



(86) Date de dépôt PCT/PCT Filing Date: 1998/04/27
(87) Date publication PCT/PCT Publication Date: 1999/04/08
(45) Date de délivrance/Issue Date: 2008/07/08
(85) Entrée phase nationale/National Entry: 2000/03/23
(86) N° demande PCT/PCT Application No.: US 1998/008491
(87) N° publication PCT/PCT Publication No.: 1999/016418
(30) Priorité/Priority: 1997/09/26 (US08/938,653)

(51) Cl.Int./Int.Cl. *A61K 33/20* (2006.01),
A61K 31/19 (2006.01), *A61K 31/20* (2006.01),
A61K 9/08 (2006.01), *A61P 31/04* (2006.01)

(72) Inventeurs/Inventors:
PAQUETTE, CATHY M., US;
STAUB, RICHARD K., US;
RICHTER, FRANCIS L., US

(73) Propriétaire/Owner:
ECOLAB, INC., US

(74) Agent: CASSAN MACLEAN

(54) Titre : LIQUIDE POUR TREMPAGE DES TRAYONS CONSTITUE DE CHLORURE AQUEUX ACIDE PRESENTANT
UNE LONGUE DUREE DE VIE DE STOCKAGE ET UNE CAPACITE DE DESINFECTION ET APPORTANT UNE
PROTECTION DES TISSUS

(54) Title: ACIDIC AQUEOUS CHLORITE TEAT DIP PROVIDING SHELF LIFE, SANITIZING CAPACITY AND TISSUE
PROTECTION

(57) **Abrégé/Abstract:**

The mastitis control teat dip composition of the invention provides rapid initial kill, a useful highly pseudoplastic rheology, a barrier/film-forming capacity, a unique antimicrobial composition that is stable over an extended period of time, and unexpected long term microbial control when compared to the prior art materials disclosed in patents and used in the marketplace. The compositions of the invention are made by combining an aqueous thickened liquid composition containing the organic components which can be combined with a simple aqueous solution of a salt of chlorous acid, preferably an alkali metal chlorite. The materials can be combined blended into a smooth viscous material and can be immediately contacted with the target animals. The compositions of the invention provide rapid initial kill, consistent long term kill and chemical and rheological stability.

**PCT**WORLD INTELLECTUAL PROPERTY ORGANIZATION
International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification ⁶ : A61K 9/00, 31/185, 31/19, 31/20, 33/00, A01N 25/24, 37/00, 37/02, 37/06, 59/00	A1	(11) International Publication Number: WO 99/16418 (43) International Publication Date: 8 April 1999 (08.04.99)
(21) International Application Number: PCT/US98/08491 (22) International Filing Date: 27 April 1998 (27.04.98) (30) Priority Data: 08/938,653 26 September 1997 (26.09.97) US (71) Applicant: ECOLAB INC. [US/US]; Ecolab Center, St. Paul, MN 55102 (US). (72) Inventors: RICHTER, Francis, L.; 6707 Sherman Lake Road, Lino Lakes, MN 55038 (US). PAQUETTE, Cathy, M.; 551 113th Lane N.W., Coon Rapids, MN 55448 (US). STAUB, Richard, K.; 16208 Goodview Trail, Lakeville, MN 55044 (US). (74) Agent: DAIGNAULT, Ronald, A.; Merchant, Gould, Smith, Edell, Welter & Schmidt, P.A., 1000 Norwest Center, 55 E. Fifth Street, St. Paul, MN 55101 (US).		(81) Designated States: AU, BR, CA, CZ, HU, JP, KE, MX, NZ, PL, PT, RO, RU, UA. Published <i>With international search report.</i>
(54) Title: ACIDIC AQUEOUS CHLORITE TEAT DIP PROVIDING SHELF LIFE, SANITIZING CAPACITY AND TISSUE PROTECTION (57) Abstract The mastitis control teat dip composition of the invention provides rapid initial kill, a useful highly pseudoplastic rheology, a barrier/film-forming capacity, a unique antimicrobial composition that is stable over an extended period of time, and unexpected long term microbial control when compared to the prior art materials disclosed in patents and used in the marketplace. The compositions of the invention are made by combining an aqueous thickened liquid composition containing the organic components which can be combined with a simple aqueous solution of a salt of chlorous acid, preferably an alkali metal chlorite. The materials can be combined blended into a smooth viscous material and can be immediately contacted with the target animals. The compositions of the invention provide rapid initial kill, consistent long term kill and chemical and rheological stability.		

**ACIDIC AQUEOUS CHLORITE TEAT DIP PROVIDING
SHELF LIFE, SANITIZING CAPACITY AND TISSUE PROTECTION**

Field of the Invention

5 The invention relates to a bovine teat dip composition that can be mixed using two parts, a simple chlorite solution and an acid or acidulant formulation, to form a stable, effective composition that can be used in routine dairy procedures.

Background of the Invention

10 Bovine mastitis is the most common and most costly disease affecting dairy herds. Some estimates suggested at least half of the dairy animal population have some degree or form of mastitis. This condition results in lowered milk yield and reduced quality. Economic loss to mastitis in the U.S. is estimated at about \$1.8
15 billion or approximately 10% of total milk sales with about two-thirds of this loss due to reduced milk production from infected cows. Mastitis is an inflammation of the mammary gland. Similarly, inflammation is one response of a tissue or organ to insult or injury. An injury caused by physical, chemical or thermal trauma can produce an inflammatory response. In the dairy cow, mastitis typically results from
20 microorganisms, usually bacteria, that invade the udder, multiply in the delicate milk producing tissues, and synthesize toxins, a by-product of bacterial metabolism. The characteristic features of inflammation are swelling, heat, redness, pain and disturbed function.

 While the animal immune system can fight intramammary infections, many
25 chronic infections remain sub-clinical (asymptomatic) and undetected unless diagnosed by laboratory testing. Sub-clinical mastitis can result in a reservoir of micro-organisms that leads to the infection of other animals within the herd. More than 80 species of microorganisms have been identified as causal agents, although approximately 95% of mastitis is caused by four pathogens; Staphylococcus aureus,
30 Streptococcus agalactiae, Streptococcus dysagalactiae, and Streptococcus uberis. Mastitis causing pathogens fall into two categories namely contagious and environmental. Contagious bacteria, such as Streptococcus agalactiae and Staphylococcus aureus, primarily colonize host tissue sites such as mammary glands,

teat canals, teat skin lesions etc. and are spread from one infected cow to another during the milking process. Environmental bacteria, often streptococci, enterococci and coliform organisms, are commonly present within the cow's surroundings from sources such as cow feces, soil, plant material, bedding or water, and infect by casual opportunistic contact with an animal during the inter-milking period. This distinction, although not exclusive, is of practical importance because different dairy herd maintenance measures are needed for the different groups of microorganisms. In all bovine mastitis cases, whatever the causal microorganism, the route of transmission of the invading pathogen into the inner gland of the udder is through the teat orifice and teat canal.

Management of dairy herds focuses attention on treatment of both established mastitis and on prevention of new intramammary infections. Therapy and hygiene are the two fundamental components of an effective mastitis control protocol. Each is applied in concert, and each operates independently. The primary effect of therapy is to increase the rate of eliminating established infections; whereas, hygiene reduces the frequency of infection by interrupting transmission vectors. We cannot present all ancillary factors that may be employed for the elimination and prevention of mastitis, however, the most effective therapy and hygiene practices are, respectively, antibiotic infusion treatment of the udder's four quarters at the end of lactation: and, post-milking teat antisepsis or "teat dipping" during lactation.

Researchers agree, and an abundance of published evidence supports the concept, that dipping teats into an effective antimicrobial solution immediately after each milking is the single most effective procedure for decreasing new intramammary infections in lactating cows. Between 1955 to 1970, Dodd and co-workers (F. K. Neave, F. H. Dodd, and R. G. Kingwell, 1966, "A Method of Controlling Udder Disease", Vet. Rec. 78:521; F. K. Neave, F. H. Dodd, R.G. Kingwell and D. R. Westgarth, 1969, "Control of Mastitis in the Dairy Herd by Hygiene and Management", J. Dairy Sci. 52:696; F. H. Dodd, D. R. Westgarth, F. K. Neave and R. G. Kingwill, 1969, "Mastitis - The Strategy of Control", J. Dairy Sci. 52:689; and F. H. Dodd, and F. K. Neave, 1970, "Mastitis Control", Proceedings, Nat'l. Inst. Res. Dairying, pp. 21-60) conducted extensive

epidemiologic investigations in commercial dairy herds. From this work, they developed the conceptual basis for modern mastitis control methods of which teat dipping is an integral component. The efficacy and value of teat dipping has since been confirmed in dozens of field trials, and it is now accepted that an effective teat
5 dip can reduce the incidence of new intramammary infections at least 50% and often up to 90%.

To reduce mastitis, commercial teat dips have been developed containing a variety of antimicrobial agents including iodophors, quaternary ammonium compounds, chlorhexidine salts, chlorine release compounds (e.g. alkali
10 hypochlorites), oxidizing compounds (e.g. hydrogen peroxide, peracids), protonated carboxylic acids (e.g. heptanoic, octanoic, nonanoic, decanoic, undecanoic acids), acid anionics (e.g. alkylaryl sulfonic acids), and chlorine dioxide (from chlorite). These agents, which have varying degrees of effectiveness, limit the transmission of mastitis by reducing pathogen populations on the teat. Teat dips, can also be divided
15 into two broad classifications. The Class I type are antimicrobial and are applied to kill microorganisms already present in the teat canal or on the surface of the teat skin. By design, their microbiological effect is immediate and their targets primarily the contagious organisms that are vectored between animals during the pre-milking, milking and post milking process. The Class II type teat dip, often referred to as a
20 "teat sealer," is a film-forming or coating composition which may or may not be antimicrobial; and, functions by developing a residual protective barrier on the teat thus providing prophylaxis by sealing the teat off from its environment. The film which forms on the surface of the teat serves as a physical barrier through which mastitis causing pathogens cannot penetrate during the intermilking period.

25 General disclosures of teat dip technology are shown in: "Current Concepts of Bovine Mastitis." 1996, Fourth Ed. National Mastitis Council, Madison WI.; P. A. Murdough and J. W. Pankey, 1993. "Evaluation of 57 Teat Sanitizers Using Excised Cow Teats", J. Dairy Sci. 76:2033-2038; J. W. Pankey et al., 1984, "Uptake on Post-milking Teat Antiseptics", J. Dairy Sci. 67:1336-1353; R. J. Farnsworth,
30 1980, "Role of Teat Dips in Mastitis Control", J. Am. Vet. Med. Assoc. 76:1116-1118; W. N. Philpot, 1979, "Control of Mastitis by Hygiene and Therapy", J. Dairy Sci. 62:168-176; W. N. Philpot and J. W. Pankey, 1978, "Hygiene in the Prevention

of Udder Infections V. Efficacy of Teat Dips Under Experimental Exposure to Mastitis Pathogens", J. Dairy Sci. 61:956-963; R. P. Natzke, 1977, "Role of Teat Dips and Hygiene in Mastitis Control", J. Amer. Vet. Med. Assoc. 170:1196-1198; W. N. Philpot and J. W. Pankey, 1975, "Hygiene in the Prevention of Udder Infections. III. Effectiveness of 59 Teat Dips for Reducing Bacterial Populations on Teat Skin", J. Dairy Sci. 58:209-216; R. J. Eberhart and J. M. Buckalew, 1972, "Evaluation of a Hygiene and Dry Period Therapy Program for Mastitis Control", J. Dairy Sci. 55:1683-1691; W. D. Schultze and J. W. Smith, 1972, "Effectiveness of Postmilking Teat Dips", J. Dairy Sci. 55:426-431; D. P. Wesen and L. H. Schultz, 1970, "Effectiveness of a Post-Milking Teat Dip in Preventing New Udder Infections". J. Dairy Sci. 53:1391-1403; and British Pat. No. 1,144,637 (Kelco Chemicals Ltd.), published March 5, 1969. U. S. Pat. No. 4,199,602 (Lentsch) issued April 22 1980, U.S. Pat. No. 4,258,056 (Lentsch) issued March 24, 1981; and U.S. Pat. No. 4,376,787 (Lentsch) issued March 15, 1983 disclose nitroalkanol, amino carboxylate/sulfonate, and sulfonate based compositions. U.S. Pat. No. 4,446,153 (Yang) issued May 1, 1984 discloses a benzyl alcohol/phenyl ethanol based composition.

Typical disclosures of intermilking or protective (barrier-type) film-forming teat dips or teat "sealers" can be found in Akers et. al., U. S. Pat. No. 3,066,071, issued November 27, 1962; Kraus, U.S. Pat. No. 3,222,252, issued December 7, 1965 (but, see Philpot et. al., J. Dairy Science 58:205-216); Coughman and Brown, U.S. Pat. No. 3,993,777, issued November 23, 1976; Pugliese, U. S. Pat. No. 4,049,830, issued September 20, 1977; and Andrews et al., U. S. Pat. No. 4,113,854, issued September 12, 1978. A teat sealer similar or identical to the Andrews et. al. film-forming composition is in commercial use and has been discussed in the dairy science literature. See, for example, R. J. Farnsworth et. al., 1980, "Use of a Teat Sealer for Prevention of Intramammary Infections in Lactating Cows", J. Am. Vet. Med. Assoc. 177:441-444; and R. J. Farnsworth et. al., 1981, "The Effect of a Teat Sealer on Coliform Mastitis", The Bovine Practitioner, No. 16, pp. 28-29. Still further examples of barrier-type film-formers for bovine teats can be found in Silver et al., U.S. Pat. No. 4,199,564, issued April 22, 1980; Dybas et al., U.S. Pat. No.

4,311,709, issued January 19, 1982; Marhavka, U.S. Pat. No. 5,017,369, issued May 21, 1991; and Schmidt et al., U.S. Pat. No. 5,503,838, issued April 2, 1996.

Persons skilled in the art of bovine mastitis treatment know that antimicrobial teat dip compositions (Class I type) which do not form protective films with barrier properties have a shortened residual time on the teat and their efficacy is quickly lost due to adsorption, ion-pairing, oxidation or simply sloughing off. Furthermore, such teat dips often fail to retard the entrance of bacteria into the teat canal during the intermilking period and provide no protection to the teat from irritation caused by wind, sun or contact abrasion. As disclosed in the art, attempts have been made to provide antimicrobial teat dips containing film-forming materials designed to form protective barriers on the skins of the teats; and, to afford continuous protection against mastitis causing pathogens, both contagious and environmental, and, from irritation caused by exposure to adverse environmental elements.

Early researchers discovered that incorporating a film-forming, protective barrier system into an antimicrobial teat dip was fraught with technical problems, either physicochemical problems with the composition or application/performance problems in practice. The best prophylactic barriers, and most environmentally durable, are water insoluble synthetic organics made up of homopolymers or heteropolymers of two or more different monomers. These are applied either from volatile solvent based compositions or by film-forming polymer latexes which are suspensions of a water insoluble polymer in water. U.S. Pat. No. 3,066,071 typifies the former type; and U.S. Pat. No. 4,113,854 discloses compositions typical of the latter kind. Typically, barrier films formed upon teats when applied from a volatile solvent based mastitis dip composition are not user, animal or environmentally friendly. In practice, these materials subject the skin of the teat to the drying, irritating effects of organic solvents. Compositions containing a film-forming polymer latex overcome volatile solvent concerns as polymer latexes are most often suspensions of a water insoluble polymer(s) in water; however, commercial latexes necessarily include stabilizers, preservatives, suspending agents etc. which add complexity; and, as such are frequently incompatible with the most preferred and most efficacious antimicrobial agents. All water insoluble polymeric films, which

generally form a flexible almost rubbery film on the teat skin, must be removed by peeling. In practice, such mastitis control compositions have not received widespread acceptance because of the inconvenient, time-consuming, often troublesome removal process prior to milking.

5 U.S. Pat. No. 3,222,252 describes a bovine teat dip consisting of vegetable oils of the drying or semi-drying types and certain fatty acid esters. In concept, this disclosure bridges the gap between synthetic polymer coatings and natural polymer coatings. In practice, oil based dips have proved to be ineffective in preventing mastitis, and they are difficult to remove from the teats. In fact, their use has tended
10 to increase the incidence of mastitis (see Philpot et. al, J. Dairy Science 58:205-216).

