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THIOGLYCEROL AND FORMALDEHYDE SULFOXYLATE STABILIZED TETRACYCLINE ANTIBIOTICS IN POLYHYDRIC ALCOHOL SOLVENTS

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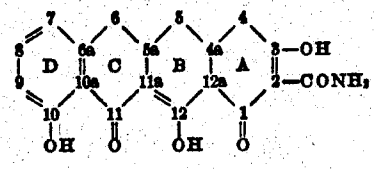
This application relates to antibiotic solutions and more particularly to stabilized solutions of the tetracycline antibiotics.

As is well known, these antibiotics are highly valuable in the treatment of disease in man and animals. In many cases it is desirable to formulate them in solution form, primarily for parenteral use, but also for oral and topical administration. Like many organic substances, however, the tetracycline antibiotics are particularly subject to oxidative decomposition in solution with the formation of highly colored impurities which render them unsuitable for the desired applications. As a result it has usually been necessary to prepare these dosage forms in a dry state for reconstitution with a solvent immediately before use. In some cases it has been possible to eliminate this procedure with its obvious attendant disadvantages by the judicious addition of certain antioxidants and the careful packaging of the solution in sealed ampoules under nitrogen gas. Thus, for example, some success has been achieved in this direction by the addition of sodium formaldehyde sulfoxylate to the antibiotic solution. However, even with this valuable additive it has not been practical to package these solutions in multi-dose containers but has rather been necessary to provide individual single-dosage ampoules. This inconvenient and expensive procedure is required because the removal of individual dosages from a multi-dose package (for example, by piercing a rubber bottle closure with a hypodermic needle, injecting air and withdrawing an individual dosage into a syringe) necessarily introduces air into the container beyond the ability of the sulfoxylate to stabilize the residual solution, which soon darkens and becomes unfit for use.

Now it has been discovered that these problems are obviated by stabilizing solutions of the tetracycline antibiotics with a combination of an alkali-metal or alkaline-earth-metal formaldehyde sulfoxylate and thioglycerol (α-monothioglycerol). With this antioxidant combination it is now possible for the first time to package already constituted solutions of the tetracycline antibiotics in multi-dose containers with the assurance that even after a substantial proportion of air has been admitted to the container during partial withdrawal of the liquid contents, the residual solution will remain stable and light in color for long periods of storage. The savings in cost and space made possible by this development are obvious.

The tetracycline antibiotics comprise a group of biologically active perhydronaphthacene derivatives having the following essential structural features. The numbering system indicated is that employed by "Chemical Abstracts."



Among the biologically active members of this series are those containing the following substituent groups.

Substituents	Common Name
4-N(CH3)2, 6-OH, 6-CH3, 12a-OH	tetracycline.
4-N(CH3)2, 5-OH, 6-OH, 6-CH3, 12a-OH	5-oxytetracycline.
4-N(CH3)2, 6-OH, 6-CH3, 7-Cl, 12a-OH	7-chlortetracycline.
4-N(CH3)2, 6-OH, 12a-OH	6-demethyltetracycline.
4-N(CH3)2, 6-OH, 7-Br, 12a-OH	6-demethyl-7-bromtetracycline.
4-N(CH3)2, 6-OH, 6-CH3	12a-deoxytetracycline.
4-N(CH3)2, 5-OH, 6-CH3, 12a-OH	6-deoxy-5-oxytetracycline.
5-OH, 6-CH3, 12a-OH, 6-OH	4-desdimethylamino-5-oxytetracycline.
4-N(CH3)2, 6-CH3, 12a-OH	6-deoxytetracycline.
4-N(CH3)2, 12a-OH	6-deoxy-6-demethyltetracycline.
4-N(CH3)2, 6-OH, 6-CH3, 7-Br, 12a-OH	7-bromotetracycline.
4-N(CH3)2, 6-OH, 7-Cl, 12a-OH	6-demethyl-7-chlortetracycline.
4-N(CH3)2, 6-CH3	6, 12a-dideoxytetracycline.
6-OH, 6-CH3, 12a-OH	4-desdimethylaminotetracycline.
6-OH, 6-CH3, 7-Cl, 12a-OH	4-desdimethylamino-7-chlortetracycline.
4-N(CH3)2, 6-OH	6-demethyl-12a-deoxytetracycline.
4-N(CH3)2, 6-CH3, 7-Cl, 12a-OH	6-deoxy-7-chlortetracycline.
4-N(CH3)2, 7-Cl, 12a-OH	6-deoxy-6-demethyl-7-chlortetracycline.
4-N(CH3)2, 6-CH3, 7Br, 12a-OH	6-deoxy-7-bromtetracycline.
4-N(CH3)2, 7-Br, 12a-OH	6-deoxy-6-demethyl-7-bromtetracycline.

