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(54) Title: STABILIZED 1α-HYDROXY VITAMIN D

(57) Abstract: The invention provides a stabilized 1α-hydroxy vitamin D ("SHVD") which is particularly well suited for pharmaceutical formulations.

STABILIZED 1α-HYDROXY VITAMIN D

This invention relates a stabilized 1α-hydroxy vitamin D ("SHVD") which is particularly well suited for pharmaceutical formulations.

Vitamin D compounds having a hydroxy group at the 1α-position have had considerable attention in recent years because of their strong vitamin D activity. These activated vitamin D compounds are, however, also known as being chemically unstable, particularly under exposure of light and in the presence of oxygen, and as having poor storage stability at higher temperatures. The compounding of activated vitamin D into a pharmaceutical formulation exacerbates these stability problems.

Despite recognition and study of various aspects of the problem as well as prior art attempts to stabilize pharmaceutical compositions of activated vitamin D, the prior art has produced very little in the way of a 1α-hydroxy vitamin D form that has a specific chemical and physical profile which provides for a stabilized compound useful for pharmaceutical formulations. It has now been found that a stabilized 1α-hydroxy vitamin D with heretofore unknown technical properties is surprisingly stable compared to known forms of activated vitamin D.

FIG. 1 is an exemplary reaction scheme for the synthesis of 1α-OH-D_2.

FIGS 2A and 2B are HPLC chromatograms showing the preparation of SHVD in accordance with the present invention.

The present invention relates to a stabilized 1α-hydroxy vitamin D form with superior technical properties and superior stability (SHVD). SHVD is particularly well adapted for use in pharmaceutical compositions or formulations. Accordingly, the present invention will now be described in detail with respect to such endeavors; however, those skilled in the art will appreciate that such a description of the invention is meant to be exemplary only and should not be viewed as limitative on the full scope thereof.

In the following description of the method of the invention, process steps are carried out at room temperature and atmospheric pressure unless otherwise specified.

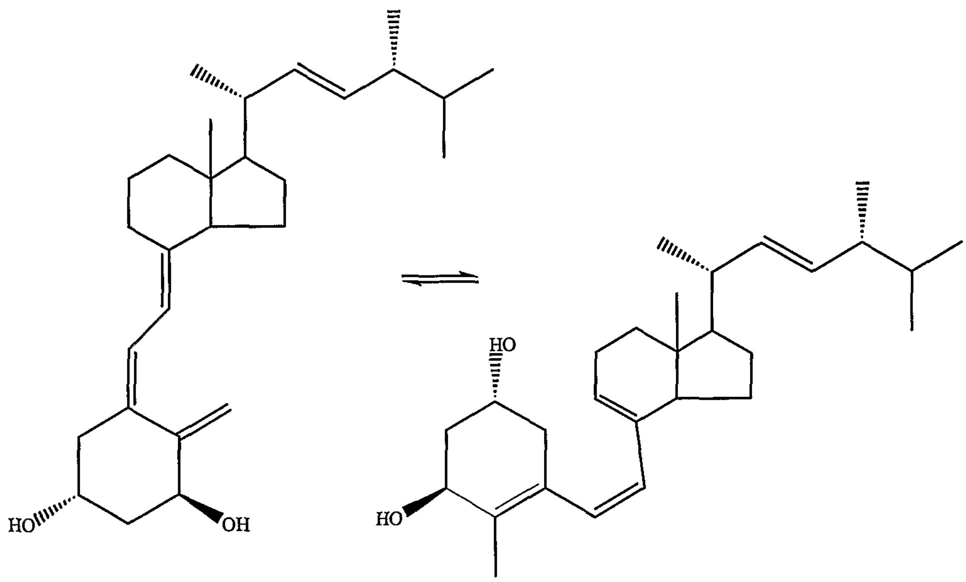
As used herein, the term "vitamin D" is intended to include vitamins D_2, D_3 and D_4 and related analogs. The term "1α-hydroxy vitamin D" refers to a vitamin D compound or analog that has a substituted hydroxy group in at least the carbon-1α position of the A ring of the compound. Those compounds that are substituted only with a hydroxy in the 1α-position, e.g., 1α-hydroxyvitamin D_2 and 1α-hydroxyvitamin D_3, are pro-drugs as they undergo further hydroxylation to form metabolically active vitamin D compounds. Those compounds that are hydroxylated in the 24 or 25 position as well as the 1α position are typically metabolically active vitamin D compounds, i.e., 1α, 24-dihydroxyvitamin D_2 and 1α, 25-dihydroxyvitamin D_3. The term "active vitamin D" or "activated vitamin D" referees to a vitamin D compound that is hydroxylated in at least the 1α, 24 or 25 position. As used herein, the term "stabilized 1α-hydroxy vitamin D" or "SHVD" is not meant to include certain commercially available vitamin D compounds, i.e., excludes 1α-hydroxyvitamin D_3, 1α,25-dihydroxyvitamin D_3, 1,24-dihydroxyvitamin D_3, 22-oxa-1,25-dihydroxyvitamin D_3 (OCT), paricalcitol (a 19-nor-vitamin D_2) and calcipotriol. The term "non-SHVD" refers to 1α-hydroxyvitamin D compounds that do not meet the purity profile and other characteristics described herein for SHVD.

