United States Patent Office

3,062,712
Patented Nov. 6, 1962

3,062,712
PREDNISOLONE SESQUIHYDRATE COMPOSITIONS

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No Drawing. Filed Nov. 9, 1959, Ser. No. 851,549
8 Claims. (Cl. 167—53)

This invention relates to a novel composition of matter, a process for the preparation thereof and to stable aqueous suspensions containing said novel composition. More particularly it relates to a novel prednisolone sesquihydrate which is stable in aqueous suspension, to a process for preparing said sesquihydrate and to stable aqueous suspensions prepared therefrom.

Stable aqueous suspensions of crystalline ingredients are truly a boon to the pharmaceutical manufacturer and the professional users of such suspensions. The manufacturer is able to prepare and supply aqueous suspensions which are free from the deleterious properties of crystal growth and aggregation. Unless aqueous pharmaceutical suspensions are free from these deleterious properties, the professional user faces major difficulties. He often finds that a suspension for injection has settled or sedimented to such an extent that resuspension is difficult if not impossible, and that accurate, uniform doses cannot be administered. He also finds that incomplete drainage from the container prevents full utilization of the contents. Syringes and needles often become clogged because particle size has increased due to crystal growth. Moreover, increased particle size in injectable suspensions can often result in pain to the patient. In the case of aqueous suspensions for use in the eye, ear or nose, crystal growth and aggregation cause related difficulties such as scratching and irritation of sensitive body surfaces and mucous membranes.

The preparation and use of aqueous suspensions of prednisolone have been accompanied by a series problem of crystal growth. In injectable aqueous suspensions the particle size of the crystals has been found to increase to such an extent that resuspending and related difficulties have resulted. Similarly, an increase in particle size in aqueous suspensions for use in the eye, ear or nose has rendered such suspensions unsuitable for use.

It is an object, therefore, of the present invention to provide a novel composition of matter which has the therapeutic activity of prednisolone but which is stable in aqueous suspensions. An additional object is to provide such a novel composition of matter which is not subject to excessive crystal growth in aqueous suspensions. A further object is to provide a process for the preparation of said novel composition of matter. A still further object is to provide aqueous suspensions containing said novel composition of matter which are stable and suitable for injection. A still further object is to provide aqueous suspensions containing prednisolone activity which are admirably suited for use in the eye, ear or nose. Other objects will be apparent to those skilled in the art to which this invention pertains.

These and additional objects have been accomplished by the provision of prednisolone sesquihydrate having the formula [$\Delta^{1,4}$ - pregnadiene - 3,20-dione-11β,17α,21-triol)$_2\cdot(H_2O)_3$], having $[\alpha]_D^{25}$ +95 +99 in dioxane,

showing retarded crystal growth in aqueous suspensions, and having interplanar spacings, obtained by X-ray diffraction using nickel-filtered copper Kα radiation, of

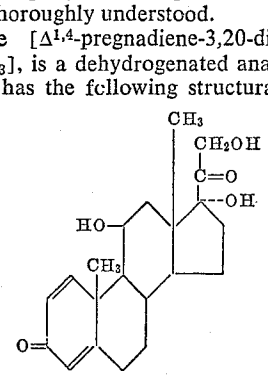
A.		
6.17	4.35	3.47
5.73	4.18	3.33
5.05	3.90	3.04
4.70	3.74	

by the provision of a process of crystallizing the prednisolone sesquihydrate from aqueous-methanol; and by the provision of stable aqueous suspensions comprising the prednisolone sesquihydrate in vehicles suitable for injection, or for use in the eye, ear or nose.

In the course of studies on the crystal growth of various preparations of prednisolone it was unexpectedly found that the product from an aqueous-methanol crystallization process did not exhibit crystal growth in various aqueous suspensions. This was unexpected in view of the deleterious crystal growth properties exhibited by preparations of crystalline prednisolone from other processes, e.g., from an aqueous-acetone process. Upon examination is was discovered that the product of the aqueous-methanol crystallization process was a novel prednisolone sesquihydrate.

The reasons for the failure of the crystals of this novel prednisolone sesquihydrate to grow in aqueous suspensions are not thoroughly understood.

