% w/w chlorhexidine), did not always result in the highest level of reduction as would be expected. Limited information with regard to emollient levels in products was available; therefore, the impact of those teat condition agents on the effectiveness of the products could not be evaluated.

The disc diffusion method used in the current study allows for an effective screening of a large number of teat disinfectant products in a short period of time. However, laboratory methods do not evaluate the true efficacy of a teat disinfectant product. Therefore, further studies must be performed to determine the ability of the products to reduce: (1) bacterial load on the teat skin, (2) new IMIs and (3) impact on teat skin condition.

Conclusion

This study has shown that there is a range of alternative teat disinfectant products available which reduce bacterial growth comparable to iodine-based products. The concentration of active ingredient did not influence the effectiveness in the majority of teat disinfectant products. Additionally, different products/ingredients were more effective against specific strains of bacteria within the study. The disc diffusion method is an effective method to screen a large number of teat disinfectant products, but field trials would be required to fully determine the products' effectiveness in reducing the bacterial load on teat skin and the ability of the products to reduce IMIs.

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