SHVD of the present invention is crystalline, substantially solvent free, storage stable and well-suited for modern therapy formulations. Studies have demonstrated that SHVD with the technical properties found in the present invention is surprisingly stable, particularly its reduced rate of conversion to the corresponding previtamin form on heat challenge or elevated temperature. SHVD in accordance with the present invention shows virtually no degradation after 6 years storage at -70°C. This corresponds to excellent storage stability when formulated in a pharmaceutical dosage form, e.g., a soft gelatin capsule. The SHVD of the present invention does not require low oxygen or inert atmosphere for storage as has been advanced by some prior art stabilized formulations.

SHVD of the present invention is also characterized in that it has an ultraviolet spectrum peak maximum (λ_{max}) at about 265 nm with a molar extinction coefficient, Σ_{265nm} of 17490 and a minimum (λ_{min}) at about 227 nm, and has a specific purity profile. SHVD has a purity equal to or greater than 98% weight-base (i.e., at least 98%) by high performance liquid chromatography (HPLC), has residual solvents of 0.5% or less, has total impurities of 1.5% or less, and has no single impurity greater than 0.5% by HPLC. The impurities include *cis*-1α-OH-D_4 and *cis*-1β-OH-D_2. While not wanting to be bound by any particular theory, it is believed that the remarkable stability of SHVD is related to its being substantially solvent free and substantially free of impurities.

The SHVD in accordance with the present invention is further characterized by a substantially reduced rate of conversion to the corresponding previtamin upon heat challenge when compared to the same non-SHVD. This stability of SHVD with respect to conversion to the corresponding previtamin D is especially surprising. A 1α-hydroxy vitamin D compound is in thermal equilibrium with its previtamin form, i.e., they are thermal isomers of each other.

By way of example, the equilibrium for 1α-OH-D_2 is illustrated below.



1a-OH-D_2 1a-OH-pre D_2

The 1α-hydroxyprevitamin D_2 has a characteristic ultraviolet spectrum peak maximum (λ_{max}) at 259 nm. Storage of the 1α-hydroxy vitamin D form in solution or at elevated temperatures produces the 1α-hydroxy previtamin D form. The isomers are in equilibrium with each other even at room temperature. Increased temperature shifts the equilibrium to the formation of the previtamin.

The SHVD of the present invention shows reduced rate of conversion to 1α-hydroxy previtamin D compared to corresponding non-SHVD compounds under the same conditions, e.g., a heat challenge. The heat challenge is suitably an elevated temperature held in air for a sustained period, which period may be up to 10 days. The elevated temperature is suitably about 60° C. Upon such heat challenge the SHVD of the present invention has a lower rate of conversion compared to that for a non-SHVD compound under the same conditions.