Prednisolone [$\Delta^{1,4}$-pregnadiene-3,20-dione-11β,17α,21-triol)$_2\cdot(H_2O)_3$], is a dehydrogenated analogue of hydrocortisone. It has the following structural formula:



It can be prepared by methods known in the art such as the method reported by Hogg et al., Journal of the American Chemical Society 77, page 4438, August 20, 1955.

The novel prednisolone sesquihydrate of the present invention can be prepared by dissolving prednisolone in aqueous-methanol, filtering if necessary to remove any insolubles, cooling to produce crystallization, filtering off the crystals, and drying said crystals, preferably to a water content of from about two to about eight percent by weight. The theoretical water content is 6.97 percent by weight; however, water contents above and below the theoretical percent are operable. A water content higher than about eight percent by weight causes wet crystals which present difficulties in handling such as in weighing and pouring. Crystals approaching the anhydrous condition are likely to grow in size in aqueous suspension. Therefore, a lower limit of water content of about two percent is preferred. It is preferred to carry out the drying by using a controlled temperature under vacuum; however, the drying can be carried out at atmospheric pressure if desired. In either case a tempera-

ture above about forty degrees centigrade should be avoided. For incorporation into suspensions the dried prednisolone sesquihydrate is comminuted to a particle size suitable for injectable and ophthalmic aqueous suspensions, preferably by micronization. A high percent of particles less than about 20 microns in size, for example, 99 percent with 90 percent less than about 10 microns, is obtained by controlling the micronization technique. However, a particle size of about twenty microns is also suitable in aqueous suspensions in that settling and sedimentation during storage are minimized and resuspendibility, container drainage and syringeability are very satisfactory. A particle size above about twenty microns usually results in a less satisfactory suspension. Subsequently, the comminuted prednisolone sesquihydrate is sterilized, e.g., by ethylene oxide gas, and incorporated into suitable aqueous vehicles using aseptic pharmaceutical techniques.

The concept and practice of the instant invention do not depend thereon, but nevertheless additional complementary active ingredients can be incorporated into a suspension containing prednisolone sesquihydrate. Said additional active ingredients include antibiotics such as neomycin, penicillins such as penicillin G, penicillin O, procaine penicillin and the like, novobiocin, bacitracin, streptomycin, dihydrostreptomycin, chloramphenicol, erythromycin, tetracycline, endomycin, polymyxin, therapeutically active derivatives of any of the foregoing, and the like, or mixtures thereof. Utility in human therapy with compositions containing an antibiotic and the prednisolone sesquihydrate, however, has not been established. Other active ingredients can also be incorporated into said suspensions, for example, vitamins, more specifically vitamin A, vitamin B_1, riboflavin, pyridoxine, ascorbic acid, vitamin B_{12} and the like. Suspensions for nasal use can contain a vaso-constrictor, such as epinephrine, phenylephrine hydrochloride, and the like.

Pharmaceutical preservatives, buffers, isotonic agents, and suspending agents can be used in the suspensions of the invention. Suitable preservatives include chlorobutanol, myristyl gamma picolinium chloride, methyl and propylparabens and sodium ethylmercurithiosalicylate. Myristyl gamma picolinium chloride has useful surfactant characteristics in addition to being a preservative. Sodium chloride is preferred as an isotonic agent. Among the preferred suspending agents are polyethylene glycol 4000 or 6000, polyvinylpyrrolidone, dextran and methylcellulose. The polyvinylpyrrolidone is characterized by having a viscosity coefficient, i.e., a K value, of 26 to 36 and a molecular weight of about 40,000.

The compositions of the instant invention are useful in the medical and related arts where compositions comprising corticosteroids are indicated, especially for inflammatory and allergic diseases.

Crystal growth was studied using a micro technique test method in which a few crystals on a microscope slide were observed, then ground between two slides (using the thumb and finger in a rotary motion) to a powder 99 percent less than 20 microns. To this ground powder was then added one drop of vehicle and the grinding continued. When a satisfactory dispersion was obtained, the slides were separated and the suspension washed onto a clean slide with a second drop of vehicle. This was covered with a cover slip, the rim of which was thickly coated with desiccator grease. This cover slip was carefully pressed down to form a water-proof seal and finally the edge was sealed with Canada balsam in xylene. Such slides can usually be stored without excess drying for prolonged periods. Observations for crystal growth are conveniently made at 10 minutes, 1 hour, 1 day, 1 week, etc. The slides can be stored at 4° C. to provide quick results by accelerating any crystal growth which may occur.

