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NEW ANTIBACTERIAL AND ANTIFUNGAL COMPOSITIONS

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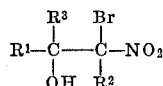
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5 Claims

ABSTRACT OF THE DISCLOSURE

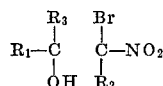
Antibacterial and antifungal compositions having as active ingredient a compound of the formula



wherein R¹ is hydrogen, alkyl of 1–12 C-atoms or phenyl alkyl of up to 10 C-atoms, R³ is hydrogen or R¹ and R³ together with the shared C atom form a cycloalkyl ring of 5–7 C-atoms and R² is hydrogen, methyl, ethyl, hydroxy methyl or bromine and their use in combating bacteria on skin and in preserving medical, toilet and cosmetic preparations from bacterial deterioration.

This invention concerns improvements in or relating to antibacterial and antifungal compositions. More particularly it relates to compositions which comprise compounds selected from a group of nitroalkanols which have been found to possess outstanding antibacterial and antifungal properties.

The invention is based on our discovery that compounds of the general formula



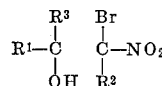
in which R¹ is hydrogen, alkyl having one to twelve carbon atoms or phenylalkyl having up to ten carbon atoms and R³ is hydrogen, or R¹ and R³ together with the shared carbon atom form a cycloalkyl ring having five to seven carbon atoms, and R² is hydrogen, methyl, ethyl, hydroxy-methyl or bromine possess outstanding antibacterial and antifungal properties, being active against a wide spectrum of bacteria and fungi. Thus, for example, they are active against the following organisms: (a) Gram-positive bacteria such as *Staphylococcus* spp., *Streptococcus* spp., *Corynebacterium pyogenes*, *Erysipelothrix rhusiopathiae* and *Mycobacterium* spp.; (b) Gram-negative bacteria, such as for example *Salmonella* spp., *Shigella* spp., *Escherichia coli*, *Aerobacter aerogenes*, *Pseudomonas pyocyanea*, *Proteus vulgaris* and *Haemophilus* spp.; (c) Dermatophytes of the Trichophyton spp. and Microsporum spp.; (d) Actinomycetes spp.; (e) yeasts such as *Candida albicans*, *Cryptococcus neoformans* and *Sporotrichum schenki*; (f) spoilage organisms such as *Pseudomonas* spp., spore-bearing bacilli, *Penicillia* spp., *Aspergillus* spp.; and *Mucor* spp. A particular feature of the compounds is their high activity against Gram-negative organisms which are difficult to control with known antibacterial agents. An extremely important feature of the

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compounds is their activity against *Pseudomonas pyocyanea* which has hitherto proved to be extremely resistant to the majority of known antibacterial agents.

It will be apparent to those skilled in the art that we have discovered outstanding antibacterial and antifungal activity in the hereinbefore defined compounds. This activity renders these compounds of great value for the control of bacteria and fungi, for example as active ingredients of compositions for the treatment of bacterial and fungal infections in human and veterinary medicine, and as preservatives in a variety of applications.

According to one aspect of the present invention there are provided compositions for use in human and veterinary medicine which comprise a compound of the general formula



in which R¹, R² and R³ are as hereinbefore defined, in association with a pharmaceutically acceptable diluent or carrier. In this specification and claims, the terms "compositions for use in human and veterinary medicine" and "medical or veterinary preparations" are to be interpreted as including compositions, such as face creams, shampoos etc., which are normally designated as toilet or cosmetic preparations.

In discussing the types of composition which fall within the scope of the present invention, it is convenient to consider (a) those compositions which are provided in order that the antibacterial and antifungal properties of the hereinbefore defined compounds may be utilised to treat bacterial and fungal infections, especially topical infections, in human and veterinary medicine, and (b) those compositions in which a compound as hereinbefore defined is included primarily as a preservative to control microbial spoilage.