1α-hydroxy vitamin D compounds may be prepared by any of the known methods of synthesis. It has been found that certain synthetic pathways provide a compound that, upon purification, may have the superior technical properties of the SHVD of the present invention. An exemplary synthetic scheme is shown in Fig. 1 illustrating the synthesis of 1α-hydroxyvitamin D_2 (also known as doxercalciferol). Generally, the synthesis includes conversion of a starting material vitamin D (i.e., the compound that is to be hydroxylated in the 1α-position) to the cyclovitamin form, hydroxylation of the cyclovitamin in the 1α-position, reconversion of the hydroxylated cyclovitamin to the *cis* and *trans* forms of the vitamin, and conversion of the *trans* form to the *cis* form. The hydroxy group in the 3-position is protected, suitably by conversion to a tosylate as a first step in the synthesis.

By way of example, as shown is Fig. 1, vitamin D_2 (or ergocalciferol) is the starting material. Vitamin D_2 is first tosylated in the 3-position to form vitamin D_2 3-tosylate. The tosylate is then converted to the cyclovitamin that is then 1α-hydroxylated. The hydroxylated cyclovitamin is then converted to *cis* and *trans* 1α-hydroxyvitamin D_2. The *trans* isomer is irradiated with ultraviolet light to convert to the *cis* isomer, i.e., 1α-hydroxyvitamin D_2.

The resulting product is purified by column chromatography. The column-purified 1α-hydroxyvitamin D_2 is then recrystallized from an organic solvent, e.g., methyl formate. The recrystallized crystals are then vacuum oven dried to a residual solvent content of <0.5% to provide SHVD. The SHVD in accordance with the present invention is particularly satisfactory when successive (generally at least two) recrystallizations are performed. Suitable solvents, in addition to methyl formate, include ethyl formate, ethyl acetate, acetone, methylethylketone, hexane, 2-propanol-hexane, pentane, heptane, diethyl ether, diisopropyl ether, methanol, ethanol acetonitrile, and combinations thereof.

The SHVD of the present invention is suitably used in pharmaceutical formulations, such as for oral use, e.g., soft gelatin capsules, solutions, tablets. The concentration of active ingredient in convenient unit dosage form is 0.5 μg to 25 μg, with a weekly dose of between 1 μg and 100 μg/week. In a soft gelatin formulation, the capsule fill suitably contains SHVD of the present invention which is dissolved in a pharmaceutically acceptable oil, e.g., fractionated coconut oil, and includes an antioxidant which may be, for example, butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT) or vitamin E. The capsule shell suitably contains gelatin, glycerin, titanium dioxide and coloring agent. The fill is typically about 58-59% by weight of the whole capsule.

The present invention is further explained by the following examples which should not be construed by way of limiting the scope of the present invention.

In the examples, UV spectra were taken on a Shimadzu UV 160V, IR spectra on a Analect DS-20, and NMR on a SEOL NMR spectrometer operating at 400 MH_3 or like instruments known in the art.

Preparation of stabilized 1α-hydroxyvitamin D_2 (SHVD$_2$)

Example 1: **<u>Preparation of 1α-hydroxyvitamin D_2 crude drug substance</u>**

<u>(1) Preparation of ergocalciferol tosylate(2)</u>

To 100 g of ergocalciferol (vitamin D_2) was added 3.1 g of 4-dimethylaminopyridine, 100 mL anhydrous pyridine, and 340 mL anhydrous dichloromethane. The flask contents were stirred until the internal reaction temperature was ≤ 5°C. Then, 122.6 g of p-toluenesulfonyl chloride was added, and the mixture stirred until all the solids were dissolved. The reaction was allowed to proceed with slow stirring under argon. After 32 hours, the completion of the reaction was monitored by TLC (silica gel plates; ethyl acetate in hexanes; 20% phosphomolybdic acid in ethanol).