Some of the aforementioned tetracycline derivatives are well known in the art, while others are described in copending applications Serial No. 773,172, filed November 12, 1958, and Serial No. 802,655, filed March 30, 1959, both assigned to the same assignee as the present invention.

Forms of the tetracycline antibiotics which are particularly suitable for solution administration are the coordination complexes which are formed with metal salts. For example, in copending application Serial No. 669,460, filed July 2, 1957, and assigned to the same assignee as the present invention, there are described compounds of oxytetracycline containing magnesium, calcium, zinc, aluminum, or combinations thereof, and compounds of tetracycline containing magnesium, calcium, aluminum or combinations thereof. In particular there are disclosed magnesium, calcium, and aluminum tetracyclines having a molar ratio of metal ion to tetracycline of about 3:1, and magnesium, calcium, zinc, and aluminum oxytetracyclines having a molar ratio of metal ion to oxytetracycline of from about 1:3 to about 3:1. As disclosed in that copending application, useful solvents are the polyhydric aliphatic alcohols and mixtures thereof. Especially satisfactory are the glycols, preferably those generally recognized as pharmaceutically acceptable, such as propylene glycol, polyethylene glycol and mixtures thereof. Glycerine is another example of a polyol which is particularly useful. Up to about 25–30% by volume of water may be incorporated in the vehicle if desired. 80% aqueous propylene glycol has been found to be a particularly convenient solvent system.

According to one preferred method of preparing such solutions, the chosen metal salt of the antibiotic is prepared in situ by dissolving the free-base form of a tetracycline antibiotic, or an acid salt thereof, in the selected polyhydric alcohol and adding a solution of a salt of the desired metal in the polyol or in water. Of course it is necessary that a salt of a pharmaceutically acceptable anion be selected, that is, one which is free from objectional side effects at the levels of ordinary use. The chloride and acetate salts, for example, are among those which are suitable. The reaction usually occurs readily at room temperature. Generally neutral to alkaline conditions are desirable, e.g. a pH range of from about 5.0 to about 10.0, preferably from a pH 8.0 to 9.0 for optimum stability. pH may be controlled by addition of base as

required, and a particularly convenient base for present purposes is monoethanolamine.

Among the formaldehyde sulfoxylates which are suitable are the pharmaceutically acceptable alkali-metal and alkaline-earth-metal derivatives, including, for example, the potassium, lithium, calcium, magnesium and strontium salts. Particularly preferred is the readily available sodium formaldehyde sulfoxylate. (The barium derivative is toxic, and will, of course, not be employed.) With as little as 0.05% (w./v.) or even less of these antioxidants useful stabilization is achieved, but it is normally preferred to employ at least about 0.1% and ordinarily it will not be necessary to employ more than about 0.5% to achieve the desired results. Similarly, concentrations of thioglycerol of about 0.1% (w./v.) or even less are beneficial but for optimum results at least about 0.2% will be preferred and normally no more than about 2% will be required.

These new therapeutic combinations are particularly suitable for parenteral administration and especially for intramuscular use. For such application, it will often be desirable to incorporate a local anaesthetic such as those which are well known to those skilled in the art. For example, lidocaine (α-diethylamino-2,6-acetoxylidide, available from the Astra Chemical Co.), may be employed at a level of about 20 mg./cc. For intramuscular use the preparations will be formulated with pyrogen-free water and filtered aseptically before packaging. Antibiotic concentrations which are particularly suitable for this mode of administration are those ranging from about 50 to about 125 mg./cc.

In addition to the parenteral applications, these stabilized solutions are also eminently suitable for oral and topical use, and may be employed, for example, in the treatment of bovine mastitis.

With the newly discovered stabilizing system, solutions of tetracycline antibiotics are found to retain their original color and potency substantially unimpaired over long periods of storage at room temperature, and also in accelerated high-temperature experiments. For optimum results it is desirable to saturate the vehicle with nitrogen and to package these dosage forms under an inert atmosphere. However, single doses may be withdrawn from multi-dose containers with attendant admission of air into the container without seriously impairing the stability of the remaining solution. For example, in accelerated tests at 50° C. for 12 days containers from which 80% of the solution has been withdrawn by hypodermic syringe retain the residual solution with little change in color or biological activity, so that it is still completely suitable for use.