(A) COMPOSITIONS FOR THE TREATMENT OF BACTERIAL AND FUNGAL INFECTIONS

The compositions according to this aspect of the invention comprise the compound hereinbefore defined formulated in any way known in the art for formulating therapeutic agents for the treatment of bacterial and fungal infections, and the pharmaceutically acceptable diluents or carriers included within the scope of the invention accordingly comprise any diluent or carrier normally used for this purpose. Particularly valuable compositions are those for topical use which for example may take the form of creams, lotions, ointments, jellies, medicated powders, solid sticks, suppositories, aerosol preparations and solutions suitable for use as eardrops. The compositions of the invention may also take the form of lozenges, boiled sweets, pastilles, mouthwashes and toothpaste for oral application to control infections in the mouth and throat.

The ointments, creams or jellies of the invention may comprise a compound of the aforementioned general formula in association with a suitable ointment base such as for example (a) a water-immiscible type which may comprise soft paraffin or low melting fatty alcohols, (b) water-miscible types which may comprise those with a self-emulsifying base, (c) the aqueous jelly type in which an aqueous solution is thickened by means of a suitable agent, for example gum tragacanth, and (d) the emulsified type in which the emulsion may be a water-in-oil or oil-in-water system and which may contain thickening

agents such as suitable waxes. The lotions of the invention may comprise the active ingredient dissolved in an aqueous or aqueous alcohol base. Hair creams which may comprise oil-in-water or water-in-oil systems, may also contain a compound of the aforementioned general formula in accordance with the invention.

Preparations which may be used to produce aerosols containing a compound of the aforementioned general formula may comprise a solution of active ingredient in a suitable solvent in association with a volatile propellant, for example, Freon (registered trademark) propellant which is a chlorofluoroalkane mixture.

It will be appreciated that only the basic ingredients of the aforementioned compositions for topical use have been detailed, and that such compositions may in addition contain other conventional additives, such as stabilising agent, emulsifying agents, odorants and perfumes.

It is to be understood that the above descriptions of types of formulation are not intended to be limiting but are given by way of example only.

Some of the compounds of the aforementioned general formula, for example 1-bromo-1-nitropentane-2-ol, exhibit a tendency to be unstable in the presence of water. Accordingly, compositions in which the pharmaceutically acceptable diluent or carrier is nonaqueous are beneficial in providing antibacterial and antifungal compositions which are stable over long periods.

Compositions of the invention may, if desired, contain other antibacterial or antifungal agents in addition to the compounds of the above general formula. Thus, for example, the compositions may contain hexachlorophene, cetrimide, cetylpyridinium chloride, or an antibiotic such as neomycin. A particularly advantageous composition of the invention is an antibacterial cream in the form of an oil-in-water emulsion comprising 2-bromo-2-nitropane-1,3-diol, cetyl-trimethyl-ammonium bromide, glyceryl monostearate (non-self-emulsifying) and liquid paraffin. The compositions of the invention may also contain, if desired, other therapeutic agents. Thus for example, in the treatment of some clinical conditions, compositions containing a steroid medicament in addition to a compound of the aforementioned general formula may be indicated. Eardrops containing prednisolone are an example.

(B) COMPOSITIONS EMPLOYING THE COMPOUNDS AS HEREINBEFORE DEFINED AS PRESERVATIVES

The wide spectrum antibacterial and antifungal activity of the hereinbefore defined compounds renders them of value as preservatives, for example in medical and veterinary preparations.

The compositions according to this aspect of the invention comprise a compound as hereinbefore defined in association with a medical or veterinary preparation, and the pharmaceutically acceptable diluents or carrier within the scope of the invention accordingly comprise any medical or veterinary preparation which it is desired to preserve. Thus, as another feature of the invention there is provided a method of preserving medical and veterinary preparations from microbial deterioration which comprises incorporating in the said preparations a nitroalkanol of the above general formula. A nitroalkanol concentration of about 0.005–0.1%, preferably about 0.005–0.05% is generally suitable. As previously stated the term "medical and veterinary preparations" includes compositions which are normally designated as toilet or cosmetic preparations. A valuable feature of the compounds of the aforementioned general formula is their activity against bacteria and fungi in preparations of a slightly alkaline pH, say 7–9. Such preparations are difficult to preserve using the conventional preservatives, for example methyl hydroxybenzoate, chloroxylenol, chlorocresol and benzoic acid, etc. Typical preparations notoriously difficult to preserve from deterioration are the laxatives based on magnesium hydroxide, antacids based on aluminium

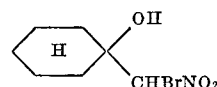
hydroxide, and shampoos based on anionic or nonionic surface active agents.