Crystal growth studies of crystalline prednisolone sesquihydrate prepared by the aqueous-methanol process and crystalline prednisolone from an aqueous-acetone process were carried out with suspensions in deionized water. Table I gives the data found in these studies, using the test method detailed above.

TABLE I

Crystal Growth in Deionized Water

Appearance Under Microscope

Sample No.	Initial	1 hour (25° C.)	1 day (25° C.)
1 [1]	<10μ	50–70% rods 10–40μ	90% rods 10–50μ.
2 [2]	<10μ	90% rods 40–400μ	100% rods 800μ.
3 [1]	<10μ	50–70% rods 10–40μ	50–80% rods 10–30μ.
4 [1]	<10μ	50% rods 10μ	70–90% rods 10–40μ.
5 [2]	99%<20, 90%<10μ	No change	50% rods 10–40μ.

[1] Crystallized from aqueous-methanol.
[2] Crystallized from aqueous-acetone.

These data show that, even in plain deionized water, prednisolone sesquihydrate prepared by the aqueous-methanol process of the invention quickly shows a superior advantage over a conventional form prepared by an aqueous-acetone process, in being free from deleterious crystal growth.

Crystal growth studies were also carried out with suspensions of prednisolone sesquihydrate in an aqueous vehicle containing 12 percent polyethylene glycol 4000, 0.6 percent neomycin sulfate, 0.02 percent myristyl gamma picolinium chloride, 0.45 percent sodium citrate and 0.1 percent polyvinylpyrrolidone. Table II gives the data found in these studies, wherein the superior advantage of the sesquihydrate is more pronounced.

TABLE II

Crystal Growth in Aqueous Vehicle

Appearance Under Microscope

Sample No.	Initial	1 hour (25° C.)	1 day (25° C.)
1 [1]	99%<20, 90%<10μ	No change	No change.
2 [2]	99%<20, 90%<10μ	50% needles 10–30μ	90% needles, 10–30μ (clumped).
3 [1]	99%<20, 90%<10μ	No change	No change.
4 [1]	99%<20, 90%<10μ	do	Do.
5 [2]	99%≤20, 10%<10μ		50% rods 20–40μ (fused in large clusters).
6 [1]	99%<20, 90%<10μ	No change	No change.
7 [1]	99%<20, 90%<10μ	do	Do.

[1] Crystallized from aqueous-methanol.
[2] Crystallized from aqueous-acetone.

These data show the marked superiority of the new sesquihydrate of prednisolone when tested for crystal growth in this vehicle. No changes occurred in the crystal size of the new form, whereas excessive growth and caking occurred in the other form.

The successful preparation of prednisolone sesquihydrate by the novel aqueous-methanol crystallization process depends on a proper ratio of the amount of water present to the amount of prednisolone used. Sufficient water must be present to hydrate all of the prednisolone. An amount of water sufficient to create a practical problem of prednisolone solubility should be avoided. The preferred water content of the aqueous methanol solvent can vary from one to ten percent by volume. A higher water content is still operative. Higher concentrations of water increase the amount of solvent necessary for solution and crystallization. The upper limit of water content depends on the variations of processing arising from practical considerations of yield and the containers and apparatus required.

The following preparations and examples are illustrative of the processes and products of this invention, and are not to be construed as limiting.

EXAMPLE 1.—PREPARATION OF PREDNISOLONE SESQUIHYDRATE

1000 grams of prednisolone N.N.R. is dissolved in a mixture of 24,950 milliliters of methanol and 250 milliliters of distilled water in a round-bottomed flask adapted to be heated and refluxed as required. Boiling for a short time is usually required to effect complete solution. The hot solution is freed of any insolubles by filtration. Cooling at about four degrees centigrade for about sixteen hours produces crystallization of the prednisolone sesquihydrate. Seeding of the solution with sesquihydrate crystals may speed up crystallization but is not critical. The crystals are recovered by filtration and dried for approximately sixteen hours at about forty degrees centigrade under vacuum (28 inches of mercury). The dried crystals are comminuted to a particle size of about 20 microns. The resulting compound is tested by infrared spectrum to confirm the existence of the sesquihydrate. A yield of 849 grams is obtained.