U.S. Pat. No. 3,993,777, referenced above, discloses a aqueous formulation of high viscosity which forms a protective film and bacteriostatic barrier about the teat which is easily removed by washing; thus, transitioning from water insoluble protective films which in practice are peeled from the teat to water washable films.
15 However, the shortfall of this teaching is the use of hydroxyethylcellulose employed as a thickener (its most typical function) in the preferred formulation which, by chance, has the characteristic of forming pliable non-brittle films upon drying. In herd practice, such cellulosic thickeners seldom perform the dual-function role of providing a tenacious barrier, being too readily removed because of its water-
20 sensitivity, wherein the performance of the antimicrobial barrier is lost. U.S. Pat. No. 4,311,709 also discloses a film-forming methycellulose having similar disadvantage as a teat dip barrier. U.S. Pat. No. 4,049,830 discloses a bovine teat dip composition which delivers an oil-in-water emulsion to the teat and, upon drying, forms an antimicrobial lipid solids barrier which remains soft and tacky for
25 prolonged periods and is water washable. Herd experience has shown that soft barriers are too easily abraded or otherwise sloughed off during the intermilking period with subsequent reduction or loss of biocidal function. U.S. Pat. No. 5,017,369 discloses antimicrobial mastitis treatment compositions which utilize a water resistant film-forming agent, polyvinyl alcohol. This art teaches away from
30 incorporation and use of thickener admixtures, suggesting that adequate viscosity can be obtained simply by adjusting the amount of polyvinyl alcohol in the composition. Such compositions are, in application, at commercial disadvantage

because polyvinyl alcohol by itself does not provide effective teat cling nor does it decrease the mobility of the dipping liquid which is manifested by excessive drain-off and loss of product; and, therefore significant reduction of barrier function and microbial performance. U.S. Pat. No. 5,503,838 overcomes this disadvantage in disclosing antimicrobial teat dip compositions containing polyvinyl alcohol in cooperation with thickening agents such as a xanthan gum. Unfortunately, neither U.S. Pat. No. 5,017,369 which typically embodies chlorhexidene gluconate or quaternary ammonium compounds as the antimicrobial; nor, U.S. Pat. No. 5,503,838 which uses iodine as the preferred biocidal agent, address the issue of teat dermal irritation caused by residual barrier films containing resident, chemically aggressive, often toxic, antimicrobial agents which remain on skin contact for extended periods of time.

Alliger, U.S. Pat. No. Re. 31,779, reissued December 25, 1984; Alliger, U.S. Pat. No. 4,330,531 (Alliger) issued May 18, 1982; Kross et al., U.S. Pat. No. 4,891,216, issued January 2, 1990; Davidson et al., U.S. Pat. No. 4,986,990, issued January 22, 1991; Davidson et al., U.S. Pat. No. 5,185,161, issued February 9, 1993; and Kross, U.S. Pat. No. 5,597,561 disclose technology embodied in a commercial composition sold as an antimicrobial barrier teat dip under the name UDDER-GOLD PLUS (Alcide Corp., Redmond, WA.). The patents disclose two aqueous solutions, described as gels, adapted to be mixed in a 50/50 proportion wherein (according to patent and product literature) chlorous acid/chlorine dioxide are generated by chemical reaction of α -hydroxy-benzene-acetic acid (mandelic acid) and sodium chlorite present in first and second gels respectively. The second gel can contain a thickener. Later patents disclose a homopolymer of 2-acrylamido-2-methylpropane sulfonic acid (polysulfonic acid) which forms a protective film over the teat. Although this composition has proven effective, it is not without problem. The herdsman is advised to mix and use the admixture for only one herd milking, discarding any extra. This attribute is likely a consequence of chlorine dioxide loss (hence, loss of antimicrobial efficacy) either from chemical incompatibility and/or off-gassing. The polysulfonic acid "gelling agent" does not immobilize the treatment on the teat and significant drippage/waste occurs. U.S. Pat. No. 5,597,561 teaches that polysulfonic acid has proven problematic because of its strong affinity

to the dermal tissue and tendency to form a solid matrix which is difficult to remove by washing.

A substantial need remains after all these attempts, for long lasting protective, film or shield forming barrier-type teat dip compositions having
5 immediate and long lasting antimicrobial effect against a wide spectrum of mastitis causing organisms.

Brief Description of the Invention

We have discovered a mastitis control treatment having important functional
10 properties which are uniquely combined into one composition. The compositions of the invention provide a rapid initial kill, long lasting antimicrobial activity, a stable chemistry and rheology. The compositions of the invention possess favorable rheology that promotes cling and immobilizes the mastitis treatment on the teat. The composition develops a barrier giving prophylactic protection and containing at least
15 one resident agent for continuous biocidal protection. The composition does not cause dermal irritation and, is removable by a simple water-washing technique. The barrier has sufficient adherence to withstand premature loss of integrity due to abrasion of environmental conditions. Compositions of this invention are mastitis control and prevention treatments often described as "teat dips," though of course
20 methods of topical aseptic application other than immersion or "flooding" might be used by the herdsman; for example, spraying, brushing, swabbing or foaming onto the teats. When employed as a teat dip, which is a particularly effective practice of application, the teats of the animal are dipped into a reservoir or receptacle containing a composition of the present invention; whereupon, the source is
25 removed, and, preferably one-half to three-fourths of the distal teat has been coated with treatment. After application (by whatever method), the resident treatment adheres onto the teat until drying occurs and development of a protective film results which provides an antimicrobial barrier and prophylactic shield thus protecting the teat from pathogens and adverse environmental factors.

30 The mastitis control compositions of the invention comprise components dissolved or suspended in an aqueous carrier or medium. The components of the compositions include a chlorine dioxide generating chemical such as sodium

chlorite, an acidulant that can contain a protic acid component and an active antimicrobial acid component, an organic film-forming agent, rheology modifying materials, a hydrotrope, an emollient, a surfactant, a buffer if needed, a colorant and other optional materials.

5 A pseudoplastic aqueous rheology is effected in the composition of the invention by admixtures of polymeric materials such as a xanthan gum and polyvinyl alcohol compositions. When shear stress is applied to the composition (i.e., dipping), product viscosity is reduced allowing easy and rapid application to the teat; and, upon the release of shear (i.e., removal of source), total viscosity
10 recovery occurs almost instantaneously immobilizing the coating, providing cling and assuring little waste by drippage. Further, said compositions have little or no viscoelastic character which thus allows the treatment to flow and to coat the teat smoothly, forming a continuous efficacious layer over the skin of the teat without formation of musilage streamers as the applicator is withdrawn. The compositions
15 flow slightly down the teat following application to form a thicker layer or "plug" across the orifice of the teat canal; and, thus cause a more effective prophylactic barrier against bacteria entering the teat canal.

 An occlusive polymeric barrier/film-forming attribute is contributed by inclusion of intermediate or fully hydrolyzed polyvinyl alcohol. Carefully
20 composed polyvinyl alcohol compositions of this invention provide to the mastitis control treatment, after drying, a balanced barrier layer which remains pliable and maintains integrity on the teat; which can be rendered antimicrobial by envelopment of biocidal agents; which does not cause irritation; and, which provides significantly improved and prolonged protection to the teat during the intermilking period by
25 structured adherence, yet does not sacrifice ease of removal prior to milking.

 A unique preferred antimicrobial composition is accomplished that can contain one fugitive biocidal agent, chlorine dioxide, and one or more non-fugitive acid biocidal agents such as a C₆₋₁₂ carboxylic acid, including heptanoic acid, pelargonic acid (nonanoic acid), etc.; an anionic sulfonate, including
30 dodecylbenzenesulfonic acid, and other acidic antimicrobials and mixtures thereof. Such admixtures provide superior cooperative antimicrobial effect. The combined agents provide an immediate, additive biocidal action to contagious mastitis causing

pathogens present on the teat when the composition is first applied. A continuous, longer term antimicrobial action is afforded by the non-volatile agents which become enveloped into the barrier film upon drying. Chlorine dioxide, formed in-situ by reacting ingredients within the composition, is present only during the initial application of treatment onto the teat and can off-gas during treatment dry-down. This effect is advantageous because the superior antimicrobial properties of chlorine dioxide are utilized to destroy the pathogens of greatest concern to the herdsman, i.e., the contagious mastitis causing organisms already on the teat and in the distal teat canal orifice; but, once applied, chlorine dioxide dissipates and thus removes the potential for severe teat skin irritation which otherwise might result from this very reactive chemical; and, for any incidental residuals in the producers milk. Heptanoic acid or nonanoic (pelargonic) acid, which are preferred antimicrobial agents within compositions of the present invention, and/or dodecylbenzenesulfonic acid, all being of non-volatile character, augment the biocidal performance of chlorine dioxide upon initial application; and, as treatment drying occurs with subsequent barrier formation, become resident within the prophylactic coating wherein the agents provide continuous and efficacious antimicrobial protection from mastitis causing environmental organisms. After the chlorine dioxide component, made in situ by the combination of the aqueous acidulant part with the aqueous chlorite part, becomes volatilized and is thus removed from the composition, the composition maintains extended antimicrobial activity and the composition maintains an effective barrier, including a milk duct plug, to environmental pathogens and soils.

Surprising and unexpected long term chlorine dioxide residence and chemical stability results in practice which is in distinct contrast to commercial embodiments of prior art, specifically and particularly the commercial antimicrobial teat dip sold under the name UDDER-GOLD PLUS (Alcide Corp. Redmond, WA), having affixed use instructions which advise the user to blend, in equal proportions, only sufficient admixture of cooperative parts (UDDER-GOLD PLUS BASE and UDDER-GOLD PLUS ACTIVATOR) for one herd milking - discarding any remainder.

We have discovered that preferred mastitis treatment compositions of this invention, once having formed chlorine dioxide in-situ from chemical reactants

blended together by the herdsman in preassigned proportion, retain this antimicrobial agent and associated antimicrobial property within the admixed teat treatment for extended periods of time. A typical useful application life span of such product being approximately one month following preparatory blending. We
5 believe this unusual chlorine dioxide stability is an additional consequence of the rheological properties of the preferred compositions whereby the chlorine dioxide gas is entrapped and held homogeneously dispersed throughout the product. Such attribute has many practical advantages for the herdsman including convenience of preblending large mixtures of teat treatment and eliminating need for repeatedly
10 preparing the exact quantity applied at each milking, reducing wasteful and costly disposal of unused, but unstable product which cannot be saved for next milking; and, enhanced safety for the user who would otherwise be exposed to off-gassing chlorine dioxide fumes during the milking process.

The compositions of the invention comprise an admixture comprised of two
15 cooperative parts: The first part, of major or equal proportion, having, in aqueous liquid mixture, an acid component and the plurality of components used in the mastitis control compositions of this invention and including all ingredients of organic structure including rheology modifiers and thickeners; emollients, humectants, conditioners and medicaments; surfactants and hydrotropes;
20 antimicrobial agents and preservatives; buffers, acidulants; chromophores and the like: The second part, of equal or minor proportion, being a chlorous acid or salt thereof, more specifically, an alkali metal chlorite: being of aqueous liquid, particulate-powder, or compressed or cast molten solid form; and, generally in practice, added to the first part of the admixture. Further, the aforementioned second
25 part must, if liquid, be readily miscible or, if solid, be easily soluble within the first part to effect rapid homogeneous blending which is of particular importance when large quantities of this admixture are prepared on site by the herdsman. Experience has taught that favorable admixture blending is best accomplished by liquids of the second part having no ordered structure, i.e. no thickened or gelled character; and,
30 by solids of the second part having large surface area. In practice, this means liquids having fluid characteristics similar to water, and solids having particulate form.

The primary function of the second part is to carry the chlorine dioxide release agent into the admixture of this mastitis control treatment; however, lesser adjuvants may be included within, e.g., alkali metal carbonate salts added to commercial chlorite solutions to improve stability. Such lesser adjuvants must not alter, to any appreciable degree, rheology properties of the second part other than characteristic of the aqueous carrier itself. Once joined, the alkaline chlorine (III)/chlorite composition of the second part is blended into the acidic solution of the first part with resulting admixture having a buffered pH of approximately 3.0; whereupon and by chemical reaction well known in the scientific art and literature, the disproportionation of chlorine (III)/chlorite occurs with a measured rate of formation of chlorine dioxide. Because this reaction begins immediately and because only very small quantities of chlorine dioxide are required for pronounced microbiocidal effect, the thus prepared mastitis control treatment is ready for application as soon as needed.

Commercial compositions of the invention are held within a user friendly dual or "duet" packaging combination designed to contain and transport parts A and B of these compositions together in predetermined and premeasured proportions and to be cooperative in the blending process wherein the user pours or otherwise causes the entire contents of package B, an equal or minor proportion, to be discharged into package A, an equal or major proportion, which then becomes the container and holding device for the admixture or final mastitis control treatment.

In summary, a first aspect of the invention provides for a physically and chemically stable mastitis treating composition, in an acidulant part and a chlorite part, that can effectively reduce microbial populations, on contact and for an extended period, said composition comprising: (a) an acidulant part comprising organic ingredients, said ingredients comprising: (i) about 0.1 to 15 parts by weight of a C_{1-6} organic monocarboxylic acid or dicarboxylic acid or mixtures thereof; (ii) about 0.1 to 15 parts by weight of an antimicrobial acid comprising a C_{6-12} carboxylic acid or salt thereof or a hydrocarbon sulfonic acid or salt thereof; (iii) about 0.1 to 20 parts by weight of an organic polymeric thickener: an organic polymeric film-forming agent or mixtures thereof; and (iv) a major proportion of water; (b) a chlorite part, substantially free of an organic component, consisting essentially of an alkali metal chlorite salt; wherein the

12a

composition exhibits rheology that promotes cling to immobilize the composition on the target surface and provides a barrier to environmental contamination.

A second aspect of the invention provides for a method of forming a mastitis treatment composition which comprises combining: (a) an acidulant part comprising organic ingredients, said ingredients comprising: (i) about 0.1 to 15 parts by weight of an C_{1-6} organic monocarboxylic acid or mixtures thereof; (ii) about 0.1 to 15 parts by weight of an antimicrobial acid comprising a C_{6-12} carboxylic acid or salt thereof; (iii) about 0.1 to 20 parts by weight of an organic polymeric thickener, an organic polymeric film-forming agent or mixtures thereof; and (iv) a major proportion of water; (b) a chlorite part substantially free of an organic component, consisting essentially of an alkali metal chlorite salt; to form an aqueous thickened teat dip material containing less than about 1500 parts per million ClO_2 and less than about 3900 parts per million oxygenated chlorine species sanitizing component: wherein the composition exhibits rheology that promotes cling to immobilize the composition on the surface of a dairy animal teat and provides a barrier to environmental contamination.

Brief Discussion of the Drawings

Figures 1A, 1B, 1C, 1D, 2A and 2B are graphical representations of the test data and comparisons shown in the tables of data in the application.

Figure 1A is a four week biocidal efficacy analysis of Example II using the food contact sanitizing protocol. Figure 1B is a four week biocidal efficacy analysis of Example II using the food contact sanitizing protocol with a 10% milk challenge. Figures 1C and 1D detail four week biocidal efficacy and analyses of the UDDER GOLD PLUS formulation obtained under similar conditions as the data in Figs. 1A and 1B respectively.

Figure 2A is a graph showing the four week efficacy analysis of the UDDER GOLD PLUS formulation using the porcine skin test protocol. Figure 2B is a four week efficacy analysis of Example II using the porcine skin test protocol.

Figure 3 is a graph showing the change in viscosity with changing shear of two exemplary teat dips, Examples II and IX, of the application compared to the UDDER GOLD PLUS formulation.

Detailed Description of the Invention

Components Used in the Mastitis Control Compositions of this Invention

10 The present invention may generally comprise in a mastitis control and prevention treatment composition a carrier, an acidulant or admixture, an antimicrobial agent or admixture, a rheology modifier or admixture, a film-forming agent or admixture, a buffer system, a hydrotrope or admixture, an emollient or admixture,

15 a surfactant or surfactant admixture, a chromophore or colorant, and optional adjuvants. The preferred compositions of this invention comprise ingredients which are generally regarded as safe, and are not of themselves or in admixture incompatible with milk or milk by-products. Likewise, ingredients may be selected for any given composition which are cooperative in their combined effects whether

20 incorporated for antimicrobial efficacy, physical integrity of the formulation or to facilitate healing and the health of the teat. Generally, the composition comprises a carrier which functions to dilute the active ingredients and facilitates application to the intended surface. The carrier is generally an aqueous medium such as water, or an organic liquid such as an oil, a surfactant, an alcohol, an ester, an ether, or an

25 organic or aqueous mixture of any of these. Water is preferred as a carrier or diluent in compositions of this invention because of its universal availability and unquestionable economic advantages over other liquid diluents.