The previously defined nitroalkanols have also been found to be of value for the preservation of fish. Thus according to a further feature of the invention there is provided a method for preserving fish from microbial deterioration which comprises treating the fish with a nitroalkanol of the previously defined general formula. In this connection the high activity of the compounds against Gram-negative bacteria such as *Pseudomonas* spp, is extremely important. A preferred procedure for preserving fish is to treat the fish with ice containing a nitroalkanol as hereinbefore defined. Thus a composition provided according to this aspect of the present invention is ice containing a nitroalkanol of the previously defined general formula. This composition can be prepared by freezing an aqueous solution of the appropriate nitroalkanol. Preferably, a thickening agent such as gelatine or a suitable dispersing agent is included in the aqueous solution in order to enhance the amount of active ingredient incorporated into the ice during freezing and to promote its uniform distribution. Ice containing about 20–70 p.p.m., preferably about 40–50 p.p.m., of the hereinbefore defined nitroalkanols, particularly 2-bromo-2-nitropropane-1,3-diol, is generally suitable.

Table 1 is a list of examples of compounds of the above general formula which may be used in accordance with the present invention but is not to be interpreted as limiting the invention in any way.

TABLE 1

- (A) $C_2H_5CH(OH)CHBrNO_2$
- (B) $CH_2(OH)CBrNO_2CH_2OH$
- (C) $nC_3H_7CH(OH)CHBrNO_2$
- (D) $CH_3CH(OH)CBrNO_2CH_2OH$
- (E) $CH_3CH(OH)CHBrNO_2$
- (F) $(CH_3)_2CHCH(OH)CBr_2NO_2$
- (G) $nC_5H_{11}CH(OH)CHBrNO_2$
- (H) $nC_6H_{13}CH(OH)CHBrNO_2$
- (I) $CH_2(OH)CNO_2BrCH_3$



- (J) $CH_3CH(OH)CBr_2NO_2$
- (K) $C_{11}H_{23}CH(OH)CHBrNO_2$
- (L) $CH_2(OH)CHBrNO_2$
- (M) $nC_4H_9CH(OH)CHBrNO_2$
- (N) $C_2H_5CH(OH)CNO_2BrCH_3$
- (O) $CH_2(OH)CNO_2BrC_2H_5$
- (P) $C_2H_5CH(OH)CBr_2NO_2$

Although we have found that compounds of the foregoing general formula as a class possess outstanding wide-spectrum antibacterial and antifungal activity, there are, as is to be expected, variations between the activities of the individual members of the class. Details of the in vitro activity of particularly active compounds from the above list against some typical Gram-positive bacteria, Gram-negative bacteria, and fungi are given in Table 2. Minimum inhibitory concentrations (m.i.c.) were determined as follows. Solutions of the compounds in water or acetone were diluted with melted nutrient agar using a three-fold dilution series from 50 μ g./ml. The surface of the nutrient agar was inoculated with 0.001 ml. amount of overnight broth cultures of the test bacteria and fungal spore suspensions using a multipoint inoculator. The lowest concentrations of compounds completely inhibiting growth were noted after 18 hours incubation at 37° C., for the bacteria and after 4 days at 26° C. for the fungi. The micro-organisms tested were:

- (1) *Staphylococcus aureus*,
- (2) *Proteus vulgaris*,
- (3) *Pseudomonas pyocyanea*,
- (4) *Shigella sonnei*,

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(5) *Canadida albican*,(6) *Trichophyton mentagrophytes*.

TABLE 2

Compound:	M.I.C. ($\mu\text{g./ml.}$)					
	1	2	3	4	5	6
A-----	50	50	50	50	16.6	16.6
B-----	16.6	16.6	16.6	16.6	50	16.6
C-----	16.6	16.6	16.6	16.6	50	5.5
D-----	16.6	16.6	16.6	16.6	50	16.6
E-----	16.6	16.6	16.6	16.6	16.6	16.6
F-----	16.6	5.5	5.5	16.6	50	16.6
G-----	50	50	50	50	50	16.6
H-----	50	16.6	16.6	50	16.6	5.5
I-----	16.6	16.6	50	16.6	50	16.6
J-----	50	50	50	50	50	16.6

Previous remarks in this specification concerning the amounts of the hereinbefore defined compounds to be used in accordance with the present invention relate particularly to the above compounds A-J.