These new formulations are suitable for administration to man and animals at conventional dosage levels for treatment of infections due to microorganisms sensitive to the contained antibiotic. For example, in the case of oxytetracycline solutions, intramuscular administration of 200–300 mg. of antibiotic daily is usually satisfactory in the case of mild or moderately severe infections. For more severe infections 300–500 mg. daily may be required. Administration in divided doses 2–3 times daily is often desirable. It has been found that no untoward side effects or impairment of potency attributable to the formaldehyde sulfoxylate or the thioglycerol are encountered.

The following examples are given by way of illustration and should not be interpreted as limiting the invention, the scope of which is defined by the appended claims.

EXAMPLE I

Oxytetracycline base (910 mcg./mg. potency), gm.	30.22
Magnesium chloride hexahydrate, gm.	12.36
Monoethanolamine, cc.	8.85
Propylene glycol, gm.	376
Water, cc.	94

The glycol is agitated for one hour while saturating with nitrogen gas, and the antibiotic is then added and the mixture stirred for 30 minutes more. Next, a solution of the magnesium chloride in the nitrogen-saturated water is slowly added. After 5 minutes' further stirring the amine is added. Throughout the addition of ingredients the tempertaure is maintained below 30° C. by cooling as required. Solution is completed by stirring under nitrogen. The resulting 500 cc. of clear, light-colored solution contains approximately 50 mg./cc. of oxytetracycline activity and has a pH of about 8.6. It is subdivided and to each portion an antioxidant is added, followed by stirring under nitrogen for 20 minutes. Samples are filled into ampoules under nitrogen and sealed. The antioxidant concentrations are as follows:

A. 0.3% w./v. sodium formaldehyde sulfoxylate plus 1.0% w./v. thioglycerol
B. 0.5% w./v. sodium formaldehyde sulfoxylate
C. 1.0% w./v. thiomalic acid

EXAMPLE II

Samples of the solutions of Example I are subjected to a temperature of 75° C. for varying periods of time up to 24 hours. After heating, each sample is diluted 1:1 with the aqueous glycol vehicle and the light absorbance is determined at 500 mμ on a Beckman spectrophotometer with a tungsten light source and a slit width setting of 0.04, employing water as the blank. Results are summarized in the table below.

Light Absorbance

	A	B	C
Initial	.445	.495	.506
2 hours at 75° C.	.426	.522	.815
4	.480	.555	∞
6	.559	.605	
8	.684	∞	
24	.830		

The relatively high color stability of solution A is apparent.

EXAMPLE III

Solutions A and B whose preparation is described in Example I are filled under nitrogen into 10 cc. multi-dose vials sealed with butyl rubber stoppers. Solution A, initially lighter in color than solution B, maintains this color even after heating at 50° C. for 12 days.

Two-cc. doses are withdrawn from fresh (unheated) vials of each solution in the standard manner, by piercing the stopper with a hypodermic needle, injecting air and withdrawing the solution into the syringe. In this manner, four such doses are withdrawn from each vial, leaving one 2-cc. dose in the stoppered vial. The residual solution A contained in the vial under a partial atmosphere of air darkens only slightly even after heating for 12 days at 50° C. In contrast, the residual solution B begins darkening within two days at room temperature, and after 12 days at room temperature is dark brown in color.

EXAMPLE IV

A stabilized preparation containing approximately 125 mg./cc. of oxytetracycline activity and suitable for intramuscular administration is prepared according to the following formulation and packaged under nitrogen:

Oxytetracycline base, gm.	75.6
Magnesium chloride hexahydrate, gm.	30.9
Monoethanolamine, cc.	22.1
Lidocaine, gm.	10
Thioglycerol, gm.	5
Sodium formaldehyde sulfoxylate, gm.	1.5
Propylene glycol, gm.	275
Water, cc.	113

Similar formulations are prepared substituting other tetracycline antibiotics, as hereinbefore described, in place of oxytetracycline, and enhanced stability attributable to the thioglycerol and sodium formaldehyde sulfoxylate is observed.