Acidulants are necessary ingredients within the mastitis control treatments of the invention to maintain the appropriate pH for dissociation of the chlorite/chlorine

30 dioxide release agent and to prevent dissociation of heptanoic, octanoic, nonanoic, decanoic and undecanoic carboxylic acids employed as non-fugitive antimicrobial agents. Carboxylic acids become increasingly biocidal as the pH falls below their

pK_a value; consequently, for the carboxylic acids mentioned above, a pH ranging from about 2.5 to 5.5, preferably from about 2.5 to 4.5 and most preferably from about 2.5 to 3.5 is desirable. The acidic component used to prepare the acidic teat dip compositions of the invention will comprise a weak inorganic acid or a weak organic acid which can be dissolved in the aqueous system of the invention to produce an acidic pH. A pH substantially less than about 1 can result in substantial irritation, while a pH greater than about 5 can unacceptably reduce the efficiency of the composition. The term "weak" as used in reference to an acidic component is intended to refer to an acid in which the first dissociation step does not proceed essentially to completion when the acid is dissolved in water at ambient temperatures at a concentration within a range useful to form the present compositions. Such inorganic and organic acids are also referred to as weak electrolytes as the term is used in Textbook of Quantitative Inorganic Analysis, I. M. Kolthoff et al., eds., The Macmillan Co. (3d ed., 1952) at pages 34-37.

Most common commercially-available weak inorganic and organic acids can be used in the invention. Preferred weak inorganic acids include phosphoric acid and sulfamic acid. Useful weak organic acids include acetic acid, hydroxyacetic acid, citric acid, tartaric acid and the like. Acidulants found useful include organic and inorganic acids such as citric acid, lactic acid, acetic acid, glycolic acid, adipic acid, tartaric acid, succinic acid, propionic acid, malic acid, alkane sulfonic acids, cycloalkane sulfonic acids, as well as phosphoric acid and the like or mixtures thereof. Preferred acidulants are those commonly referred to as C₂₋₆ alpha-hydroxycarboxylic acids, that group of acids which contain a hydroxy function in the alpha position directly adjacent to the carbon atom bearing the carboxyl function, examples of alpha-hydroxymonocarboxylic acids being glycolic, lactic and hydroxybutanoic acid; and, examples of hydroxydicarboxylic acids being malic and tartaric acids. We have found a surprising interaction between the acidulant material and a second antimicrobial acid composition. Preferably, the acidulant material comprises the C₂₋₆ alpha-hydroxy carboxylic acid in combination with a secondary antimicrobial acid composition. The second antimicrobial acid composition can comprise a C₆₋₁₂ carboxylic acid or a hydrocarbon sulfonic acid composition. These

materials work together to provide a cooperative antimicrobial action which effects initial kill from chlorine dioxide contributed by the acidulated chlorite and a long lasting kill in the barrier layer from the carboxylic acid/sulfonic acid material. This cooperation of ingredients is an important aspect of the invention.

5 Used in personal care products, alpha-hydroxycarboxylic acids absorb moisture from the atmosphere and therefore, when applied topically, increase moisture content and plasticity of the stratum corneum. They have had significant impact on skin treatment due to their ability to reduce corneocyte adhesion and accelerate cell proliferation within the basal layers. Though mechanism of action is
10 not yet fully understood, alpha-hydroxycarboxylic also are thought to stimulate synthesis of collagen and mucopolysaccharides in the dermis. At use levels under 10%, skin care benefits are derived through a continued pattern of product usage. Continued use of products with alpha-hydroxycarboxylic acids levels below 10% has been shown to result in gradual reduction of fine lines and an improvement in
15 skin texture through accelerated desquamation. Although conjecture at present, it is believed that some, if not all of these advantages may also be transferred upon the bovine teat skin. By incorporating an alpha-hydroxycarboxylic acid, healing may be accelerated; and, by "smoothing" the dermal surface, cleaning and asepsis may be improved. The most preferred alpha- hydroxycarboxylic acid for compositions of
20 this invention is lactic acid.

Numerous inorganic and organic antimicrobial agents may be utilized in teat dip compositions including (but not limited to) chlorine and bromine release compounds (e.g. alkali and alkaline earth hypochlorites and hypobromites, isocyanurates, chlorinated derivatives of hydantoin, sulfamide, amine, etc.), iodine
25 release complexes of surfactants or polymers such as polyvinylpyrrolidone (termed iodophors), quaternary ammonium compounds, chlorhexidine salts, peroxide and peroxyacid compounds, protonated short chain carboxylic acids, acidified anionic surfactants and chlorine dioxide. Of these typically applied antimicrobial agents which have been investigated for control of bovine mastitis, protonated short chain
30 (C_{6-12})carboxylic acids, acidified alkylaryl sulfonates and chlorine dioxide are proven efficacious against mastitis causing microorganisms; and, are preferred in compositions of the present invention. More specifically, dodecylbenzene sulfonic

acid, protonated C₆₋₁₂ carboxylic acids and chlorine dioxide are especially preferred antimicrobial agents.

The composition of the invention may also contain one or more rheology modifiers, to enhance viscosity, or thicken and cause the aqueous treatment to cling
5 to the surface skin of the teat. Clinging enables the composition to remain in contact with transient and resident pathogenic bacteria for longer periods of time, promoting microbiological efficacy and resisting waste because of excessive dripping. The rheology modifier may be a film former or act cooperatively with a film-forming agent to form a barrier that provides additional protection. Water soluble or water
10 dispersible rheology modifiers that are useful can be classified as inorganic or organic. The organic thickeners can further be divided into natural and synthetic polymers with the latter still further subdivided into synthetic natural-based and synthetic petroleum-based.

Inorganic thickeners are generally compounds such as colloidal magnesium
15 aluminum silicate (VEEGUM[®]), colloidal clays (Bentonites), or silicas (CAB-O-SILS[®]) which have been fumed or precipitated to create particles with large surface to size ratios. Natural hydrogel thickeners of use are primarily vegetable derived exudates. For example, tragacanth, karaya, and acacia gums; and extractives such as caragheenan, locust bean gum, guar gum and pectin; or, pure culture fermentation
20 products such as xanthan gum are all potentially useful in the invention. Chemically, all of these materials are salts of complex anionic polysaccharides. Synthetic natural-based thickeners having application are cellulosic derivatives wherein the free hydroxyl groups on the linear anhydro-glucose polymers have been etherified or esterified to give a family of substances which dissolve in water and
25 give viscous solutions. This group of materials includes the alkyl and hydroxylalkylcelluloses, specifically methylcellulose, hydroxyethylmethylcellulose, hydroxypropylmethylcellulose, hydroxybutylmethylcellulose, hydroxyethylcellulose, ethylhydroxyethylcellulose, hydroxypropylcellulose, and carboxymethylcellulose. Synthetic petroleum-based water soluble polymers are prepared by direct
30 polymerization of suitable monomers of which polyvinylpyrrolidone, polyvinylmethylether, polyacrylic acid and polymethacrylic acid, polyacrylamide, polyethylene oxide, and polyethyleneimine are representative.

All thickeners do not work with equal effectiveness in this invention. Preferred aqueous thickening agents which are more useful in this invention are those which are extremely pseudoplastic (non-Newtonian, rapid relaxation), tend not to develop a rigid three-dimensional structure from interpolymer interactions, have a low or negligible viscoelastic character and possess a high gel strength. Such rheological properties are manifested in a teat dip composition which has a smooth flowing appearance, is easy to pour and apply onto the teat, coats uniformly without forming muscilage streamers as the applicator is withdrawn and remains firmly in place without significant sag. Examples of preferred rheology modifiers are xanthan gum and the hydroxylalkylcelluloses. Generally, the concentration of thickener used in the present invention will be dictated by the final composition any by the method of teat application. Spraying or misting requires a lower composition viscosity for easy and effective application of treatment than dipping. Film-forming barrier dips typically require high apparent viscosity necessary to form thick coatings on teats which insures improved prophylactic effect.

For compositions of this invention designed to provide a barrier for prophylactic protection, additional film-forming agents are included which typically work in conjunction with thickeners. In fact, many of the aforementioned rheology modifiers are themselves film formers of greater or lesser effectiveness; however, a preferred grade of polyvinyl alcohol when used with preferred thickeners such as xanthan gum or hydroxyalkylcelluloses affords particularly useful properties to compositions of this teaching, most notably the development of "balanced" films on treated teats which are sufficiently water-sensitive to be stripped off with conventional udder washing, but capably adherent to the teat skin to withstand premature loss of integrity between milkings and intrinsically resistant to environmental exposure; and, in addition, are of such structure as to successfully occlude antimicrobial agents within the film matrix for continuing biocidal effect against mastitis causing organisms. The success of the barriers thus formed by compositions of this invention are, in part, a consequence of a hydrophobic-hydrophilic balance, caused when non-volatile ingredients, especially fatty acids, surfactants and hydrotropes, become resident throughout the film and whose

individual properties become additive with those characteristics of the thickeners and film formers. Such inclusions also plasticize the film and render it pliable.

Polyvinyl alcohol compositions can be used as a film former. Variation of film flexibility, water sensitivity, ease of solvation, viscosity, film strength and
5 adhesion can be varied by adjusting molecular weight and degree of hydrolysis. The preferred polyvinyl alcohol for use in compositions herein has a degree of hydrolysis greater than 92%, preferably greater than 98%, most preferably greater than 98.5%; and, has a molecular weight (Mn) that falls in the range of between about 15,000 and 100,000, but preferably between 40,000 and 70,000 corresponding to a solution
10 viscosity (4% wt aqueous solution measured in centipoise (cP) at 20°C by Hoeppler falling ball method) of 12-55 cP and 12-25 cP respectively.

The classical definition of a buffered solution is one containing both a weak acid and its conjugate weak base, whose pH changes only slightly on addition of acid or alkali. The weak acid becomes a buffer when alkali is added, and the weak
15 base becomes a buffer when acid is added. Maintenance of the pH of compositions described in the present invention is necessary to minimize undesirable chemical changes which may inhibit the microbiological efficacy of the antimicrobial agent or cause toxic or irritating effect upon the teat. Any compatible organic or inorganic material or mixture of materials which has the desired effect of maintaining the
20 composition pH within prescribed ranges can be utilized as the buffering agent or system in the instant invention. Of primary concern are pH shifts caused by naturally occurring chemicals brought into the composition, after application onto the teat, by skin exudations, milk or environmental soils; and, pH drifting which sometimes accompanies chemical equilibriums established within compositions as
25 ingredients are changed or concentrations varied.

In general, the pH of bovine mastitis control treatments can vary from a low of about pH 2.0 to a maximum of approximately 11.0 depending primarily upon the choice of antimicrobial agent being incorporated in the composition because optimal efficacy normally occurs with a specific, narrow, pH range. Therefore the buffering
30 agent or system is chosen accordingly. The preferred pH range of compositions of this invention is typically from 2.5 to 5.5 most preferably, about 2.5 to 3.5 - the lower value being a limit to prevent excessive irritation on the teat surface; and, the

upper limit set to enhance chlorine dioxide formation and maintain antimicrobial effect of the protonated carboxylic acid(s) and/or acidified anionic surfactant. A typical and preferred buffer system would be citric acid and its alkali metal salt. However, any acidulant and corresponding conjugate weak base could be used.

5 Solubilizing agents called hydrotropes or couplers may be generally used in compositions of the present invention to maintain physical single phase integrity and storage stability. To this end, any number of ingredients known to those skilled in formulation art may be employed, such as monofunctional and polyfunctional alcohols. These preferably contain from about 1 to about 6 carbon atoms and from 1
10 to about 6 hydroxy groups. Examples include ethanol, isopropanol, n-propanol, 1, 2-propanediol, 1, 2-butanediol, 2-methyl-2, 4-pentanediol, mannitol and glucose. Also useful are the higher glycols, polyglycols, polyoxides, glycol ethers and propylene glycol ethers. Additional useful hydrotropes include the free acids and alkali metal salts of sulfonated alkylaryls such as toluene, xylene, cumene and
15 phenol or phenol ether or diphenyl ether sulfonates; alkyl and dialkyl naphthalene sulfonates and alkoxylated derivatives. The most preferred hydrotrope for the most preferred embodiments of this invention is 1-octane sulfonate or mixtures of 1-octane sulfonate and 1,2-octane disulfonate manufactured and held for proprietary use under the name NAS by Ecolab Inc, St. Paul, MN.

20 Teat dip compositions of the present invention generally also comprise an emollient and/or humectant to lubricate, condition and generally reduce and promote the healing of irritation on the teat surface of application which may result either from the antimicrobial agent, from the mechanical action of the milking machine or from environmental conditions such as wind chill, dehydration, abrasion and
25 sunburn. Any water soluble or dispersible skin conditioning agent may be used in this present invention. Compositions such as polyhydric alcohols are useful in the invention including glycerin, sorbitol, mannitol, and propylene glycol and its homopolymers; fatty acid esters of simple monohydric alcohols including isopropyl palmitate or isopropyl myristate and similar esters; polyol esters of fatty acids; and,
30 ethoxylated lanolins, vegetable oils, and similar natural sourced derivatives such as aloe. Preferred emollients to be used in the invention include glycerin, sorbitol, and propylene glycol.

The surfactant or surfactant admixture of the present invention can be selected from compatible water soluble or water dispersible nonionic, or anionic surface-active agents; or mixtures of each or both types. Nonionic and anionic surfactants offer diverse and comprehensive commercial selection, low price; and, most important, excellent deterative effect - meaning surface wetting. Surface - active or "wetting agents" function to increase the penetrant activity of the invention into the tissue surface at risk from mastitis causing pathogens. Nonionic surfactants useful in the invention are generally characterized by the presence of an organic hydrophobic group and an organic hydrophilic group and are typically produced by the condensation of an organic aliphatic, alkyl aromatic or polyoxyalkylene hydrophobic compound with a hydrophilic alkaline oxide moiety which in common practice is ethylene oxide or a polyhydration product thereof, polyethylene glycol. Practically any hydrophobic compound having a hydroxyl, carboxyl, amino, or amido group with a reactive hydrogen atom can be condensed with ethylene oxide, or its polydration adducts, or its mixtures with alkoxylenes such as propylene oxide to form a nonionic surface-active agent. The length of the hydrophilic polyoxyalkylene moiety which is condensed with any particular hydrophobic compound can be readily adjusted to yield a water dispersible or water soluble compound having the desired degree of balance between hydrophilic and hydrophobic properties.

Useful nonionic surfactants in the present invention include: Block polyoxypropylene-polyoxyethylene polymeric compounds based upon propylene glycol, ethylene glycol, glycerol, trimethylolpropane, and ethylenediamine as the initiator reactive hydrogen compound. Examples of polymeric compounds made from a sequential propoxylation and ethoxylation of initiator are commercially available under the trade name PLURONIC[®] manufactured by BASF Corp. PLURONIC[®] compounds are difunctional (two reactive hydrogens) compounds formed by condensing ethylene oxide with a hydrophobic base formed by the addition of propylene oxide to two hydroxyl groups of propylene glycol. This hydrophobic portion of the molecule weighs from about 1,000 to about 4,000. Ethylene oxide is then added to sandwich this hydrophobe between hydrophilic groups, controlled by length to constitute from about 10% by

weight to about 80% by weight of the final molecule. TETRONIC® compounds are tetra-functional block copolymers derived from the sequential additional of propylene oxide and ethylene oxide to ethylenediamine. The molecular weight of the propylene oxide hydrotype ranges from about 500 to about 7,000; and, the hydrophile, ethylene oxide, is added to constitute from about 10% by weight to about 80% by weight of the molecule.

Also useful nonionic surfactants include the condensation products of one mole of alkyl phenol wherein the alkyl constituent, contains from about 8 to about 18 carbon atoms with from about 3 to about 50 moles of ethylene oxide. The alkyl group can, for example, be represented by diisobutylene, di-amyl, polymerized propylene, isoctyl, nonyl, and di-nonyl. Examples of commercial compounds of this chemistry are available on the market under the trade name IGEPAL® manufactured by Rhone-Poulenc and TRITON® manufactured by Union Carbide.

Likewise useful nonionic surfactants include condensation products of one mole of a saturated or unsaturated, straight or branched chain alcohol having from about 6 to about 24 carbon atoms with from about 3 to about 50 moles of ethylene oxide. The alcohol moiety can consist of mixtures of alcohols in the above delineated carbon range or it can consist of an alcohol having a specific number of carbon atoms within this range. Examples of like commercial surfactant are available under the trade name NEODOL® manufactured by Shell Chemical Co. and ALFONIC® manufactured by Vista Chemical Co.