Some of the compounds A-J, for example compounds A, C, E and F have a tendency to be unstable in the presence of water and are accordingly preferably selected for use in nonaqueous systems. Compounds B and D are substantially stable in the presence of water and are accordingly preferred compounds. Compound C, 1-bromo-1-nitropentan-2-ol, is a preferred compound for use in non-aqueous systems. Compound B, 2-bromo-2-nitropropane-1,3-diol is the especially preferred compound for use in accordance with the present invention.

Compounds F and H, i.e., 1,1-dibromo-3-methyl-1-nitrobutan-2-ol and 1-bromo-1-nitro-octan-2-ol are new compounds. They may be prepared by reacting the sodium salts of the corresponding nitroalkanols with bromine, preferably using an inert organic liquid such as ether as the reaction medium. The required sodium salts may be prepared from the nitroalkanols by reaction with sodium methoxide in methanol.

The following non-limitative examples illustrate the invention.

EXAMPLE 1

Compositions for the treatment of hemorrhoids containing the following ingredients were prepared.

(a) Ointment:	Percent w./w.
Ephedrine hydrochloride -----	0.25
Allantoin -----	0.5
Lignocaine hydrochloride -----	0.5
2-bromo-2-nitropropane-1,3-diol -----	0.2
White soft paraffin -----	90.0
Wool fat -----	5.0
Water -----	3.55
(b) Suppositories:	
Ephedrine hydrochloride -----	0.25
Allantoin -----	0.5
Lignocaine hydrochloride -----	0.5
2-bromo-2-nitropropane-1,3-diol -----	0.2
Water -----	4.0
Fatty ester suppository base to 100.0.	

The ephedrine hydrochloride, lignocaine hydrochloride and 2-bromo-2-nitropropane-1,3-diol were dissolved in the water. This solution was blended with the remaining ingredients and the mixture homogenized.

EXAMPLE 2

Creams in the form of oil-in-water emulsions suitable for treating infected skin lesions were prepared from the ingredients listed below. The method employed was to add a mixture of the glyceryl monostearate and liquid paraffin at 55° C. to a stirred aqueous solution of the rest of the ingredients at 50° C.

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(a)

Percent w./w.

2-bromo-2-nitropropane-1,3-diol -----	0.1
Cetyl-trimethyl-ammonium bromide B.P. -----	0.5
5 Glyceryl monostearate (non-self-emulsifying) -----	10.0
Liquid paraffin -----	30.0
Water to 100.0.	

(b)

10 2-bromo-2-nitropropane-1,3-diol -----	0.1
Cetyl-trimethyl-ammonium bromide B.P. -----	0.5
Cetylpyridinium chloride B.P. -----	0.2
Glyceryl monostearate (non-self-emulsifying) -----	10.0
Liquid paraffin -----	30.0
15 Aqueous phosphate buffer solution (pH 5.0) to 100.0.	

EXAMPLE 3

Ointments (a), (b) and (c) were prepared containing 0.1% w./w. of the following active ingredients in a base consisting of Emulsifying Ointment, B.P.

- (a) 2-bromo-2-nitropropane-1,3-diol
 (b) 1-bromo-1-nitropentan-2-ol
 (c) 1,1-dibromo-3-methyl-1-nitrobutan-2-ol.

EXAMPLE 4

Ointments were prepared containing the following ingredients:

(a)

Percent w./w.

30 1-bromo-1-nitropentan-2-ol -----	0.2
Oleyl alcohol -----	5.0
White soft paraffin to 100.0.	

(b)

35 1-bromo-1-nitropentan-2-ol -----	0.1
Liquid paraffin -----	5.0
White soft paraffin to 100.0.	

Similar preparations to (a) and (b) were prepared substituting 2-bromo-2-nitropropane-1,3-diol for 1-bromo-1-nitropentan-2-ol.