Over a period of approximately 20 minutes, the reaction mixture was dripped into a chilled beaker containing saturated sodium bicarbonate. The reaction mixture was rinsed into a separatory funnel with 900 mL isopropyl ether. The mixture was extracted with isopropyl ether by shaking, and the organic and aqueous layers separated. The isopropyl ether extracts were washed with dilute hydrochloric acid, saturated sodium bicarbonate, and saturated sodium chloride. The combined isopropyl ether extracts were dried over 250 g of anhydrous magnesium sulfate and filtered. The isopropyl ether extracts were combined and concentrated *in vacuo*. The crude tosylate was suitable for use in the solvolysis below.

<u>(2) Preparation of 6(R)-methoxy-3,5-cyclovitamin D_2(3)</u>

To a stirred mixture of 1950 mL of dry methyl alcohol and 500 g of sodium bicarbonate was added the ergocalciferol tosylate from Step 1 dissolved and rinsed in 250 mL dichloromethane. The stirred mixture was refluxed under an argon atmosphere for 18-

22 hours with an internal temperature of about 56 °C. After about 20 hours, the extent of reaction was monitored by TLC (silica gel; solvents 20% phosphomolybdic acid in ethanol).

The reaction mixture was cooled to 25-30°C, and the sodium bicarbonate was removed by filtration. The reaction flask, filtered sodium bicarbonate and filtration flask were rinsed with 1000 mL of isopropyl ether. The organic filtrate was concentrated *in vacuo*, maintaining the temperature of the bath at 50 °C. The semi-solid residue was diluted with isopropyl ether. This isopropyl ether solution was washed three times with water and once with saturated sodium chloride, and then dried over magnesium sulfate. The magnesium sulfate was removed by filtration and rinsed with isopropyl ether. The combined organic filtrate and the isopropyl ether rinse of the filtration flask were concentrated *in vacuo* for 4-18 hours to yield the 6(R)-methoxycyclovitamin.

(3) Preparation of 1α-hydroxy-6(R)-methoxy-3,5-cyclovitamin D_2(4)

To a stirred suspension of 12.3 g of freshly ground selenium dioxide in 1500 mL of 1,2-dichioroethane was added 81 mL of anhydrous *t*-butyl hydroperoxide (5-6 M in decane). After stirring under positive argon atmosphere for 3-4 hours at ambient temperature, 18 mL of pyridine was added. The 6(R)-methoxy-3,5-cyclovitamin D_2 from Step 2, dissolved in 1,2-dichloroethane, was added dropwise into the chilled reaction flask followed by a rinse of 1,2-dichloroethane. The reaction continued with stirring at an internal temperature of <5°C, and the reaction progress was monitored at 15-minute intervals by TLC. The reaction was considered complete when the starting 6(R)-methoxy-3,5-cyclovitamin D_2 was either no longer visible or did not appear to change between two consecutive monitoring thin layer chromatograms.

The reaction was quenched with 10% sodium hydroxide. The aqueous and organic layers were separated, and the aqueous layer was extracted with isopropyl ether. This isopropyl ether extract was added to the organic layer from the reaction mixture. The combined extracts were dried with anhydrous magnesium sulfate and filtered. The flask and residual magnesium sulfate were rinsed with isopropyl ether, and the combined isopropyl ether extracts and isopropyl ether rinse of the filtration flask were concentrated *in vacuo*.

The product was dissolved in 500 mL of 5% ethyl acetate in hexanes, purified on a silica gel (60/230-400 Mesh) in a column (75 mm I.D.), and eluted with approximately

4000 mL 5% ethyl acetate in hexanes, followed by 8000 mL 20% ethyl acetate in hexanes under positive argon pressure. Fractions of approximately 200 mL each were collected during the elution. The fractions were analyzed by silica gel TLC. The fractions containing the 1α-hydroxy-6(R)-methoxy-3,5-cyclovitamin D_2 were identified, combined, and concentrated *in vacuo.*