Condensation products of one mole of saturated or unsaturated, straight or branched chain carboxylic acid having from about 8 to about 18 carbon atoms with from about 6 to about 50 moles of ethylene oxide. The acid moiety can consist of mixtures of acids in the above delineated carbon atoms range or it can consist of an acid having a specific number of carbon atoms within the range. Examples of commercial compounds of this chemistry are available on the market under the trade name NOPALCOL® manufactured by Henkel Corporation and LIPOPEG® manufactured by Lipo Chemicals, Inc. In addition to ethoxylated carboxylic acids, commonly called polyethylene glycol esters, other alkanoic acid esters formed by reaction with glycerides, glycerin, and polyhydric (saccharide or sorbitan/sorbitol) alcohols have application in this invention. All of these ester moieties have one or

more reactive hydrogen sites on their molecule which can undergo further acylation or ethylene oxide (alkoxide) addition to control the hydrophilicity of these substances.

Other useful surfactants are nonionics made by adding ethylene oxide to ethylene glycol to provide a hydrophile of designated molecular weight; and, then adding propylene oxide to obtain hydrophobic blocks on the outside (ends) of the molecule. The hydrophobic portion of the molecule weighs from about 1,000 to about 3,100 with the central hydrophile comprising 10% by weight to about 80% by weight of the final molecule. These "reverse" PLURONIC®'s are manufactured by the BASF Corporation under the trade name PLURONIC® surfactants. Likewise, the TETRONIC® surfactants are produced by the BASF Corporation by the sequential addition of ethylene oxide and propylene oxide to ethylenediamine. The hydrophobic portion of the molecule weighs from about 2,100 to about 6,700 with the central hydrophile comprising 10% by weight to 80% by weight of the final molecule.

Tertiary amine oxides corresponding to the general formula:



can be used wherein the $\text{ } \text{ } \text{O}$ bond is a conventional representation of a semi-polar bond; and R^1 , R^2 , and R^3 may be aliphatic, aromatic, heterocyclic, alicyclic groups or a combination of such groups thereof. Generally, for amine oxides of detergent interest, R^1 is an alkyl radical of from about 8 to about 24 carbon atoms; R^2 and R^3 are selected from the group consisting of alkyl or hydroxyalkyl of 1-3 carbon atoms and mixtures thereof; R^4 is an alkylene or a hydroxyalkylene group containing 2 to 3 carbon atoms; and n ranges from 0 to about 20. Useful water soluble amine oxide surfactants are selected from the coconut or tallow dimethyl amine oxides.

Also useful in the present invention are surface active substances which are categorized as anionics because the charge on the hydrophobe is negative; or surfactants in which the hydrophobic section of the molecule carries no charge

unless the pH is elevated to neutrality or above (e.g. carboxylic acids). Carboxylate, sulfonate, sulfate and phosphate are the polar (hydrophilic) solubilizing groups found in anionic surfactants. Of the cations (counterions) associated with these polar groups, sodium, lithium and potassium impart water solubility and are most preferred in compositions of the present invention. Examples of suitable synthetic, water soluble anionic compounds are the alkali metal (such as sodium, lithium and potassium) salts or the alkyl mononuclear aromatic sulfonates such as the alkyl benzene sulfonates containing from about 5 to about 18 carbon atoms in the alkyl group in a straight or branched chain, e.g., the salts of alkyl benzene sulfonates or of alkyl naphthalene sulfonate, dialkyl naphthalene sulfonate and alkoxylated derivatives. Other anionic detergents are the olefin sulfonates, including long chain alkene sulfonates, long chain hydroxyalkane sulfonates or mixtures of alkenesulfonates and hydroxyalkane-sulfonates and alkylpoly (ethyleneoxy) ether sulfonates. Also included are the alkyl sulfates, alkyl poly (ethyleneoxy) ether sulfates and aromatic poly (ethyleneoxy) sulfates such as the sulfates or condensation products of ethylene oxide and nonyl phenol (usually having 1 to 6 oxyethylene groups per molecule).

Complexed iodines offer the advantage of being chromophoric, i.e. easily visible when applied onto the teat. Other antimicrobial agents do not have this feature; therefore, compositions of this invention may include a water soluble or dispersible coloring agent (dye or pigment or mixtures) which renders the composition chromophoric, having sharp contrast to teat skin and permitting the dairy herd manager to visually discern that the teats have been treated.

Alternatively, the compositions of the invention may be comprised of any number of optional ingredients, i.e. adjuvants. Depending upon the benefits provided, adjuvants may partially or wholly displace the carrier in the composition. Generally, in accordance with the invention, there may be included within this composition formulary adjuvants which assist in the application of the invention with respect to physical and chemical stability, barrier film formation, teat health maintenance, performance, physical form and manufacturing process anesthetics. Of course, these functions may be accomplished exclusively by composition ingredients already described or admixtures thereof; however, formulary or application or

performance situations may occur requiring additional effect which may be accomplished by introducing an additional inorganic or organic agent or agents and mixtures thereof into the composition.

The compositions of the invention may optionally include medicaments, for example sunscreens such as paraamino benzoic acid and healing agents such as allantoin or urea to provide curative action and stimulation of formation of new tissue; preservatives such as methyl paraben, propyl paraben, sorbic and benzoic acids or salts thereof to retard bacterial growth and prolong shelf life; antioxidants such as BHT (butylated hydroxytoluene), BHA (butylated hydroxyanisole), TBHQ (tert-butylhydroquinone), or propyl gallate to retard oxidative or hydrolytic degradation; sequestering agents such as aminopolyacetates, polyphosphonates, aminopolyphosphonates, polycarboxylates, and condensed phosphates; dispersants or suspending agents having polyelectrolytic character such as polyacrylate and similar polycarboxylates of homopolymeric or copolymeric structure; and manufacturing processing agents, for example defoam additives employed to facilitate blending and mixing.

A wide variety of ingredients useful in mastitis control treatment can be included in the compositions hereof. This list is not intended to be exhaustive and other optional ingredients, which may not be listed, but which are well known in the art, may also be utilized in the composition. The examples are not intended to be limited in any way. In certain cases, some of the individual adjuvants may overlap other categories. The adjuvants employed will be selected so as not to interfere with the antimicrobial action of the composition and to avoid physical or chemical instability of the product.

Table "Bovine Mastitis Treatment Admixture Compositions", below, provides guidelines for consistent concentrations in accordance with this invention.

<u>INGREDIENT</u>	<u>USEFUL AMOUNT (WT.%)</u>	<u>PREFERRED (WT.%)</u>	<u>MORE PREFERRED (WT.%)</u>
CARRIER	40.0 - 98.0	50.0 - 98.0	60.0 - 98.0
BLEND OF ACIDULANT AND/OR ANTI- MICROBIAL	1.0 - 12.0	1.0 - 10.0	1.0 - 8.0
RHEOLOGY MODIFIER	0.0 - 10.0	0.01 - 7.5	0.1 - 5.0
FILM FORMER	0.0 - 12.0	0.01 - 8.0	0.1 - 4.0
BUFFER	0.0 - 15.0	0.01 - 10.0	0.1 - 5.0
HYDROTROPE	0.0 - 20.0	0.0 - 15.0	0.1 - 10.0
EMOLLIENT	0.5 - 60.0	1.0 - 40.0	1.5 - 20.0
SURFACTANT	0.0 - 60.0	0.01 - 40.0	0.1 - 20.0
COLORANT	0.0 - 1.0	0.001 - 0.8	0.002 - 0.6
OPTIONAL ADJUVANT	0.0 - 5.0	0.1 - 4.0	0.1 - 3.0

The following examples and data are provided to illustrate preferred embodiments of the invention and contain a best mode.

5

Example I

Example I is a representative embodiment of this invention which illustrates a bovine mastitis treatment having barrier film properties. Antimicrobial effect is provided by generation of chlorine dioxide caused by disproportionation of chlorite ion upon admixture of composition base with activator and by the resident biocidal activity of protonated nonanoic acid.

10

A 16 kilogram batch of the following experimental base formula part and 1 kilogram of the ClO_2^- part was prepared by blending the ingredients as shown.

Base Formula (Part I) (pH = 2.8):

Ingredients	Wt%	Grams
Glycerin, 96%	5.00	800.05
Isopropanol, 99%	2.00	320.05
Nonanoic (Pelargonic) Acid	1.50	240.05
Lactic Acid, 88%	2.95	472.03
*Xanthan Gum (KELTROL [®] K5C151)	0.30	48.02
Deionized Water	60.76	9721.62
Potassium Benzoate	0.20	32.01
KOH, 45%	0.29	46.40
Octane Sulfonate	17.00	2720.02
**ELVANOL [®] Premix, 10%	10.00	1600.00
TOTAL	100.00	16000.25

5

* KELTROL[®] K5C151 is a grade of xanthan gum manufactured by Kelco.

** ELVANOL[®] Premix: 10% aqueous solution of ELVANOL[®] 90-50.

ELVANOL[®] 90-50 is a grade of polyvinyl alcohol manufactured by E. I. duPont.

10

Activator ClO_2^- Formula (Part II) (pH = 12.3):

Ingredients	Wt %	Grams
Deionized Water	50.00	500.00
Sodium Chlorite, 25%	50.00	500.00
TOTAL	100.00	1000.00

About 3376 grams of the base formula part were blended with about 92.89 grams of the ClO_2^- part (the ClO_2^- is 0.32% of total composition). The viscosity (Brookfield Model DV-II Viscometer, spindle No. 1, 20 rpms at 25°C) was about 304 cps.

15

Example II

20

Example II is a further example of this invention which differs from Example I in that the resident biocidal activity is contributed by dodecylbenzene sulfonic acid.

The hydrotropic agent, octane sulfonate, is not required in this formula for physical stability.

A seven kilogram batch of the following base formula part and a one kilogram batch of the ClO_2^- activator part was prepared.

5

Base Formula (Part II) (pH = 2.9)

Ingredients	%	Grams
Glycerin, 96%	5.00	350.10
Dodecylbenzene Sulfonic Acid, 97%	2.00	140.01
Xanthan Gum *KELTROL® K5C151	0.30	21.01
Deionized Water	78.14	5469.90
Lactic Acid, 88%	2.95	206.51
KOH, 45%	1.41	98.73
Potassium Benzoate	0.20	14.00
**ELVANOL™ Premix, 10%	10.00	700.06
TOTAL	100.00	7000.32

-
- * KELTROL® K5C151 is a grade of xanthan gum manufactured by Kelco.
 ** ELVANOL™ Premix: 10% aqueous solution of ELVANOL® 90-50.
 ELVANOL® 90-50 is a grade of polyvinyl alcohol manufactured by E. I. duPont.

10

Activator ClO_2^- Formula (Part II) (pH = 12.3):

Ingredients	%	Grams
Deionized Water	50.00	500.00
Sodium Chlorite, 25%	50.00	500.00
TOTAL	100.00	1000.00

About 3376 grams of the base formula part were mixed with about 92.98 grams of the ClO_2^- activator part. The stable rheology and pH of the combined formula is shown as follows:

Viscosity:	As Mixed	pH
	Brookfield (#1, 20 rpm @ 25°C)	
Brookfield: initial	290 cps	3.08
Brookfield: 1 week	296 cps	3.09
Brookfield: 2 weeks	n/a	n/a
Brookfield: 3 weeks	291 cps	2.97
Brookfield: 4 weeks	294 cps	3.10

5

Example III

Example III is a representative composition of the invention illustrating a bovine mastitis treatment without a barrier film property and having no thickening nor film-forming agents. A surfactant, NEODOL[®] 25-9, is included for teat cleansing and surface wetting.

10

A two hundred gram batch of the following experimental base formula and a 1 kilogram batch of the ClO_2^- activator part was made.

Base Formula (Part I) (pH = 2.7):

Ingredients	Wt%	Grams
Glycerin, 96%	5.00	10.0
C ₁₂₋₁₅ alcohol (9 mole) ethoxylate NEODOL [®] 25-9	0.50	1.00
Pelargonic Acid	0.50	1.00
Lactic Acid, 88%	2.95	5.90
Deionized Water	75.44	150.88
Octane Sulfonate	7.00	14.00
KOH, 45%	0.61	1.22
Dye -F, D & C Green #3-1.0% aq. active	4.00	8.00
*Pigment, 5.0%	4.00	8.00
TOTAL	100.00	200.00

15

* Pigment: PYLAKLOR[®] Yellow LX-10192 and Permanent Green S-722 (50:50 blend) manufactured by Pylam Products Co. Inc.

29

Activator ClO_2^- Formula (Part II) (pH = 12.3):

Ingredients	Wt%	Grams
Deionized Water	50.00	500.00
Sodium Chlorite, 25%	50.00	500.00
TOTAL	100.00	1000.00

About 200 grams of the base formula were mixed with about 5.5 grams of the ClO_2^- activator part. pH of final mixture is about 2.9.

Example IV

Example IV is a further modification of Example III using sorbitol as an emollient in place of glycerin. A two hundred gram batch of the following experimental base formula and a 1 kilogram batch of the ClO_2^- activator part was made.

Base Formula (Part I) (pH = 2.7):

Ingredients	Wt%	Grams
Sorbitol, 70%	1.00	2.00
C_{12-15} alcohol 9 mole ethoxylate NEODOL [®] 25-9	0.50	1.00
Pelargonic Acid	0.50	1.00
Lactic Acid, 88%	2.95	5.90
Deionized Water	79.49	158.98
Octane Sulfonate	7.00	14.00
KOH, 45%	0.56	1.12
Dye-F, D & C Green #3, 1.0% aqueous active	4.00	8.00
*Pigment, 5.0%	4.00	8.00
TOTAL	100.00	200.00

* Pigment: PYLAKLOR[®] Yellow LX-10192 and Permanent Green S-722 (50:50 blend) manufactured by Pylam Products Co. Inc.

Activator ClO_2^- Formula (Part II) (pH = 12.3):

Ingredients	Wt%	Grams
Deionized Water	50.00	500.00
Sodium Chlorite, 25%	50.00	500.00
TOTAL	100.00	1000.00

- 5 About 200 grams of the base formula were mixed with about 5.5 grams of the ClO_2^- activator part. pH of final mixture is about 2.9.

Example V

- Example V is a further composition of the invention illustrating a bovine mastitis treatment without a barrier film property, again having no thickening nor film-forming agents and using phosphoric acid as the acidulant. A surfactant, NEODOL[®] 25-9, is included for teat cleansing and surface wetting. A two hundred gram batch of the following experimental base formula and a 1 kilogram batch of the ClO_2^- activator part was made.

Base Formula (Part I) (pH = 2.7):

15

Ingredients	%	Grams
Sorbitol, 70%	1.00	2.00
C ₁₂₋₁₅ alcohol 9 mole ethoxylate - NEODOL [®] 25-9	0.50	1.00
Pelargonic Acid	0.50	1.00
Phosphoric Acid, 75%	1.00	2.00
Deionized Water	81.32	162.64
Octane Sulfonate	7.00	14.00
KOH, 45%	0.68	1.36
Dye-F, D & C Green #3, 1.0%	4.00	8.00
*Pigment 5.0%	4.00	8.00
TOTAL	100.00	200.00

* Pigment: PYLAKLOR[®] Yellow LX-10192 and Permanent Green S-722 (50:50 blend) manufactured by Pylam Products Co. Inc.

Activator ClO_2^- Formula (Part II) (pH = 12.3):

Ingredients	%	Grams
Deionized Water	50.00	500.00
Sodium Chlorite, 25%	50.00	500.00
TOTAL	100.00	1000.00

- 5 About 200 grams of the base formula were mixed with about 5.5 grams of the ClO_2^- activator part. pH of final mixture is about 2.9.

Example VI

- 10 Examples VI, VII and VIII are compositional variations of Example I which contain the homologous carboxylic acids octanoic, decanoic and a mixture thereof respectively.

A two hundred gram batch of the following experimental base formula and a 1 kilogram batch of the ClO_2^- activator part was prepared.

Base Formula (Part I) (pH = 2.7):

Ingredients	Wt%	Grams
Glycerin, 96%	5.00	10.05
Isopropanol, 99%	2.00	4.01
Octanoic Acid	1.50	3.02
Lactic Acid, 88%	2.95	5.90
Xanthan Gum KELTROL [®] K5C151	0.30	0.61
Deionized Water	60.93	121.90
Potassium Benzoate	0.20	0.40
KOH, 40%	0.12	0.25
Octane Sulfonate	17.00	34.02
*ELVANOL [®] Premix, 10%	10.00	20.09
TOTAL	100.00	200.25

15

* ELVANOL[®] Premix: 10% aqueous solution of ELVANOL[®] 90-50.
ELVANOL[®] 90-50 is a grade of polyvinyl alcohol manufactured by E. I. duPont.

32

Activator ClO_2^- Formula (Part 2) (pH = 12.3):

Ingredients	Wt%	Grams
Deionized Water	50.00	500.00
Sodium Chlorite, 25%	50.00	500.00
TOTAL	100.00	1000.00

5 The mixed product made with 100 grams of Part 1 formula and 2.75 grams of Part 2 is about pH 2.9.

Example VII

A 200 gram batch of the following experimental base formula and a 1 kilogram batch of the ClO_2^- activator part was prepared.