EXAMPLE 2.—PREPARATION OF PREDNISOLONE SESQUIHYDRATE

Following the procedure of Example 1, 500 grams of prednisolone N.N.R. is dissolved in a mixture of 9000 milliliters of methanol and 1000 milliliters of distilled water. A yield of 445 grams of dry sesquihydrate is obtained. The crystals are tested by infrared spectrum to confirm the existence of the sesquihydrate.

The novel prednisolone hydrate was subjected to various chemical and physical tests to confirm the existence of the sesquihydrate. Table III gives the weight loss data compared with water determination by the Karl Fischer method.

TABLE III

Prednisolone sesquihydrate	Weight loss at 60° centigrade in vacuo, percent	Water determination (Karl Fischer method), percent
Lot 12, 034-2	6.68	6.43, 6.84
Lot 12, 035-2	6.24	6.29, 6.30
Lot GF 787	6.71	6.75, 6.75

The equivalence of the percent of weight loss and the percent of water by the Karl Fischer method establishes that the prednisolone sesquihydrate samples do not contain a stoichiometrically significant amount of methanol. The magnitude of the water content establishes the prednisolone hydrate to be the sesquihydrate, the theoretical water content for the sesquihydrate being 6.97 percent.

Moisture equilibration data afford further evidence that the sesquihydrate is an entity. A sample of the sesquihydrate was exposed at fifty percent relative humidity and 25 degrees centigrade for 36 hours with no change in weight, showing that no additional water beyond the sesquihydrate-equivalent amount was added. A second sample of the sesquihydrate was dried to constant weight and then exposed at fifty percent relative humidity and 25 degrees centigrade. The exposed material equilibrated with the water present and returned to the weight of the original sample.

The novel prednisolone sesquihydrate was tested by elemental analyses to confirm the elemental formula. Table IV contains the data.

TABLE IV

Prednisolone sesquihydrate	Calculated, percent		Found, percent	
	Carbon	Hydrogen	Carbon	Hydrogen
Lot 12, 034-2	65.09	8.06	65.21	8.26
Lot 12, 035-2	65.09	8.06	65.49	8.61
Lot GF 787	65.09	8.06	65.11	8.13

These data establish that the formula

$$(\text{prednisolone})_2 \cdot (H_2O)_3$$

is correct.

The novel prednisolone sesquihydrate and an anhydrous sample of prednisolone were examined by infrared analysis. Table V contains the data.

TABLE V

Infrared Absorption Bands of Prednisolone Sesquihydrate and Anhydrous Prednisolone

Sesquihydrate frequency	Anhydrous form frequency
cm.$^{-1}$	cm.$^{-1}$
3,500	3,450
3,360	3,370
1,703	1,709
1,655	1,650
1,615	1,610
1,597	1,594
1,410	1,412
1,350	1,395
1,303	1,347
1,270	1,322
1,240	1,300
1,203	1,278
1,195	1,232
1,165	1,220
1,130	1,137
1,105	1,152
1,051	1,135
1,039	1,110
1,002	1,092
946	1,059
933	1,038
890	1,002
857	976
823	950
812	925
763	900
737	886
	863
	815
	743
	700

These data establish the sesquihydrate to be an entity differing from the commercial anhydrous form heretofore available.

EXAMPLE 3.—AQUEOUS SUSPENSIONS FOR INJECTION

A vehicle containing the following ingredients:

	Grams
Sodium citrate	15.75
Polyvinylpyrrolidone	5.0
Methylparaben	1.57
Propylparaben	0.157

Distilled water, q.s. ad 1000 milliliters.

is prepared and sterilized by a conventional sterile filtration technique. The sterile vehicle is then mixed under aseptic conditions with ten grams of prednisolone sesquihydrate which has been previously comminuted to a particle size of about twenty microns and sterilized by exposure to ethylene oxide gas. The mixture is passed through a sterile colloid mill and filled under aseptic conditions into sterile vials. The vials are then plugged and sealed. The resulting suspension is sterile and remains stable under handling and storage conditions for at least twelve months at room temperature.