(4) Preparation of *cis-* and *trans-* 1α-hydroxyvitamin D_2(5 and 6)

A stirred solution of the lα-hydroxy-6(R)-methoxy-3,5-cyclovitamin D_2 from Step 3 in 120 mL of dimethyl sulfoxide and 130 mL of glacial acetic acid was heated to about 60° C and stirred at this temperature under an argon atmosphere for 60 minutes. The reaction was monitored periodically by silica gel TLC for the consumption of the reactant lα-hydroxy-6(R)-methoxy-3,5-cyclovitamin D_2. Upon completion, the reaction was cooled to 20 - 25°C. The reaction was quenched by pouring the mixture and an isopropyl ether rinse into saturated sodium bicarbonate and water held at a temperature ≤10°C. Isopropyl ether was added and the mixture was stirred. This mixture was then rinsed into a separatory funnel with 950 mL isopropyl ether and extracted by shaking. The aqueous and organic layers were separated. The aqueous layer was extracted with isopropyl ether. The organic layer from the reaction mixture was combined with this isopropyl ether extract and washed with saturated sodium bicarbonate three times, water three times, and saturated sodium chloride once. The organic extracts were dried over magnesium sulfate, filtered, and the cake magnesium sulfate and filtration flask were rinsed with isopropyl ether. The combined isopropyl ether filtrates were concentrated *in vacuo,* maintaining the temperature of the bath at about 35 °C. Residual solvent was removed from the remaining residue by vacuum.

The product, dissolved in 350 mL dichloromethane, was purified on silica gel (60/230-400 mesh) in a column (75 mm I.D.) eluted with 40% ethyl acetate in hexanes under argon. Fractions of approximately 125 mL were collected. The fractions were analyzed by silica gel TLC eluted with 40% ethyl acetate in hexanes. The fractions containing *cis-* and *trans*-lα-OH-D_2 were identified, combined, and concentrated *in vacuo.*

(5) Preparation of *cis*-1α-hydroxyvitamin D_2(5)

The *cis-* and *trans*-lα-OH-D_2 mixture from Step 4 and 1.1 g of 9-acetylanthracene were dissolved in 2000 mL methyl alcohol and transferred with methyl alcohol rinses totaling 2000 mL to a photolysis chamber and diluted with additional methyl alcohol to a total of 13.7 L. The photolysis was run to completion, approximately 2-6 hours. The product solution and 400 mL methyl alcohol rinse were concentrated *in vacuo*, maintaining the temperature of the bath around 35 °C.

The residual solvent was removed under high vacuum. The product, dissolved in dichloromethane, was purified on silica gel (60/230-400 mesh) in a column (75 mm I.D.) eluted with 40% ethyl acetate in hexanes and ethyl acetate under argon pressure. Fractions (125 mL) were collected with 40% ethyl acetate in hexanes elution, then fractions (200 mL) were collected with 100% ethyl acetate elution. The fractions were analyzed by silica gel TLC eluted with 40% ethyl acetate in hexanes. The fractions containing the *cis*-lα-OH-D_2 were identified, combined, and concentrated *in vacuo*, The residual solvent was removed under high vacuum. A typical yield for this step was 15-30 g (15-30% overall yield from the starting material, ergocalciferol).

Example 2: Purification of crude drug substance to yield $SHVD_2$

The column-purified *cis*-lα-hydroxyvitamin D_2 (crude drug substance) was successively recrystallized from methyl formate.

For each recrystallization, the column-purified crude drug substance (usually 15-30 g) was resuspended in 3500 mL methyl formate in a round bottom flask. This flask was attached to a rotary evaporator with a chilled condenser and slowly rotated in a water bath at 40°C until the solids dissolve. The solution was concentrated in the rotary evaporator, maintained at 40°C until crystals began to form. When crystals were observed, the flask was placed in a –20°C freezer to cool for 48-72 hours.