10

Base Formula (Part 1) (pH = 2.6):

Ingredients	Wt%	Grams
Glycerin, 96%	5.00	10.05
Isopropanol, 99%	2.00	4.00
Decanoic Acid	1.50	3.03
Lactic Acid, 88%	2.95	5.90
Xanthan Gum KELTROL® K5C151	0.30	0.61
Deionized Water	60.93	121.93
Potassium Benzoate	0.20	0.40
KOH, 40%	0.12	0.25
Octane Sulfonate	17.00	34.10
*ELVANOL® Premix, 10%	10.00	20.00
TOTAL	100.00	200.27

* ELVANOL® Premix: 10% aqueous solution of ELVANOL® 90-50.
ELVANOL® 90-50 is a grade of polyvinyl alcohol manufactured by E. I. duPont.

15

Activator ClO_2^- Formula (Part 2)(pH = 12.3):

Ingredients	Wt%	Grams
Deionized Water	50.00	500.00
Sodium Chlorite, 25%	50.00	500.00
TOTAL	100.00	1000.00

The mixed product made with 100 grams of the Base Part 1 Formula combined with 2.75 grams of the activator Part 2 ClO_2^- formula. The mixture is about pH 2.9.

Example VIII

- 5 A 200 gram batch of the following experimental base formula and a 1 kilogram batch of the ClO_2^- activator part was prepared.

Base Formula (Part 1) (pH = 2.7):

Ingredients	Wt%	Grams
Glycerin, 96%	5.00	10.04
Isopropanol, 99%	2.00	4.01
*KORTACID [®] - C8:C10 (3:1)	1.50	3.03
Lactic Acid, 88%	2.95	5.90
Xanthan Gum KELTROL [®] K5C151	0.30	0.61
Deionized Water	60.93	121.90
Potassium Benzoate	0.20	0.40
KOH, 40%	0.12	0.24
Octane Sulfonate	17.00	34.04
**ELVANOL [®] Premix, 10%	10.00	20.01
TOTAL	100.00	200.18

- 10 * KORTACID[®]: 3:1 Octanoic/decanoic blend manufactured by Akzo Chemical.
 ** ELVANOL[®] Premix: 10% aqueous solution of ELVANOL[®] 90-50.
 ELVANOL[®] 90-50 is a grade of polyvinyl alcohol manufactured by E. I. duPont.

Activator ClO_2^- Formula (Part 2) (pH = 12.3):

15

Ingredients	Wt%	Grams
Deionized Water	50.00	500.00
Sodium Chlorite, 25%	50.00	500.00
TOTAL	100.00	1000.00

The mixed product made using 100 grams of Part 1 and 2.75 grams of Part 2 is about pH 2.9.

34

Example IX

Example IX is an additional variation of Example I containing heptanoic acid and n-propanol in place of nonanoic and isopropanol respectively.

A 1000 gram batch of this experimental base formula was prepared; and,
 5 1000 grams of the ClO_2^- part was prepared by blending the ingredients as shown.
 Two typical admixtures of parts I and II were then prepared as illustrated.

Base Formula (Part I) (pH = 2.8):

Ingredients	Wt%	Grams
Glycerin, 96%	5.00	50.00
n-Propanol, 99%	1.50	15.03
Heptanoic Acid	1.00	10.03
Lactic Acid, 88%	2.95	29.51
KELTROL [®] K5C151	0.30	3.02
Deionized Water	71.13	711.38
Potassium Benzoate	0.20	2.03
KOH, 45%	0.42	4.21
Octane Sulfonate	7.50	75.00
ELVANOL [®] Premix, 10%	10.00	100.07
TOTAL	100.00	1000.28

Activator Formula (Part II) (pH = 12.0):

10

Ingredients	Wt%	Grams
Deionized Water	75.0	750
Sodium Chlorite, 25%	25.0	250
TOTAL	100.00	1000.00

Typical Mix Ratios:

Base (g)	Activator (g)
100	2.754 (6.25% active) → 0.16% NaClO_2
3376	93 (6.25% active) → 0.16% NaClO_2

15

Example X

A 500 gram batch of the following experimental base formula was prepared for preliminary testing. This example is similar to Example I with n-propanol and half of the NaClO_2 amount.

35

Base Formula (Part I) (pH = 2.8):

Ingredients	Wt%	Grams
Glycerin, 96%	5.00	25.00
n-Propanol, 99%	1.50	7.50
Pelargonic Acid	1.50	7.50
Lactic Acid, 88%	2.95	14.75
*KELTROL [®] K5C151	0.30	1.50
Deionized Water	71.13	266.30
Potassium Benzoate	0.20	1.00
KOH, 45%	0.29	1.25
Octane Sulfonate	17.00	85.00
**ELVANOL [®] Premix, 10%	10.00	50.00
***Pigment	8.00	40.00
TOTAL	100.00	500.00

* KELTROL[®] K5C151 is a grade of xanthan gum manufactured by Kelco.

5 ** ELVANOL[®] Premix: 10% aqueous solution of ELVANOL[®] 90-50.

ELVANOL[®] 90-50 is a grade of polyvinyl alcohol manufactured by E. I. duPont.

*** Pigment: PYLAKLOR[®] Yellow LX-10192 and Permanent Green S-722 (50:50 blend) manufactured by Pylam Products Co. Inc.

10

Activator Formula (Part II) (pH = 12.0):

Ingredients	Wt%	Grams
Deionized Water	75.00	375.00
Sodium Chlorite, 25%	25.00	125.00
TOTAL	100.00	500.00

Typical Mix Ratios:

Base (g)	Activator (g)
3376	186 g (6.25% active) → 0.32% NaClO ₂
3376	93 (6.25% active) → 0.16% NaClO ₂

15

Example XI

A 1000 gram batch of the following experimental base formula was prepared for testing. This composition is similar to Example III with NAS and thickener.

36

Base Formula (Part I) (pH = 2.7):

Ingredients	Wt%	Grams
Glycerin, 96%	5.00	50.00
NEODOL [®] 25-9	.50	5.00
Pelargonic Acid	0.50	5.00
Lactic Acid, 88%	2.95	29.50
KELTROL [®] K5C151	0.10	1.00
Deionized Water	79.45	794.50
Octane Sulfonate	7.00	70.00
KOH, 45%	0.50	5.00
*Pigment 5.0%	4.00	40.00
TOTAL	100.00	1000.0

- 5 * Pigment: PYLAKLOR[®] Yellow LX-10192 and Permanent Green S-722 (50:50 blend) manufactured by Pylam Products Co. Inc.

Activator Formula (Part II) (pH =12.3):

10

Ingredients	Wt%	Grams
Deionized Water	50.00	500.00
Sodium Chlorite, 25%	50.00	500.00
TOTAL	100.00	1000.00

Typical Mix Ratios:

Base (g)	Activator (g)
450	12.39 (12.5% active) → 0.32% NaClO ₂
225	6.2 (12.5% active) → 0.32% NaClO ₂

Example XII

- 15 A 1000 gram batch of the following experimental base formula was prepared for testing. This composition is similar to Example IV with NAS.

Base Formula (Part I) (pH = 2.7):

Ingredients	Wt%	Grams
Sorbitol, 70% active	1.00	10.00
NEODOL [®] 25-9	0.50	5.00
Pelargonic Acid	0.50	5.00
Lactic Acid, 88% active	2.95	29.50
Deionized Water	83.49	834.90
Deionized Water	79.45	794.50
NAS-FAL	7.00	70.00
KOH, 45%	0.56	5.60
*Pigment 5.0%	4.00	40.00
TOTAL	100.00	1000.00

5

* Pigment: PYLAKLOR[®] Yellow LX-10192 and Permanent Green S-722 (50:50 blend) manufactured by Pylam Products Co. Inc.

10

Activator Formula (Part II) (pH = 12.3):

Ingredients	Wt%	Grams
Deionized Water	50.00	500.00
Sodium Chlorite, 25% active	50.00	500.00
TOTAL	100.00	1000.00

Typical Mix Ratios:

Base (g)	Activator (g)
450	12.39 (12.5% active) → 0.32% NaClO ₂
225	6.2 (12.5% active) → 0.32% NaClO ₂

15

Example XIII

A 1500 gram batch of the following experimental base formula was prepared for testing. Similar to Example III with NAS, thickener and heptanoic acid.

Base Formula (Part I) (pH = 2.7):

Ingredients	Wt%	Grams
Glycerin, 96%	5.00	75.00
NEODOL [®] 25-9	0.50	7.50
Heptanoic Acid	0.50	7.50
Lactic Acid, 88%	2.95	44.25
KELTROL [®] K5C151	0.10	1.50
Deionized Water	80.45	1206.75
NAS-FAL	6.00	90.00
KOH, 45%	0.50	7.50
*Dye/Pigment 5.0%	4.00	60.00
TOTAL	100.00	1500.00

5

* Pigment: PYLAKLOR[®] Yellow LX-10192 and Permanent Green S-722 (50:50 blend) manufactured by Pylam Products Co. Inc.

10

Activator Formula (Part II) (pH = 12.0):

Ingredients	Wt%	Grams
Deionized Water	75.00	75.00
Sodium Chlorite, 25%	25.00	25.00
TOTAL	100.00	100.00

Typical Mix Ratios:

Base (g)	Activator (g)
50	1.377 (6.25% active) → 0.16% NaClO ₂
100	2.754 (6.25% active) → 0.16% NaClO ₂
450	12.39 (6.25% active) → 0.16% NaClO ₂

15

Example XIV

A 1500 gram batch of the following experimental base formula was prepared for testing. Similar to Example IV with NAS and heptanoic acid.

Base Formula (Part I) (pH = 2.7):

Ingredients	Wt%	Grams
Sorbitol, 70%	1.00	15.00
NEODOL® 25-9	0.50	7.50
Heptanoic Acid	0.50	7.50
Lactic Acid, 88%	2.95	44.50
Deionized Water	80.45	1206.75
NAS-FAL	6.00	90.00
KOH, 45%	0.54	8.10
*Dye/Pigment 5.0%	4.00	60.00
TOTAL	100.00	1500.00

5

* Pigment: PYLAKLOR® Yellow LX-10192 and Permanent Green S-722 (50:50 blend) manufactured by Pylam Products Co. Inc.

10

Activator Formula (Part II) (pH = 12.0):

Ingredients	Wt%	Grams
Deionized Water	75.00	75.0
Sodium Chlorite, 25%	25.00	25.0
TOTAL	100.00	100.00

Typical Mix Ratios:

Base (g)	Activator (g)
50	1.377 (6.25% active) → 0.16% NaClO ₂
100	2.754 (6.25% active) → 0.16% NaClO ₂
450	12.39 (6.25% active) → 0.16% NaClO ₂

15

Germicidal and Detergent Sanitizing Action of Disinfectants Testingcreated from AOAC Method 960.09

A test to determine the efficacy of antimicrobial products used for sanitizing precleaned, nonporous food contact surfaces.

20

Culture Media

1. Nutrient Agar A:

40

2. Nutrient Agar B (French Slants):

Subculture Media

1. Tryptone Glucose Extract Agar (TGE):

2. Neutralized Tryptone Glucose Extract Agar:

5 The neutralizer used should be appropriate for test substance inactivation.

Reagents

1. Neutralizer Blanks:

a. A mixture of 49.5 mL Chambers solution and 49.5 mL of 1%

aqueous $\text{Na}_2\text{S}_2\text{O}_3$

10

2. Phosphate Buffer Stock (0.25M)

3. Phosphate Buffer Dilution Water

Apparatus

1. Glassware

15 250 ml Erlenmeyer flasks, 100 ml volumetric flasks, pipettes, glass beads, 20x150 and 25x150 mm test tubes. Sterilize for 20 minutes at 121°C or in a dry air oven at 180°C for 180 minutes.

2. Petri Dishes

Sterile disposable petri dishes, 15x100 mm.

20 3. French Bottles (milk dilution bottles)

175 ml flint glass bottles

4. Water Bath

Constant temperature water bath that can maintain a test temperature $\pm 2^\circ\text{C}$ of required test temperature. Monitor temperature throughout the test.

25 5. Transfer Loops

Suitable metal or plastic disposal transfer loops

6. Sterile Buchner Funnel Containing Whatman No. 2 Filter Paper

Staphylococcus aureus ATCC 653830 *Escherichia coli* ATCC 11229

Maintain on Nutrient Agar A slants at 4°C with transfers to new stock slants once per month (reference SOP MS031 for culture maintenance). From the stock culture

41

slant, make >3 and <30 consecutive transfers on nutrient agar slants with incubation at $37^{\circ}\text{C} \pm 2^{\circ}\text{C}$ for 20-24 hours. If only one daily transfer has been missed, no special procedures are required; if 2 or more daily transfers are missed, repeat with 3 daily transfers.

5

Inoculate french slants by washing the growth from the Nutrient Agar A slant into 99 ml of phosphate buffer as follows: Use 5 ml of buffer on the slant and rinse this into the balance of the 99 ml of buffer. Mix this suspension well and add 2 ml of suspension to each french slant. Tilt the slant back and forth to cover the surface.

10 Remove the excess suspension Aseptically. Incubate the slants at $37^{\circ}\text{C} \pm 2^{\circ}\text{C}$ for 18-24 hours.

Remove the culture from the agar surface using 3 ml of phosphate buffer and sterile glass beads rotated back and forth to remove the growth. Filter the suspension
15 through a sterile Buchner funnel containing Whatman No. 2 filter paper that has been prewet with 1 ml of phosphate buffer. Collect the suspension in a sterile test tube.

Standardize the culture suspension by dilution using sterile phosphate buffered water
20 to yield 10×10^9 organisms per milliliter. 10×10^9 organisms/ml corresponds roughly to % transmittance readings of 0.1% to 1.0%T at 580 nm. It is recommended that each individual operator determine what %T readings they need to achieve a 10×10^9 organisms/ml culture suspension prior to performing this test since validity of the test is based on having the proper inoculum.

25

Operating Technique

Dispense 99 ml of test substance into a sterile 250 ml Erlenmeyer flask. Prepare triplicate flasks for each test substance to be tested. Place flasks containing test substance into a 25°C temperature water and let rest ≥ 20 minutes or until reaches
30 test temperature.

Operating Technique with milk challenge

- Dispense 90 ml of test substance and 10 mL of milk challenge into a sterile 250 ml Erlenmeyer flask, mix and remove 1.0mL. Prepare duplicate flasks for each test substance to be tested. Place flasks containing test substance into a 25°C temperature water and let rest ≥ 20 minutes or until reaches test temperature

Enumerate inoculum numbers in sterile phosphate buffer. Enumeration of inoculum numbers will be performed as follows:

10	Prepared Test Culture	1:100 $\rightarrow 10^0$	1:100 $\rightarrow 10^{-2}$	1:100 $\rightarrow 10^{-4}$	1:100 $\rightarrow 10^{-6}$
----	-----------------------------	-----------------------------	--------------------------------	--------------------------------	--------------------------------

- From the 10^{-6} dilution, plate 1 ml (10^6) and 0.1 ml (10^7) in quadruplicate. Use pour plate technique with TGE medium. Invert and incubate at $37^\circ\text{C} \pm 2^\circ\text{C}$ for 48 hours.

- Whirl the test flasks and add 1 ml of culture to 99 ml of the test substance dilution midway between the side of the flask and the center. Avoid touching the sides of the flask with the pipette. Transfer 1 ml portions to the appropriate neutralizer (based on inactivation of the test substance) after 15 seconds exposure and mix well. Longer exposure times may be used depending on the test substance, previous testing and/or the study.

- For regulatory documentation testing, plate 1 ml (10^{-1}) and 0.1 ml (10^{-2}) from the neutralizer blank tube in quadruplicate. For non-regulatory testing, serial dilutions may be performed in sterile phosphate buffer to yield dilutions of 10^{-1} , 10^{-3} , and 10^{-5} . These dilutions are usually single platings. Use pour plate technique with neutralized (appropriate for test substance inactivation) TGE medium. Invert and incubate at $37^\circ\text{C} \pm 2^\circ\text{C}$ for 48 hours.

Controls

- 30 1. Phenol Resistance Method
Determine the resistance of the test system to phenol according to standard procedures. The test system must meet the resistance specified in that SOP.
2. Neutralization Method (created from ASTM E 1054-91)

43

Duplicate neutralization method check(s) should be performed on each test system. If more than one use-solution concentration is used, test the most concentrate solution. Testing should be performed as follows:

Test A = Add 1 ml of test substance use-solution to 9 ml of the neutralizer and mix.