EXAMPLE V

The following pharmaceutical solutions are prepared and found to exhibit enhanced stability attributable to their content of thioglycerol and a formaldehyde sulfoxylate:

(A)

Glycerine, U.S.P., liter	1
Calcium acetate monohydrate, gm.	20.66
Oxytetracycline hydrochloride, gm.	60.2
Potassium formaldehyde sulfoxylate, gm.	2
Thioglycerol, gm.	4
10% sodium hydroxide in glycerine, to pH 9, cc. approx.	150
Glycerine, U.S.P., to make 2 liters.	

(B)

Gylcerine, U.S.P., cc.	500
Aluminum chloride hexahydrate, gm.	40.7
Tetracycline hydrochloride, gm.	27.55
Thioglycerol, gm.	20
Lithium formaldehyde sulfoxylate, gm.	5
10% sodium hydroxide in glycerine, to pH 9	--
Glycerine, U.S.P., to make 1 liter.	

(C)

Glycerine, U.S.P., cc.	500
Sorbitol, gm.	50
Zinc chloride, gm.	5.94
Oxytetracycline hydrochloride, gm.	33.5
Thioglycerol, gm.	10
Sodium formaldehyde sulfoxylate, gm.	3
10% sodium hydroxide in glycerine, to pH 8.5, cc. approx.	72
Glycerine, U.S.P., to make 1 liter.	

(D)

Polyethylene glycol 300, cc.	500
Magnesium chloride tetrahydrate, gm.	28.19
Tetracycline hydrochloride, gm.	27.55
Thioglycerol, gm.	10
Sodium formaldehyde sulfoxylate, gm.	3
Triethanolamine, cc.	25
10% sodium hydroxide in glycerine, to pH 8.5, cc. approx.	125
Polyethylene glycol 300, to make 1 liter.	

(E)

Propylene glycol, cc.	800
Benzocaine, gm.	51
Calcium chloride, anhydrous, gm.	3.46
Tetracycline hydrochloride, gm.	5.1
Thioglycerol, gm.	2

Calcium formaldehyde sulfoxylate, gm.	1
10% sodium hydroxide in propylene glycol, to pH 8.5, cc. approx.	15
Propylene glycol, to make 1 liter.	

(F)

Glycerine, U.S.P., cc.	500
70% aqueous sorbitol, gm.	100
Aluminum chloride hexahydrate, gm.	24
Oxytetracycline base, gm.	46
Ethanol, cc.	80
Thioglycerol, gm.	5
Strontium formaldehyde sulfoxylate, gm.	2
10% sodium hydroxide in glycerine, to pH 8	--
Glycerine, U.S.P., to make 1 liter.	

(G)

Glycerine, U.S.P., cc.	500
Calcium acetate monohydrate, gm.	20.66
Oxytetracycline hydrochloride, gm.	30.1
Thioglycerol, gm.	10
Magnesium formaldehyde sulfoxylate, gm.	3
10% sodium hydroxide in glycerine, to pH 9	--
Glycerine, U.S.P., to make 1 liter.	

What is claimed is:

1. A pharmaceutical composition comprising a tetracycline antibiotic dissolved in a pharmaceutically acceptable polyhydric alcohol solvent, and as stabilizer for said tetracycline antibiotic, a mixture of thioglycerol and a pharmaceutically acceptable formaldehyde sulfoxylate selected from the group consisting of the alkali-metal and alkaline-earth-metal salts, the weight ratio of sulfoxylate salt to thioglycerol being from about 5:2 to about 1:20.

2. A composition as in claim 1 wherein said antibiotic is selected from the group consisting of magnesium, calcium and aluminum tetracyclines having a molar ratio of metal ion to tetracycline of about 3:1 and magnesium, calcium, aluminum and zinc oxytetracyclines having a molar ratio of metal ion to oxytetracycline of from about 1:3 to about 3:1.

3. A composition as in claim 1 wherein said solvent contains up to about 30% by volume of water.

4. A composition as in claim 1 wherein said formaldehyde sulfoxylate is present in a weight/volume concentration ranging from about 0.1 to about 0.5% and said thioglycerol is present in a weight/volume concentration ranging from about 0.2 to about 2%.

5. A pharmaceutical composition suitable for intramuscular administration comprising a solution containing magnesium oxytetracycline having a molar ratio of magnesium to oxytetracycline of about 1, about 0.3% w./v. sodium formaldehyde sulfoxylate and about 1% w./v. thioglycerol in about 80% aqueous propylene glycol.

References Cited in the file of this patent

UNITED STATES PATENTS

2,719,812 Hanus ..... Oct. 4, 1955