After the crystals obtained from the recrystallization had air dried for 5 - 10 minutes, the crystals were milled to a powder, and then, transferred to a crystallization dish. The dish with crystals was placed in a vacuum oven set at about 55 °C for 72 -120 hours. The amount of drug substance obtained ($SHVD_2$) was usually 5 - 10 g, which represents a yield of 5 - 10 % of the starting material, ergocalciferol.

Evidence that the purification procedure improved the purity of the crude drug substance was demonstrated by the HPLC chromatogram in FIG. 2. The first chromatogram (A) was obtained with column-purified crude drug substance. Chromatogram (B) shows the final product. As shown in FIG. 2, the levels of the impurities with retention times of approximately 37.7 and 41 minutes were reduced by recrystallization.

Example 3: Determination of residual solvents of $SHVD_2$

A method for quantitating residual solvents with toluene was used. The target analytes included methanol, methyl formate, methylene chloride, hexane, diisopropyl either, 1,1-dichloroethane and ethyl acetate.

A gas chromatographic method was used with helium as the carrier gas, split injections/run, and run in single ion mode (SIM mode). It required a 60 m x 0.25 mm ID, DB-624 capillary column with a film thickness of 1.4 µm, a split/splitless injector, a gas chromatograph capable of ramping from 50 °C to 250 °C, and a mass selective detector. The chromatograph was set to detect ions 31, 32, 43, 45, 49, 60, 61, 62, 84, 86 and 87 at a dwell time of 25 ms per ion.

An external calibration was used, consisting of a blank, matrix blank and a 4-point curve. A curve was prepared at concentration of 1 µg/g, 10 µg/g and 50 µg/g. A 10 mg sample of *cis*-1α-OH-D_2 was prepared in toluene. Results from the samples were calculated using the linear curve generated above. The lower limit for all analytes was 100 µg/g. The results indicated that the amount of residual solvents in the *cis*-1α-OH-D_2 ($SHVD_2$) was ≤0.5%

Example 4: Storage stability testing of $SHVD_2$

The stability of samples from four lots of $SHVD_2$ was examined at 6-month intervals during long-term storage at –70°C. All lots were stored in the container-closure system selected as packaging for the drug substance. Stability was assessed by examining samples for changes in (1) weight percent lα-OH-D_2 content, (2) percent related substances content, and (3) impurity profile, using HPLC. In addition, all lots were examined for changes in UV spectrum by UV spectrophotometry.

Results of this study showed no significant differences in mean weight % lα-OH-D_2 content between baseline and 6 or 12 months. The weight % lα-OH-D_2 content for all lots examined was >98.0% at all time points. Impurity profile chromatography demonstrated no new peaks at 6 or 12 months relative to baseline. The percent related substances remained <2.0%.

Example 5: Preparation of $SHVD_2$ soft gelatin capsule fill

BHA was added to fractionated coconut oil (FCO), heated and stirred until the BHA was completely dissolved. This solution was then cooled. $SHVD_2$ was accurately weighed into a suitable container, dehydrated alcohol was added, and the solution was stirred until the $SHVD_2$ was dissolved. FCO was weighed into a stainless steel mixing tank. The BHA/FCO solution was added to the FCO while stirring. Then $SHVD_2$/dehydrated alcohol solution was added and the solution was stirred until uniform. The vessel containing the $SHVD_2$/dehydrated alcohol solution was rinsed with dehydrated alcohol and the rinses were added to the FCO/$SHVD_2$/dehydrated alcohol mix. The mixing tank containing this mix was sealed, a vacuum was applied, and the contents were mixed (deaerated). The resulting solution was transferred through filters to receivers and blanketed with nitrogen.