EXAMPLE 5

Solutions for use as eardrops were prepared containing the following ingredients:

(a)

Percent w./w.

45 1-bromo-1-nitropentan-2-ol -----	0.2
Isopropyl myristate to 100.0.	

(b)

50 1-bromo-1-nitropentan-2-ol -----	0.2
Prednisolone -----	0.5
Isopropyl myristate to 100.0.	

Similar preparations to (a) and (b) were prepared substituting 2-bromo-2-nitropropane-1,3-diol for 1-bromo-1-nitropentan-2-ol.

EXAMPLE 6

2-bromo-2-nitropropane-1,3-diol was used as a preservative in the following compositions:

(a)

Percent w./w.

65 2-bromo-2-nitropropane-1,3-diol -----	0.02
Mist. Mag. Hydrox. B.P. to 100.0.	

(b)

70 2-bromo-2-nitropropane-1,3-diol -----	0.02
Emulsion of liquid paraffin and Mist. Mag. Hydrox. B.P. to 100.0.	

(c)

2-bromo-2-nitropropane-1,3-diol -----	0.02
Magnesium hydroxide -----	1.6
Flavoring, colouring and dispersing agents -----	0.3
75 Aluminium hydroxide gel B.P.C. to 100.0.	

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(d)

Maize oil B.P.C. -----	48.0
Acacia -----	10.0
Flavouring -----	0.4
2-bromo-2-nitropropane-1,3-diol -----	0.02
Water to 100.0.	

(e)

Aluminium hydroxide paste equivalent to 2.0% Al_2O_3	
Kaolin -----	20.0
2-bromo-2-nitropropane-1,3-diol -----	0.02
Flavouring, colouring and dispersing agents -----	20.4
Water to 100.0.	

EXAMPLE 7

A medicated dusting powder was prepared containing the following ingredients:

	Percent w./w.
1-bromo-1-nitropentan-2-ol -----	0.2
Magnesium oxide -----	0.5
Nongelling starch to 100.0.	

EXAMPLE 8

A shampoo incorporating 2-bromo-2-nitropropane-1,3-diol as a preservative was prepared from the following ingredients:

	Percent w./w.
Triethanolamine lauryl sulphate -----	20.0
Lauric isopropanolamide -----	3.0
2-bromo-2-nitropropane-1,3-diol -----	0.02
Perfume and colouring -----	0.1
Distilled water to 100.0.	

EXAMPLE 9

A toothpaste incorporating 2-bromo-2-nitropropane-1,3-diol as a preservative was prepared from the following ingredients:

	Percent w./w.
Dicalcium phosphate -----	48.0
Sodium lauryl sulphate -----	1.5
Glycerine -----	30.0
Sodium carboxymethylcellulose -----	0.7
Saccharin -----	0.1
Flavouring -----	0.1
2-bromo-2-nitropropane-1,3-diol -----	0.02
Distilled water to 100.0.	

EXAMPLE 10

Water containing 50 p.p.m. 2-bromo-2-nitropropane-1,3-diol and 100 p.p.m. gelatin was frozen to form blocks of ice suitable for preserving fish.

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We claim:

1. A method of combatting aerobic pathogenic bacteria on the surface of the skin which comprises topically applying to the skin a non-toxic pharmaceutically acceptable composition comprising an antibacterially effective amount of 2-bromo-2-nitropropane-1,3-diol incorporated in a pharmaceutically acceptable topically applicable carrier.

2. The method of claim 1 wherein the pharmaceutically acceptable composition comprises from 0.005 to 0.1% by weight of 2-bromo-2-nitropropane-1,3-diol.

3. The method of claim 1 wherein the pharmaceutically acceptable composition comprises from 0.005 to 0.1% by weight of 2-bromo-2-nitropropane-1,3-diol incorporated in a pharmaceutically acceptable ointment base.

4. The method of claim 3 wherein the ointment base is an oil-in-water emulsion ointment base.

5. A method of controlling aerobic pathogenic gram-negative bacteria on the surface of the skin which comprises topically applying to the skin an antibacterially effective amount of 2-bromo-2-nitropropane-1,3-diol incorporated in a pharmaceutically acceptable, topically applicable carrier.

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