5 Add 0.1 ml of $\sim 10^{-3}$ cfu/ml test system suspension, mix.

Test B = Add 1 ml of test substance diluent to 9 ml of the neutralizer and mix. Add 0.1 ml of $\sim 10^{-3}$ cfu/ml test system suspension, mix.

Test C = Add 0.1 ml of $\sim 10^{-3}$ cfu/ml test system suspension to 9 ml of phosphate buffered dilution water and mix.

10

Let tests stand for 30 minutes. then enumerate by plating 0.1 and 1 ml using pour plate technique and incubating 48 hours at est system specific temperature.

15 The data obtained will show the neutralizer to be effective if $a \cong c$. The neutralizer will be observed not be detrimental to the cells if $b \cong c$.

3. Diluent Control

Plate 1 ml of diluent used in the test. Incubate at $37^{\circ}\text{C} \pm 2^{\circ}\text{C}$ for 48 hours.

TABLE 1

Zero Time Results

Escherichia coli ATCC 11229

20

Test Substance	Organic Challenge	Time Exposure	Log Reduction
Ex. II Prepared (06-19-97)	No Challenge	15 seconds	6.38
Ex. II Prepared (06-19-97)	10% Milk	15 seconds	6.38
UDDER GOLD PLUS Prepared (06-19-97)	No Challenge	15 seconds	6.38
UDDER GOLD PLUS Prepared (06-19-97)	10% Milk	15 seconds	6.38

44

TABLE 1A*Staphylococcus aureus* ATCC 6538

Test Substance	Organic Challenge	Time Exposure	Log Reduction
Ex. II Prepared (06-19-97)	No Challenge	15 seconds	5.87
Ex. II Prepared (06-19-97)	10% Milk	15 seconds	5.87
UDDER GOLD PLUS Prepared (06-19-97)	No Challenge	15 seconds	5.87
UDDER GOLD PLUS Prepared (06-19-97)	10% Milk	15 seconds	5.87

Conclusions:

- 5 When freshly prepared both the composition of Example II and UDDER GOLD PLUS achieved a greater than 5 log reduction after 15 seconds with and without a 10% milk challenge against both *Staphylococcus aureus* ATCC 6538 and *Escherichia coli* ATCC 11229.

One Week Results

10

TABLE 1B*Escherichia coli* ATCC 11229

Test Substance	Organic Challenge	Time Exposure	Log Reduction
Ex. II Prepared (06-19-97)	No Challenge	15 seconds	5.93
Ex. II Prepared (06-19-97)	10% Milk	15 seconds	5.93
UDDER GOLD PLUS Prepared (06-19-97)	No Challenge	15 seconds	1.42
UDDER GOLD PLUS Prepared (06-19-97)	10% Milk	15 seconds	0.61

45

TABLE 1C*Staphylococcus aureus* ATCC 6538

Test Substance	Organic Challenge	Time Exposure	Log Reduction
Ex. II Prepared (06-19-97)	No Challenge	15 seconds	5.82
Ex. II Prepared (06-19-97)	10% Milk	15 seconds	5.82
UDDER GOLD PLUS Prepared (06-19-97)	No Challenge	15 seconds	1.40
UDDER GOLD PLUS Prepared (06-19-97)	10% Milk	15 seconds	0.59

After one week, Example II achieved a >5 log reduction after 15 seconds with and without a 10% milk challenge against both *Staphylococcus aureus* ATCC 6538 and *Escherichia coli* ATCC 11229. UDDER GOLD PLUS achieved a 1.42 log reduction against *Escherichia coli* ATCC 11229 in 15 seconds without milk, while achieving a 0.61 log reduction with the 10% milk challenge. Against *Staphylococcus aureus* ATCC 6538 UDDER GOLD PLUS achieved a log reduction of 1.42 without milk, and 0.59 with a 10% milk challenge. Two Weeks Results

TABLE 1D*Escherichia coli* ATCC 11229

Test Substance	Organic Challenge	Time Exposure	Log Reduction
Ex. II Prepared (06-19-97)	No Challenge	15 seconds	5.74
Ex. II Prepared (06-19-97)	10% Milk	15 seconds	4.00
UDDER GOLD PLUS Prepared (06-19-97)	No Challenge	15 seconds	2.06
UDDER GOLD PLUS Prepared (06-19-97)	10% Milk	15 seconds	0.26

46

TABLE 1E*Staphylococcus aureus* ATCC 6538

Test Substance	Organic Challenge	Time Exposure	Log Reduction
Ex. II Prepared (06-19-97)	No Challenge	15 seconds	5.75
Ex. II Prepared (06-19-97)	10% Milk	15 seconds	5.75
UDDER GOLD PLUS Prepared (06-19-97)	No Challenge	15 seconds	2.09
UDDER GOLD PLUS Prepared (06-19-97)	10% Milk	15 seconds	0.32

After two weeks, Example II achieved a >5 log reduction after 15 seconds with and without a 10% milk challenge against *Staphylococcus aureus* ATCC 6538. Against *Escherichia coli* ATCC 11229 a reduction of 4.00 was seen with the 10% milk challenge while a 5.74 log reduction was seen without the challenge. UDDER GOLD PLUS achieved a 2.06 log reduction against *Escherichia coli* ATCC 11229 in 15 seconds without milk, while achieving a 0.26 log reduction with the 10% milk challenge. Against *Staphylococcus aureus* ATCC 6538 UDDER GOLD PLUS achieved a log reduction of 2.09 without milk, and 0.32 with a 10% milk challenge.

Three Weeks Results

TABLE 1F*Escherichia coli* ATCC 11229

Test Substance	Organic Challenge	Time Exposure	Log Reduction
Ex. II Prepared (06-19-97)	No Challenge	15 seconds	5.67
Ex. II Prepared (06-19-97)	10% Milk	15 seconds	3.91
UDDER GOLD PLUS Prepared (06-19-97)	No Challenge	15 seconds	1.86
UDDER GOLD PLUS Prepared (06-19-97)	10% Milk	15 seconds	0.26

47

TABLE 1G*Staphylococcus aureus* ATCC 6538

Test Substance	Organic Challenge	Time Exposure	Log Reduction
Ex. II Prepared (06-19-97)	No Challenge	15 seconds	5.76
Ex. II Prepared (06-19-97)	10% Milk	15 seconds	5.76
UDDER GOLD PLUS Prepared (06-19-97)	No Challenge	15 seconds	1.78
UDDER GOLD PLUS Prepared (06-19-97)	10% Milk	15 seconds	0.36

After three weeks, Example II achieved a >5 log reduction after 15 seconds with and without a 10% milk challenge against *Staphylococcus aureus* ATCC 6538. Against *Escherichia coli* ATCC 11229 a reduction of 3.91 was seen with the 10% milk challenge while a 5.67 log reduction was seen without the challenge. UDDER GOLD PLUS achieved a 1.86 log reduction against *Escherichia coli* ATCC 11229 in 15 seconds without milk, while achieving a 0.26 log reduction with the 10% milk challenge. Against *Staphylococcus aureus* ATCC 6538 UDDER GOLD PLUS achieved a log reduction of 1.78 without milk, and 0.36 with a 10% milk challenge.

Four Weeks Results

TABLE 1H*Escherichia coli* ATCC 11229

Test Substance	Organic Challenge	Time Exposure	Log Reduction
Ex. II Prepared (06-19-97)	No Challenge	15 seconds	>5.5
Ex. II Prepared (06-19-97)	10% Milk	15 seconds	>5.5
UDDER GOLD PLUS Prepared (06-19-97)	No Challenge	15 seconds	1.2
UDDER GOLD PLUS Prepared (06-19-97)	10% Milk	15 seconds	0.1

TABLE 11*Staphylococcus aureus* ATCC 6538

Test Substance	Organic Challenge	Time Exposure	Log Reduction
Ex. II Prepared (06-19-97)	No Challenge	15 seconds	>6.0
Ex. II Prepared (06-19-97)	10% Milk	15 seconds	5.3
UDDER GOLD PLUS Prepared (06-19-97)	No Challenge	15 seconds	1.3
UDDER GOLD PLUS Prepared (06-19-97)	10% Milk	15 seconds	0.6

- 5 After four weeks, Example II achieved a >5 log reduction after 15 seconds with and without a 10% milk challenge against both *Staphylococcus aureus* ATCC 6538 and *Escherichia coli* ATCC 11229. UDDER GOLD PLUS achieved a 1.2 log reduction against *Escherichia coli* ATCC 11229 in 15 seconds without milk, while achieving a 0.6 log reduction with the 10% milk challenge. Against *Staphylococcus aureus*
- 10 ATCC 6538 UDDER GOLD PLUS achieved a log reduction of 1.3 without milk, and 0.6 with a 10% milk challenge.

Porcine Skin Test

- An analysis was made to determine the antimicrobial activity of teat dips applied to porcine skin inoculated with *Staphylococcus aureus* ATCC 6538 and *Escherichia coli* ATCC 11229. Using the following Test Method:
- 15

1. One inch squares of sterile lyophilized porcine skin (Corethium™2, Johnson & Johnson UK) were rehydrated in sterile distilled water for 1 hour.
2. Hydrated skin squares were dipped into each teat dip formulation for 10 seconds and hung in a vertical position to allow excess to drain off.
- 20 3. The squares were then placed in a sterile petri dish and inoculated with 10 microliters of a 24 hour broth culture for the organism to be tested. After inoculation, samples were allowed a contact time of 5 minutes.

4. Skin squares were then put in a tube containing 20 mL of the appropriate neutralizer.
5. Samples were then vortexed and dilutions of 10^{-1} , 10^{-3} , and 10^{-5} were plated in order to enumerate the survivors.
- 5 6. Plates were incubated at 37°C for 48 hours.
7. A neutralization test was also performed.

Method Parameters:

Test Substance	Active Ingredients
ClO ₂ Example II Formulation Prepared 6/19/97)	2.00% LAS ClO ₂
UDDER GOLD PLUS (Prepared 6/19/97)	ClO ₂
Water Control	None

Test Systems: *Staphylococcus aureus* ATCC 6538
 10 *Escherichia coli* ATCC 11229

Test Temperature: Room Temperature

Exposure Time: 5 minutes

Subculture Medium: Tryptone Glucose Extract Agar

Incubation: 37°C for 48 hours

15 Results:

TABLE 2A

Zero Time

Staphylococcus aureus ATCC 6538

Test Substance	Exposure Time to Teat Dip (min)	Log Reduction
ClO ₂ Example II Formula (Prepared 6/19/97)	5	2.73
UDDER GOLD PLUS (Prepared 6/19/97)	5	No Reduction
Water Control	5	N/A

TABLE 2B*Escherichia coli* ATCC 11229

Test Substance	ExposureTime to Teat Dip (min)	Log Reduction
ClO ₂ Example II Formula (Prepared 6/19/97)	5	1.78
UDDER GOLD PLUS (Prepared 6/19/97)	5	1.32
Water Control	5	N/A

5

Results:

One Week
TABLE 2C*Staphylococcus aureus* ATCC 6538

10

Test Substance	ExposureTime to Teat Dip (min)	Log Reduction
UDDER GOLD PLUS (Prepared 6/19/97)	5	No Reduction
ClO ₂ Example II Formula (Prepared 6/19/97)	5	3.48
Water Control	5	N/A

TABLE 2D*Escherichia coli* ATCC 11229

15

Test Substance	ExposureTime to Teat Dip (min)	Log Reduction
UDDER GOLD PLUS (Prepared 6/19/97)	5	0.20
ClO ₂ Example II Formula (Prepared 6/19/97)	5	1.83
Water Control	5	N/A

Results:

Two WeeksTABLE 2E

5

Staphylococcus aureus ATCC 6538

Test Substance	ExposureTime to Teat Dip (min)	Log Reduction
UDDER GOLD PLUS (Prepared 6/19/97)	5	No Reduction
ClO ₂ Example II Formula (Prepared 6/19/97)	5	3.52
Water Control	5	N/A

Escherichia coli ATCC 11229

10

TABLE 2F

Test Substance	ExposureTime to Teat Dip (min)	Log Reduction
UDDER GOLD PLUS (Prepared 6/19/97)	5	0.08
ClO ₂ Example II Formula (Prepared 6/19/97)	5	0.60
Water Control	5	N/A

15 Results:

Three WeeksTABLE 2G*Staphylococcus aureus* ATCC 6538

Test Substance	ExposureTime to Teat Dip (min)	Log Reduction
UDDER GOLD PLUS (Prepared 6/19/97)	5	No Reduction
ClO ₂ Example II Formula (Prepared 6/19/97)	5	2.26
Water Control	5	N/A

52

TABLE 2H*Escherichia coli* ATCC 11229

Test Substance	ExposureTime to Teat Dip (min)	Log Reduction
UDDER GOLD PLUS (Prepared 6/19/97)	5	0.77
ClO ₂ Example II Formula (Prepared 6/19/97)	5	1.73
Water Control	5	N/A

5

Results:

Four WeeksTABLE 2I*Staphylococcus aureus* ATCC 6538

Test Substance	ExposureTime to Teat Dip (min)	Log Reduction
UDDER GOLD PLUS (Prepared 6/19/97)	5	0.19
ClO ₂ Example II Formula (Prepared 6/19/97)	5	2.78
Water Control	5	N/A

10

TABLE 2J*Escherichia coli* ATCC 11229

Test Substance	ExposureTime to Teat Dip (min)	Log Reduction
UDDER GOLD PLUS (Prepared 6/19/97)	5	0.07
ClO ₂ Example II Formula (Prepared 6/19/97)	5	1.72
Water Control	5	N/A

15

These test results show the compositions of the invention are superior in these anti-microbial tests to a quality commercial teat dip. These data are representative in anti-microbial properties of the exemplary formulations of the invention.

The teat dip compositions of the invention using nonanoic acid and a sulfonate material in a formulation with 2-propanol, were tested for their properties in coating and forming a useful antimicrobial layer that maintains a sufficient

20

quantity of material on the animal. A simulated test was conducted involving a test tube. In the method a Kimax® brand glass test tube (20mm x 150mm) is weighed, and dipped approximately 2 inches into the teat dip being tested. The test tube is then removed from the dip and placed on hanging rack for 10 minutes above a beaker that was previously weighed. At the end of 10 minutes the beaker and the test tube are each weighed again and the data is entered into the table. The dry weight is obtained by allowing the resulting teat dip film to dry for 24 hrs.

Simulated Teat Dipping Test
UDDER GOLD PLUS

	Wet Wt. (g)	Wet Wt. (g)	Dry Wt. (g)	Dry Wt. (g)
Sample Name	UDDER GOLD PLUS 1	UDDER GOLD PLUS 2	UDDER GOLD PLUS 1	UDDER GOLD PLUS 2
Beaker Tare	110.3762	106.9215	110.3762	106.9215
Test Tube Tare	24.5295	18.9019	24.5295	18.9019
Beaker @ 10 min.	111.7136	108.1773	110.5015	107.0377
Test Tube @ 10 min.	24.6747	19.0028	24.5459	18.9151
Product on Test Tube	0.1452	0.1009	0.0164	0.0132
Product in Beaker	1.3374	1.2558	0.1253	0.1162
% Product Retained	9.79%	7.44%	11.57%	10.20%
% Product Lost	90.21%	92.56%	88.43%	89.80%

Comments: Wet Wt. = 10 minutes
Dry Wt. = 24 hours

Simulated Teat Dipping Test Composition of Example I

	Wet Wt. (g)	Wet Wt. (g)	Dry Wt. (g)	Dry Wt. (g)
Sample Name	Example I	Example I	Example I	Example I
Beaker Tare	108.0534	107.0201	108.0534	107.0201
Test Tube Tare	25.0294	24.9161	25.0294	24.9161
Beaker @ 10 min.	108.3413	107.3925	108.1192	107.1081
Test Tube @ 10 min.	25.1733	25.0665	25.0596	24.9432
Product on Test Tube	0.1439	0.1504	0.0302	0.0271
Product in Beaker	0.2879	0.3724	0.0658	0.0880
% Product Retained	33.33%	28.77%	31.46%	23.54%
% Product Lost	66.67%	71.23%	68.54%	76.46%

- | | | | |
|---|-----------|--|---|
| 5 | Comments: | Wet Wt. = 10 minutes
Dry Wt. = 24 hours | Formula Example I = 2.5% 2-propanol
1.5% C9
17.0% NAS |
|---|-----------|--|---|

A brief examination of the tabulated data show that the rheology of the material of Example I maintains a larger quantity of material on the simulated animal when compared to the rheology of the commercial teat dip formulation. These data suggest that the composition of Example I would be somewhat more effective in mastitis treatment because the formulations of the invention would maintain a larger quantity of treating composition in a longer lasting film than the commercial materials. In other testing we have found that the identity of the antibacterial material, carboxylic acid, fatty acid, phosphoric acid or sulfonic acid, does not significantly change the rheology of the material and a fully formulated material having rapid initial kill and long term kill can be formulated in a long lasting film-forming composition.