Example 6: Stability testing: $SHVD_2$ v. non-$SHVD_2$

Approximately 1 mg of $SHVD_2$ or non-stabilized 1α-hydroxyvitamin D_2 (non-$SHVD_2$) is placed in each of 9 (12 X 75mm) test tubes. The test tubes are placed in an oven at about 60°C. One test tube of each of the two vitamin D_2 preparations is removed at 0, 3, 6, 9, 12, 18, 24, 36 and 48 hours of incubation. The contents in each test tube are dissolved in ethanol to produce a 1 mg/mL solution. Each solution (10μL) is analyzed by the following HPCL conditions: a YMC Pack C8 column (4.6 x 250 mm, 5μm) with a mobile phase of acetonitrile: methanol: water (60:20:20) at a flow rate of 1.0 mL/min. The rate of appearance of the peak migrating with a relative retention time of approximately 0.9 compared with the 1α-hydroxyvitamin D_2 peak is greater with the non-$SHVD_2$ than with $SHVD_2$.

Example 7: Stability testing: $SHVD_2$ v. commercially available 1α-OH-D_2

Approximately 1 mg of $SHVD_2$ or commercial 1α-hydroxyvitamin D_2 is placed in each of 2 (12 x 75 mm) test tubes. One test tube from each of the two preparations is placed in an oven at about 60°C. For analysis, the contents in each test tube are dissolved in ethanol to produce a 1 mg/mL solution. Each solution (10μL) is analyzed by the following HPLC conditions: a YMC Pack C8 column (4.6 x 250 mm, 5μm) with a mobile phase of acetonitrile: methanol: water (60:20:20) at a flow rate of 1.0 mL/min. The rate of appearance of the peak migrating with a relative retention time of approximately 0.9 compared with the 1α-hydroxyvitamin D_2 peak is greater with the commercial 1α-hydroxyvitamin D_2 than with $SHVD_2$.

Example 8: Stability testing of $SHVD_2$ v. non-$SHVD_2$

Approximately 1 mg of stabilized 1α-hydroxyvitamin D_2 (doxercalciferol) ($SHVD_2$) was placed in each of two test tubes (12 x 75 mm). One sample was placed in a dessicator containing an atmosphere of ethanol vapors to increase the residual solvent content of the material to form non-stabilized doxercalciferol (non-$SHVD_2$). The material in the dessicator was heated at 60°C for 48 hours. The other sample was not subjected to the ethanol atmosphere, thereby retaining its stabilized form, but was heated at 60° C for 48 hours. The contents in each test tube were dissolved in ethanol to produce a 1 mg/mL solution. Each solution (10 μL) was analyzed by the following HPLC conditions: a YMC Pack C8 column (4.6 x 250 mm, 5 μm) with a mobile phase of acetonitrile: methanol: water (60:20:20) at a flow rate of 1.0 mL/min. The rate of appearance of the peak migrating with a relative retention time of approximately 0.9 compared with the doxercalciferol peak is greater with the non-$SHVD_2$ material than with $SHVD_2$.

Example 9: Stability testing: 1α,24(S)-dihydroxyvitamin D_2 containing > 0.5% residual solvents (a non-$SHVD_2$)

Approximately 1 mg of 1α,24(S)-dihydroxyvitamin D_2 containing >0.5% residual solvents (non-SHVD) was placed in a capped vial and heated at 60°C for 48 hours. The contents of the vial were dissolved in ethanol to produce a 1 mg/mL solution. The solution (10μL) was analyzed by the following HPLC conditions: a YMC Pack Pro C18 column (4.6 x 150 mm, 5 μm) with a mobile phase of acetonitrile: water (50:50) at a flow rate of 1.0 mL/min. After heating the non-SHVD, a compound with a relative retention time of approximately 0.85 compared with standard 1α,24(S)-dihydroxyvitamin D_2 was observed.

In summary, the present invention provides SHVD, a stabilized 1α-hydroxyvitamin D form with superior technical properties and superior stability. The novel form of the present invention is crystalline, substantially solvent free, stable and well suited for modern therapy formulations.

While the present invention has now been described and exemplified with some specificity, those skilled in the art will appreciate the various modifications, including variations, additions, and omissions, which may be made in what has been described. Accordingly, it is intended that these modifications also be encompassed by the present invention and that the scope of the present invention be limited solely by the broadest interpretation that lawfully can be accorded the appended claims.