EXAMPLE 4.—AQUEOUS SUSPENSION FOR NASAL USE

A vehicle containing the following ingredients:

	Grams
Polyethylene glycol 4000	120
Sodium citrate	4.5
Myristyl gamma picolinium chloride	0.2
Polyvinylpyrrolidone	1.0
Neomycin sulfate	6.0
Phenylephrine hydrochloride	2.75

Distilled water, q.s. ad 1000 milliliters.

is prepared and filter sterilized. The sterile vehicle is then mixed with from about 2.5 grams to 5 grams of pred-

nisolone sesquihydrate which has been previously air-micronized to a particle size less than about ten microns and sterilized by exposure to ethylene oxide gas. The mixture is passed through a sterile colloid mill and filled under aseptic conditions into bottles adapted for nasal spray use. The resulting suspension is sterile and remains stable under handling and storage conditions for at least eighteen months at room temperature.

EXAMPLE 5.—AQUEOUS SUSPENSION FOR PARENTERAL USE AND FOR THE EYE AND EAR

Following the procedure of Example 4, but omitting the phenylephrine hydrochloride, one obtains a stable suspension suitable for use in the eye, ear and for injection.

EXAMPLE 6.—AQUEOUS SUSPENSION FOR PARENTERAL USE

A vehicle containing the following ingredients:

	Grams
Myristyl gamma picolinium chloride	0.2
Polyethylene glycol 4000	10.0
Sodium chloride	0.45
Sodium citrate	1.5
Distilled water, q.s. ad 1000 milliliters.	

is prepared and filter sterilized. The sterile vehicle is then mixed with from about 10 grams to about 100 grams of prednisolone sesquihydrate which has been previously comminuted to a particle size of about twenty microns and sterilized with ethylene oxide gas. The mixture is passed through a sterile colloid mill and filled under aseptic conditions into sterile vials. The vials are then plugged and sealed. The resulting suspension is sterile and remains stable under handling and storage conditions for at least twelve months at room temperature.

EXAMPLE 7.—AQUEOUS SUSPENSION FOR PARENTERAL USE

Three liters of vehicle identical in composition to the vehicle of Example 3 is prepared and filter sterilized. Thirty grams of micronized, sterilized prednisolone sesquihydrate is added under aseptic conditions. Thereafter 387 grams of sterilized procaine penicillin G of a particle size less than about ten microns preferably coated with 1.25% lecithin, and 1200 grams of sterilized procaine penicillin G of a particle size between about forty microns and about sixty microns preferably coated with 4% lecithin (process disclosed in U.S. Patent 2,694,665) are added under aseptic conditions. The mixture is passed through a sterile colloid mill and filled under aseptic conditions into sterile vials. The vials are then plugged and sealed. The resulting suspension is sterile and remains stable under handling and storage conditions for at least twelve months at refrigerator temperature.

EXAMPLE 8.—AQUEOUS SUSPENSION FOR PARENTERAL USE

Following the procedure of Example 7, a stable suspension is prepared by replacing the procaine penicillin G by calcium novobiocin in an amount sufficient to give a concentration of 200 milligrams per milliliter of suspension.

EXAMPLE 9.—AQUEOUS SUSPENSION FOR PARENTERAL OR TOPICAL USE

Following the procedure of Example 7, a stable suspension is prepared by replacing the procaine penicillin G by tetracycline base in an amount sufficient to give a concentration of 200 milligrams per milliliter of suspension.

EXAMPLE 10.—AQUEOUS SUSPENSION FOR PARENTERAL USE

Three liters of vehicle identical in composition to the vehicle of Example 3 is prepared and filter sterilized.

Thereafter 984 grams of sterile dihydrosterptomycin sulfate is dissolved in about 1500 milliliters of this vehicle and 30 grams of micronized, sterilized prednisolone sesquihydrate is added under aseptic conditions. This mixture is then combined under sterile conditions with 350 grams of sterilized procaine penicillin G of a particle size less than about 10 microns, preferably coated with 1.25% lecithin, and 350 grams of sterilized procaine penicillin G of a particle size between about 40 microns and 60 microns, preferably coated with 4% lecithin (U.S. Patent 2,694,665). The mixture is adjusted to a final volume of three liters using additional sterile vehicle as required, then processed through a sterile colloid mill and filled under aseptic conditions into vials. The vials are then plugged and sealed. The resulting suspension is sterile and remains stable under handling and storage conditions for at least twelve months at refrigerator temperature. This suspension is admirably suited for veterinary use.