Detailed Discussion of the Drawings

Figure 1A and Figure 1B are a graphical representations of the data shown in the tables 1A - 1I showing the results of a four week biocidal efficacy analysis of Example II using the food contact sanitizing protocol preceding the data in the

specification. Clearly, from zero time through four weeks the composition of Example II had a kill substantially greater than a 5 log reduction in microbial populations of both model microorganisms. This data was taken without a milk challenge. However, Figure 1B shows, within experimental error, similar properties using the same food contact sanitizing protocol with a 10% milk soil challenge. The week 2 and week 3 kill of E. coli is not easily explained in view of the 5.5 log reduction obtained in week 4 for that model microorganism. However, overall the results are strikingly successful in reducing microorganism populations on food contact surfaces.

Figure 1C and Figure 1D are graphical representations of data showing the results of the four week biocidal food contact sanitizing efficacy analysis of a commercial teat dip composition sold under the tradename UDDER GOLD PLUS.

The superiority in performance of the composition of Example II when compared to the UDDER GOLD PLUS compositions is marked particularly in weeks 1 through 4.

Figure 2A, a graphical representation of the data shown in tables 2A - 2J is a four week efficacy analysis of the UDDER GOLD PLUS chlorine dioxide teat dip formulation measured using the procein skin protocol at pages 51 to 52 in the specification. In all test periods the material failed to achieve greater than a 2 log reduction.

Figure 2B, prepared from the corresponding data given in the forementioned tables, shows a four week efficacy analysis of Example II using the porcine skin protocol. Clearly, under these more difficult conditions using the porcine skin substrate as a test vehicle, the log reductions in kill are not as great as those for food contact surfaces, however, the biocidal efficacy of Example II exceeded that of the well regarded UDDER GOLD PLUS chlorine dioxide formulations.

Figure 3, shows a graph of the change in viscosity measured in centipoise plotted against shear (rpm) for UDDER GOLD PLUS and Example II and Example IX of the invention. The examples of the invention should show classic non-Newtonian or nonlinear viscosity with respect to shear. At low shear, i.e., low rpm, the viscosity is high. As shear increases, viscosity drops. In sharp contrast, viscosity of the UDDER GOLD PLUS formulation shows much less pseudoplastic

behavior with threshold shear stress. The viscosity is substantially constant as shear or rpm increases. This viscosity information is consistent with the data of the previous table and demonstrates that the viscoelastic behavior of the compositions of the invention are likely to coat and adhere to teat tissue more efficiently than

5 UDDER GOLD PLUS. Since the materials are sheared somewhat upon application, the materials will flow on to the teat surface. However, when shear is removed, the teat dips obtain high viscosity and tend to adhere more tenaciously than a composition such as UDDER GOLD PLUS having linear viscosity. The substantial difference in viscosity at low shear (low rpm) demonstrates that the materials should

10 coat the teat with substantially more material and after drying have a more effective environmental barrier.

The procedure is as follows. Place ~ 500 mls of the experimental teat dip in 600 ml beaker. Record viscosity measurements at various rpms. Apparatus: Brookfield RVT Viscometer, spindle #1 and 2, various rpm (see chart below). Start

15 Temperature: 73.6° F- Stop Temperature: 73.8° F

RPM	Example II	Example IX	U.Gold Plus	Spindle
5	940	802	244	1
10	595	501	240	1
20	374	315	234	1
50	247	210	230	2
100	156	141	224	2

The above specification, example and data provide a clear basis for understanding the operation of the compositions and methods of the invention.

20 While the invention can be embodied in a variety of specific examples and processes, the invention resides in the claims hereinafter appended. In the claims, the proportions are expressed in parts by weight per each one hundred parts of the claimed antimastitis composition as a whole.

WE CLAIM:

1. A physically and chemically stable mastitis treating composition, in an acidulant part and a chlorite part, that can effectively reduce microbial populations, on contact and for an extended period, said composition comprising:

5 (a) an acidulant part comprising organic ingredients, said ingredients comprising:

(i) about 0.1 to 15 parts by weight of a C_{1-6} organic monocarboxylic acid or dicarboxylic acid or mixtures thereof;

10 (ii) about 0.1 to 15 parts by weight of an antimicrobial acid comprising a C_{6-12} carboxylic acid or salt thereof or a hydrocarbon sulfonic acid or salt thereof;

(iii) about 0.1 to 20 parts by weight of an organic polymeric thickener, an organic polymeric film-forming agent or mixtures thereof; and

15 (iv) a major proportion of water;

(b) a chlorite part, substantially free of an organic component, consisting essentially of an alkali metal chlorite salt;

20 wherein the composition exhibits rheology that promotes cling to immobilize the composition on the target surface and provides a barrier to environmental contamination.

2. The composition of claim 1 wherein the alkali metal chlorite comprises sodium chlorite.

3. The composition of claim 1 wherein the organic monocarboxylic acid comprises lactic acid.

25 4. The composition of claim 1 wherein the C_{6-12} carboxylic acid comprises a fatty acid having 7, 8, 9, 10 or 11 carbon atoms or a mixture of two or more acids thereof.

5. The composition of claim 1 wherein the C_{6-12} carboxylic acid comprises decanoic acid.

30

6. The composition of claim 1 wherein the C_{6-12} carboxylic acid comprises heptanoic acid.

7. The composition of claim 1 wherein the C₆₋₁₂ carboxylic acid comprises octanoic acid.

8. The composition of claim 1 wherein the C₆₋₁₂ carboxylic acid comprises nonanoic acid.

5 9. The composition of claim 1 which additionally comprises a hydrocarbon sulfonic acid comprising an alkyl sulfonic acid or an alkylbenzene sulfonic acid wherein the alkyl group has 6 to 18 carbon atoms.

10 10. The composition of claim 1 in subpart (iii) wherein the composition comprises a mixture of about 0.05 to 2 parts by weight of a xanthan thickener and about 0.1 to 5 parts by weight of a film-forming agent selected from the group consisting of a polyvinyl alcohol, a polyvinyl acetate and mixtures thereof.

11. The composition of claim 10 wherein the organic polymeric thickening agent comprises a material that imparts pseudoplastic behavior with threshold shear stress to the final composition.

15 12. The composition of claim 1 wherein the composition additionally comprises about 1 to 10 parts of an emollient.

13. The composition of claim 1 wherein the pH of the final material blended from the acidulant part and the chlorite part has a pH about 2 to 4.5.

20 14. The composition of claim 1 wherein the pH of the final material blended from the acidulant part and the chlorite part has a pH about 2 to 3.8.

15. The composition of claim 1 wherein the chlorite part is a solid inorganic part.

16. The composition of claim 1 wherein the chlorite part is an aqueous inorganic part.

25 17. The composition of claim 15 wherein the chlorite part is a solid tablet comprising about equal proportions of sodium carbonate and sodium chlorite.

18. The composition of claim 15 wherein the chlorite part is a powdered inorganic part.

30 19. The composition of claim 10 wherein the polyvinyl alcohol has a molecular weight (M_n) of about 15 x 10³ to 100 x 10³ and a degree of hydrolysis of about 92 to 99%.

20. A method of forming a mastitis treatment composition which comprises combining:

(a) an acidulant part comprising organic ingredients, said ingredients comprising:

- 5 (i) about 0.1 to 15 parts by weight of an C_{1-6} organic monocarboxylic acid or mixtures thereof;
- (ii) about 0.1 to 15 parts by weight of an antimicrobial acid comprising a C_{6-12} carboxylic acid or salt thereof;
- 10 (iii) about 0.1 to 20 parts by weight of an organic polymeric thickener, an organic polymeric film-forming agent or mixtures thereof; and
- (iv) a major proportion of water;

(b) a chlorite part substantially free of an organic component, consisting essentially of an alkali metal chlorite salt; to form an aqueous thickened teat dip material containing less than about 1500 parts per million ClO_2 and less than

15 about 3900 parts per million oxygenated chlorine species sanitizing component: wherein the composition exhibits rheology that promotes cling to immobilize the composition on the surface of a dairy animal teat and provides a barrier to environmental contamination.

20 21. The method of claim 20 wherein the alkali metal chlorite comprises sodium chlorite.

22. The method of claim 20 wherein the organic acid comprises lactic acid.

25 23. The method of claim 20 wherein the C_{6-12} carboxylic acid comprises a fatty acid having 7, 8, 9, 10 or 11 carbon atoms or a mixture of two or more acids thereof.

24. The method of claim 20 wherein the C_{6-12} carboxylic acid comprises heptanoic acid.

25. The method of claim 20 wherein the C₆₋₁₂ carboxylic acid comprises octanoic acid.

26. The method of claim 20 wherein the C₆₋₁₂ carboxylic acid comprises nonanoic acid.

5 27. The method of claim 20 wherein the C₆₋₁₂ carboxylic acid comprises decanoic acid.

28. The method of claim 20 wherein the teat dip additionally comprises a hydrocarbon sulfonic acid comprising an alkyl sulfonic acid or an alkylbenzene sulfonic acid wherein the alkyl group has 9 to 18 carbon atoms.

10 29. The method of claim 27 in subpart (iii) wherein the composition comprises a mixture of about 0.05 to 2 parts by weight of a xanthan thickener and about 0.1 to 5 parts by weight of a film-forming agent selected from the group consisting of a polyvinyl alcohol, a polyvinyl acetate and mixtures thereof.

15 30. The method of claim 20 wherein the organic polymeric thickening agent comprises a material that imparts pseudoplastic behavior with threshold shear to the final composition.

31. The method of claim 20 wherein the composition additionally comprises about 1 to 10 parts of an emollient.

20 32. The method of claim 20 wherein the pH of the final material blended from the acidulant part and the chlorite part has a pH about 2 to 4.5.

33. The method of claim 20 wherein the pH of the final material blended from the acidulant part and the chlorite part has a pH about 2 to 3.8.

34. The method of claim 20 wherein the chlorite part is a solid inorganic part.

25 35. The method of claim 20 wherein the chlorite part is an aqueous inorganic part.

36. The method of claim 20 wherein the chlorite part is a solid tablet comprising equal proportions of sodium carbonate and sodium chlorite.

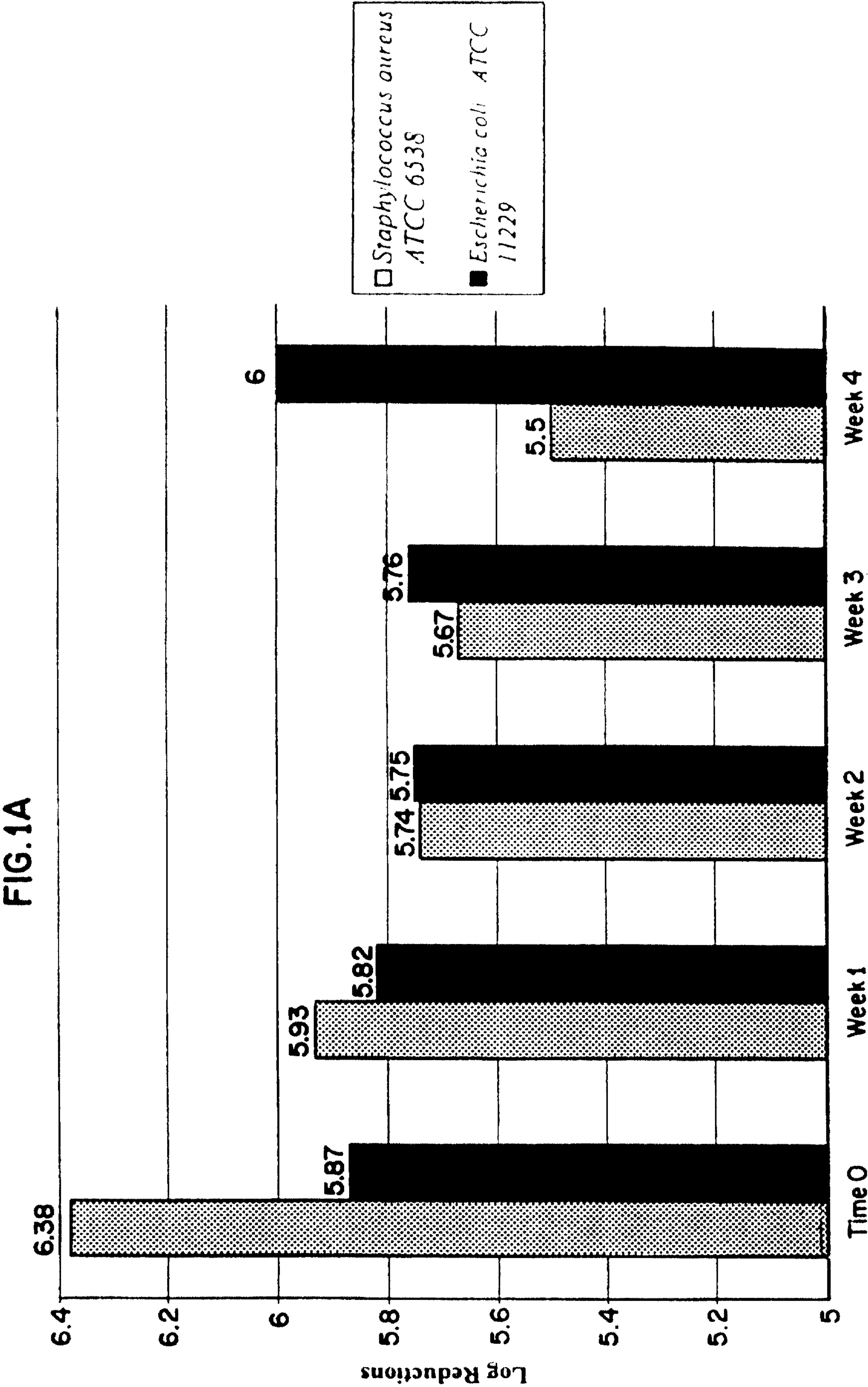
37. The method of claim 30 wherein the chlorite part is a powdered inorganic part.

38. The method of claim 29 wherein the polyvinyl alcohol has a molecular weight (Mn) of about 15×10^3 to 100×10^3 , and a degree of hydrolysis of about 92 to 99%.