This application is a continuation-in-part of our application Serial No. 675,255, filed July 31, 1957, now abandoned.

What is claimed is:

1. A stable aqueous suspension for topical and parenteral administration comprising prednisolone sesquihydrate having the formula $[(\Delta^{1,4}\text{-pregnadiene-3,20-dione-11}\beta,17\alpha,21\text{-triol})_2\cdot(H_2O)_3]$, having a particle size no more than about 20 microns and a pharmaceutically acceptable vehicle.

2. A stable veterinary aqueous suspension for topical and parenteral administration comprising prednisolone sesquihydrate having the formula $[(\Delta^{1,4}\text{-pregnadiene-3,20-dione-11}\beta,17\alpha,21\text{-triol})_2\cdot(H_2O)_3]$, having a particle size no more than about 20 microns and an antibiotic.

3. A stable veterinary aqueous suspension for topical and parenteral administration comprising prednisolone sesquihydrate having the formula $[(\Delta^{1,4}\text{-pregnadiene-3,20-dione-11}\beta,17\alpha,21\text{-triol})_2\cdot(H_2O)_3]$, having a particle size no more than about 20 microns and at least one member of the group consisting of procaine penicillin, dihydrostreptomycin, streptomycin, neomycin, polymyxin, tetracycline and calcium novobiocin.

4. A stable aqueous suspension for parenteral administration comprising prednisolone sesquihydrate having the formula $[(\Delta^{1,4}\text{ - pregnadiene - 3,20 - dione - 11}\beta,17\alpha,21\text{-triol})_2\cdot(H_2O)_3]$, having a particle size no more than about 20 microns, a preservative, a suspending agent and a surfactant.

5. A stable veterinary aqueous suspension for parenteral administration comprising prednisolone sesquihydrate having the formula $[(\Delta^{1,4}\text{-pregnadiene-3,20-dione-11}\beta,17\alpha,21\text{-triol})_2\cdot(H_2O)_3]$, having a particle size no more than about 20 microns, procaine penicillin G and dihydrostreptomycin.

6. A stable veterinary aqueous suspension for parenteral administration comprising prednisolone sesquihydrate having the formula $[(\Delta^{1,4}\text{-pregnadiene-3,20-dione-11}\beta,17\alpha,21\text{-triol})_2\cdot(H_2O)_3]$, having a particle size no more than about 20 microns, procaine penicillin G, dihydrostreptomycin, polyvinylpyrrolidone, a preservative and a buffer.

7. A stable veterinary aqueous suspension for parenteral administration comprising prednisolone sesquihydrate having the formula $[(\Delta^{1,4}\text{-pregnadiene-3,20-dione-11}\beta,17\alpha,21\text{-triol})_2\cdot(H_2O)_3]$, having a particle size no more than about 20 microns, neomycin sulfate, a preservative, polyethylene glycol of molecular weight about 4000 and a surfactant.

8. A composition of matter consisting of prednisolone sesquihydrate having the formula $[(\Delta^{1,4}\text{-pregnadiene-3,20-dione-11}\beta,17\alpha,21\text{-triol})_2\cdot(H_2O)_3]$, having a particle size of no more than about 20 microns and being free of deleterious crystal growth in aqueous suspension.

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UNITED STATES PATENT OFFICE
CERTIFICATE OF CORRECTION

Patent No. 3,062,712 November 6, 1962

Jack K. Dale et al.

It is hereby certified that error appears in the above numbered patent requiring correction and that the said Letters Patent should read as corrected below.

Column 1, line 39, for "series" read -- serious --; line 64, after "+95" insert -- to --; column 2, line 23, for "is" read -- it --; column 9, line 6, for "June 9, 1957" read -- June 9, 1956 --.

Signed and sealed this 23rd day of April 1963.

(SEAL)
Attest:

ERNEST W. SWIDER
Attesting Officer

DAVID L. LADD
Commissioner of Patents