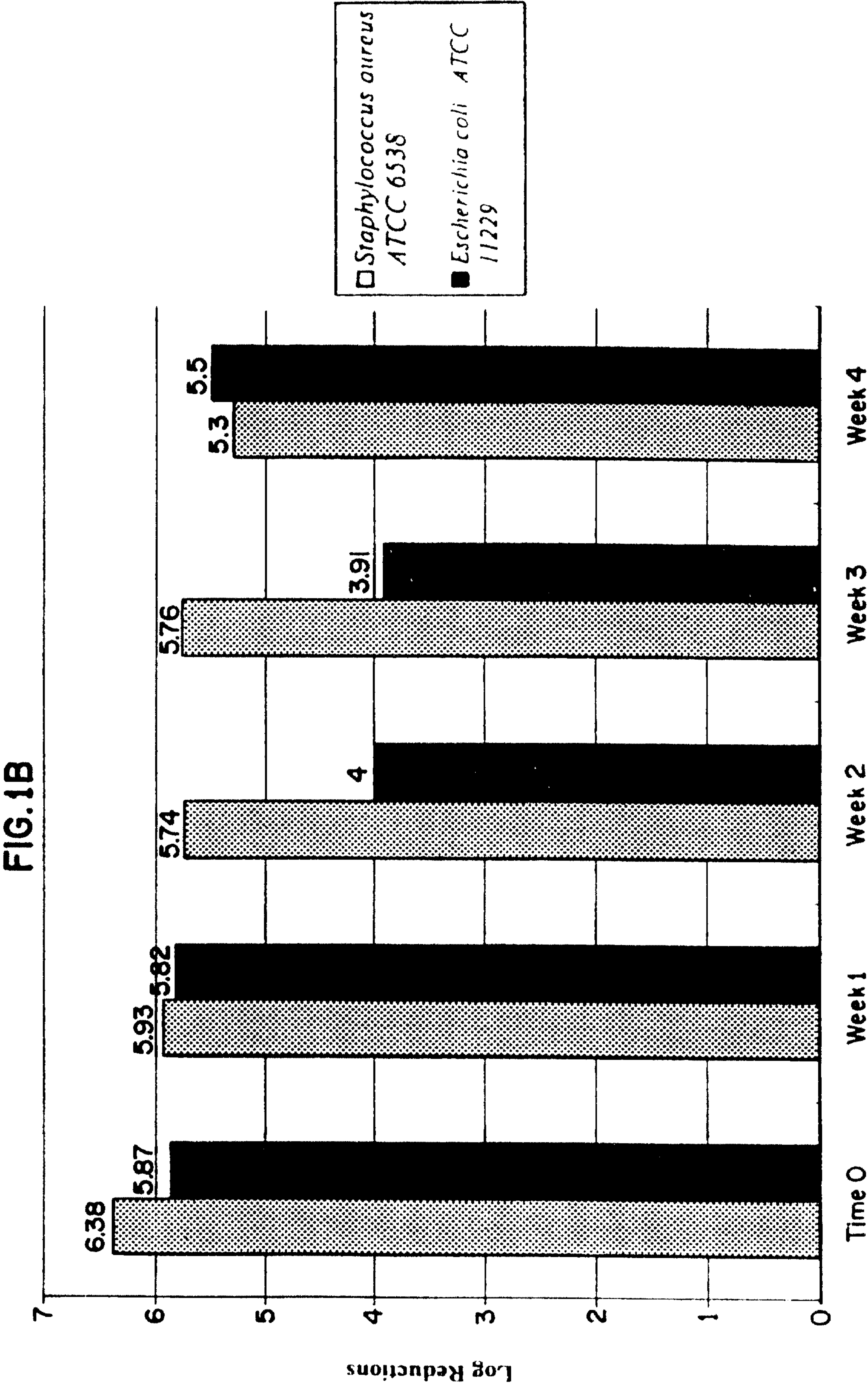
5

CASSAN MACLEAN
Suite 401, 80 Aberdeen Street
Ottawa, Ontario
K1S 5R5

Agents for the Applicant

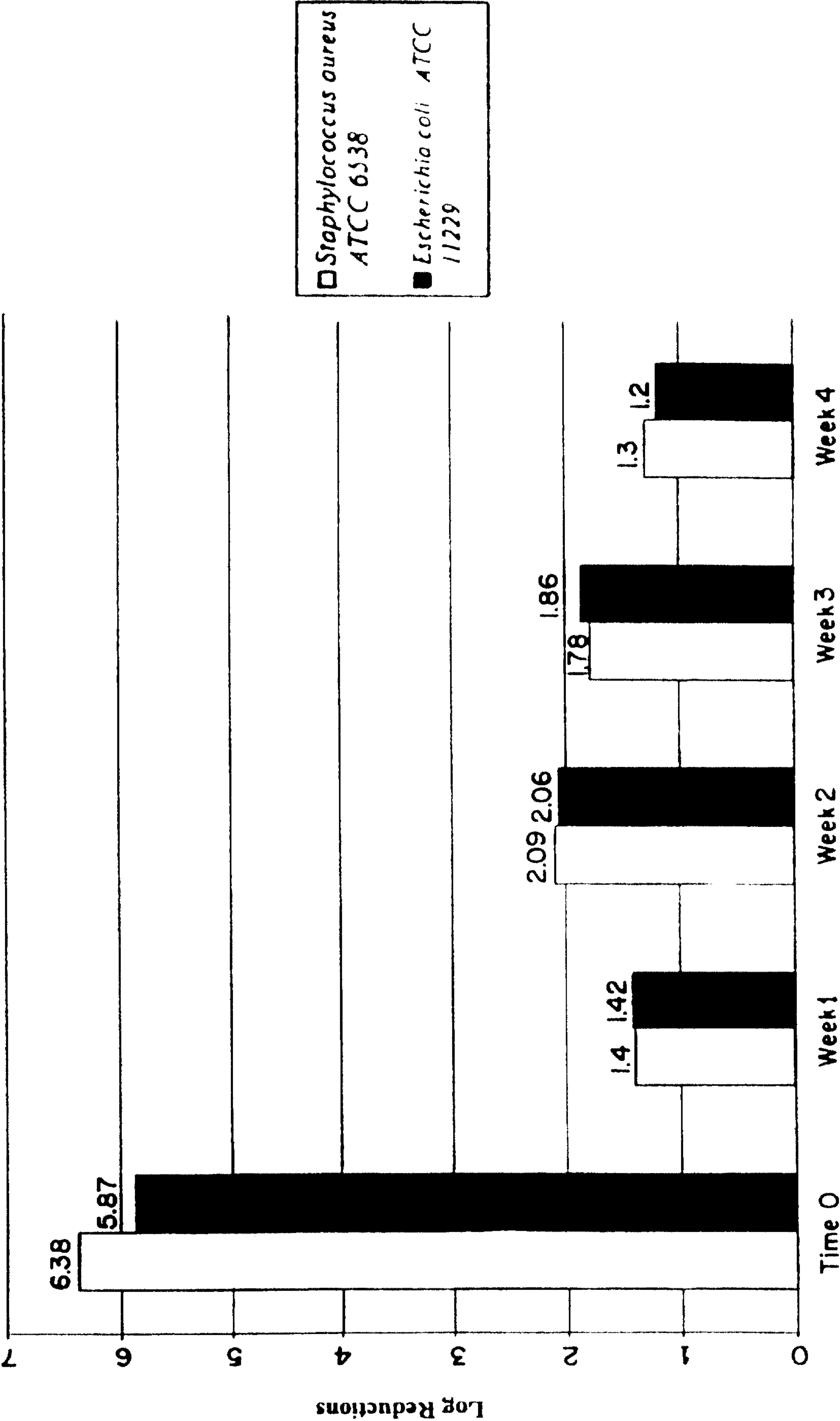


2/7

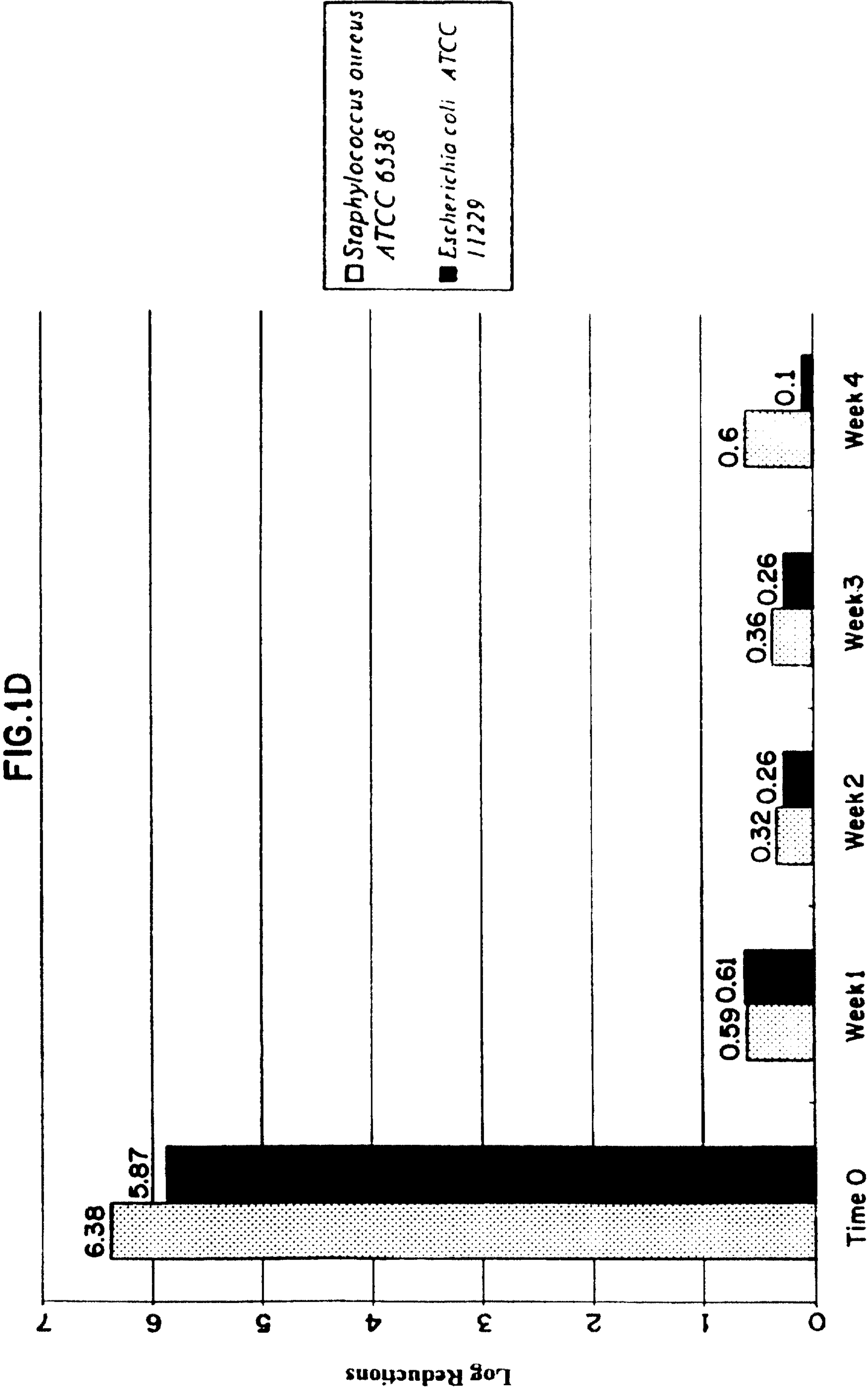


3/7

FIG.1C

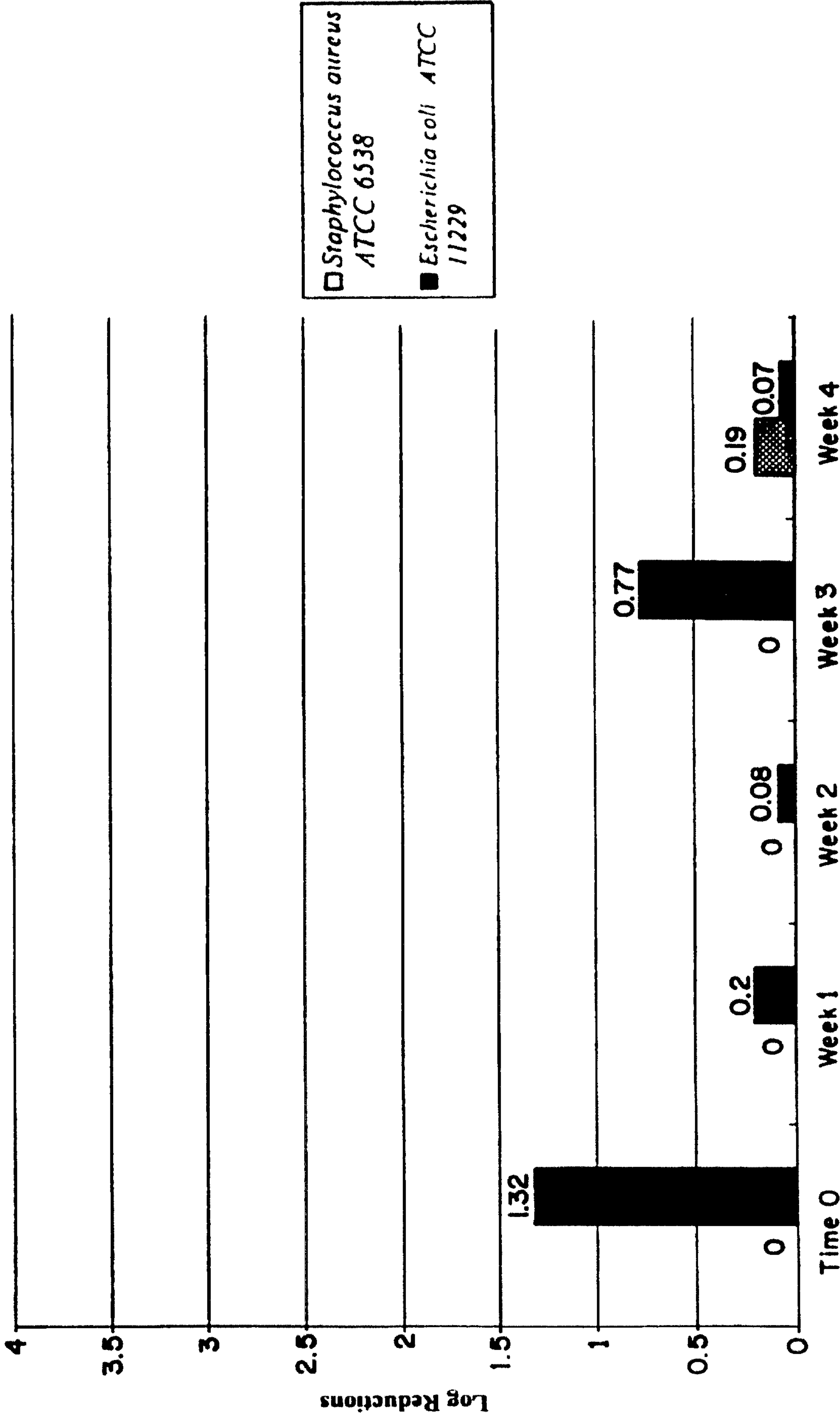


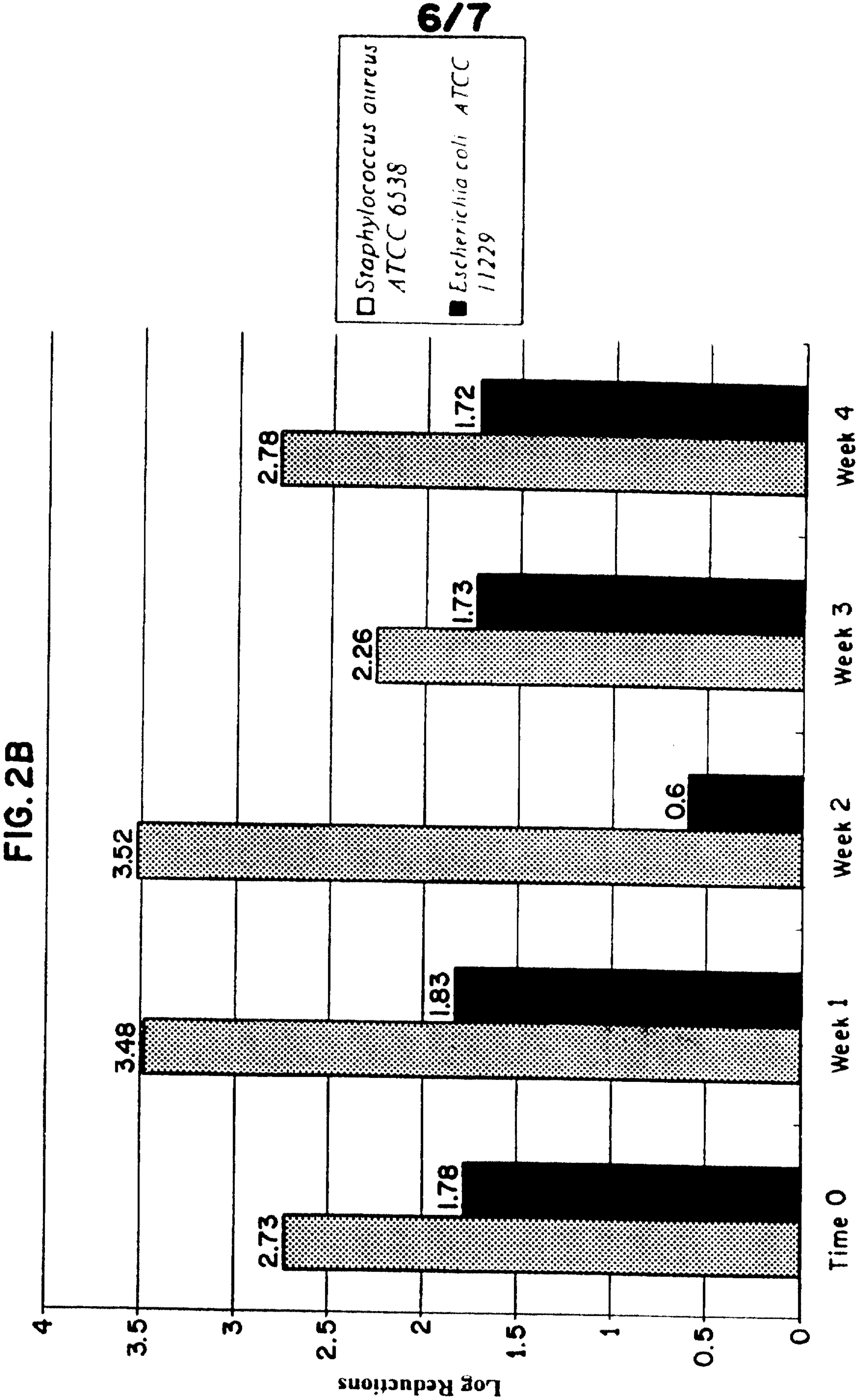
4/7



5/7

FIG. 2A





6/7

FIG. 3