CLAIM(S)

We claim:

1. A stabilized 1α-hydroxy vitamin D (SHVD) characterized by a purity equal to or greater than 98% by a weight-based HPLC assay, residual solvents of 0.5% or less, a total impurity of 1.5% or less, and no single impurity of greater than 0.5%.

2. The stabilized 1α-hydroxy vitamin D of claim 1 wherein the impurity is 1α-hydroxyvitamin D_4.

3. The stabilized 1α-hydroxy vitamin D of claim 1 wherein the rate of conversion of the vitamin D form to the previtamin form upon heat challenge is less than that of non-stabilized 1α-hydroxy vitamin D (non-$SHVD_2$) under the same conditions.

4. A pharmaceutical composition comprising the SHVD of claim 1.

5. The composition of claim 4, which is a soft gelatin, capsule.

6. The composition of claim 4 which is a solution.

7. The composition of claim 4, in unit dosage form, having a content of active component of 0.5 μg-25 μg.

8. A composition which comprises a solution of an effective amount of the SHVD of claim 1 in an oil, the solution contained in a soft gelatin capsule.

9. The composition of claim 8 wherein the oil is fractionated coconut oil.

10. A composition which comprises a solid pharmaceutical preparation of an effective amount of the SHVD of claim 1, wherein the solid pharmaceutical preparation is in the form of a tablet, a capsule, a granule or a powder.

11. A medicament for the treatment of a disease due to abnormality in calcium absorption, transportation or metabolism, hyperproliferative cellular activity, immune response imbalance or inflammatory response imbalance which comprises an effective amount of the SHVD of claim 1 as an active ingredient.

12. A method of treating a disease due to abnormality in calcium absorption, transportation or metabolism, hyperproliferative cellular activity, immune response imbalance or inflammatory response imbalance which comprises administering to a subject in need thereof, an effective amount of the SHVD of claim 1 as an active ingredient.

13. The method of claim 12 wherein the disease is psoriasis.

14. A stabilized 1α-hydroxy vitamin D (SHVD) characterized by reduced rate of conversion to a 1α-hydroxy previtamin D when heat challenged compared to that of non-SHVD under identical conditions.

15. The stabilized 1α-hydroxy vitamin D of claim 14 wherein the heat challenge occurs at a constant temperature of about 60°C for a sustained period.

16. A stabilized 1α-hydroxy vitamin D (SHVD) which is substantially pure, substantially solvent free and storage stable.

17. A pharmaceutical compound comprising a stabilized 1α-hydroxy vitamin D (SHVD) characterized by a substantially reduced rate of conversion to its corresponding previtamin form.

18. A method of making stabilized 1α-hydroxy vitamin D (SHVD) comprising: a) tosylating the hydroxy group in the 3-position of a starting material which is a vitamin D compound to which a hydroxy is to be added in the 1α-position; b) converting the tosylated form to a cyclovitamin; c) hydroxylating the cyclovitamin in the 1α-position; d) converting the cyclovitamin to the *cis* and *trans* vitamin forms; e) irradiating the *trans*

vitamin form to yield the *cis* form; f) recrystallizing the *cis* form in an organic solvent, and vacuum oven drying the recrystallized form for 72 – 120 hours and at 55 °C to yield the SHVD.

19. A stabilized 1α-hydroxy vitamin D (SHVD) synthesized by the method of claim 18.

20. A method of making stabilized 1α-hydroxy vitamin D (SHVD) comprising successively recrystallizing at least three times, a crude hydroxyvitamn D product from an organic solvent.

21. A method as claimed in claim 20, wherein the recrystallizing step includes two recrystallizations.

22. A method as claimed in claim 21, wherein the organic solvent is methyl formate, ethyl formate, ethyl acetate, acetone, methylethylketone, hexane, 2-propanol-hexane, pentane, heptane, diethyl ether, diisopropyl ether, methanol, ethanol acetonitrile, or combinations thereof.

FIG. 1

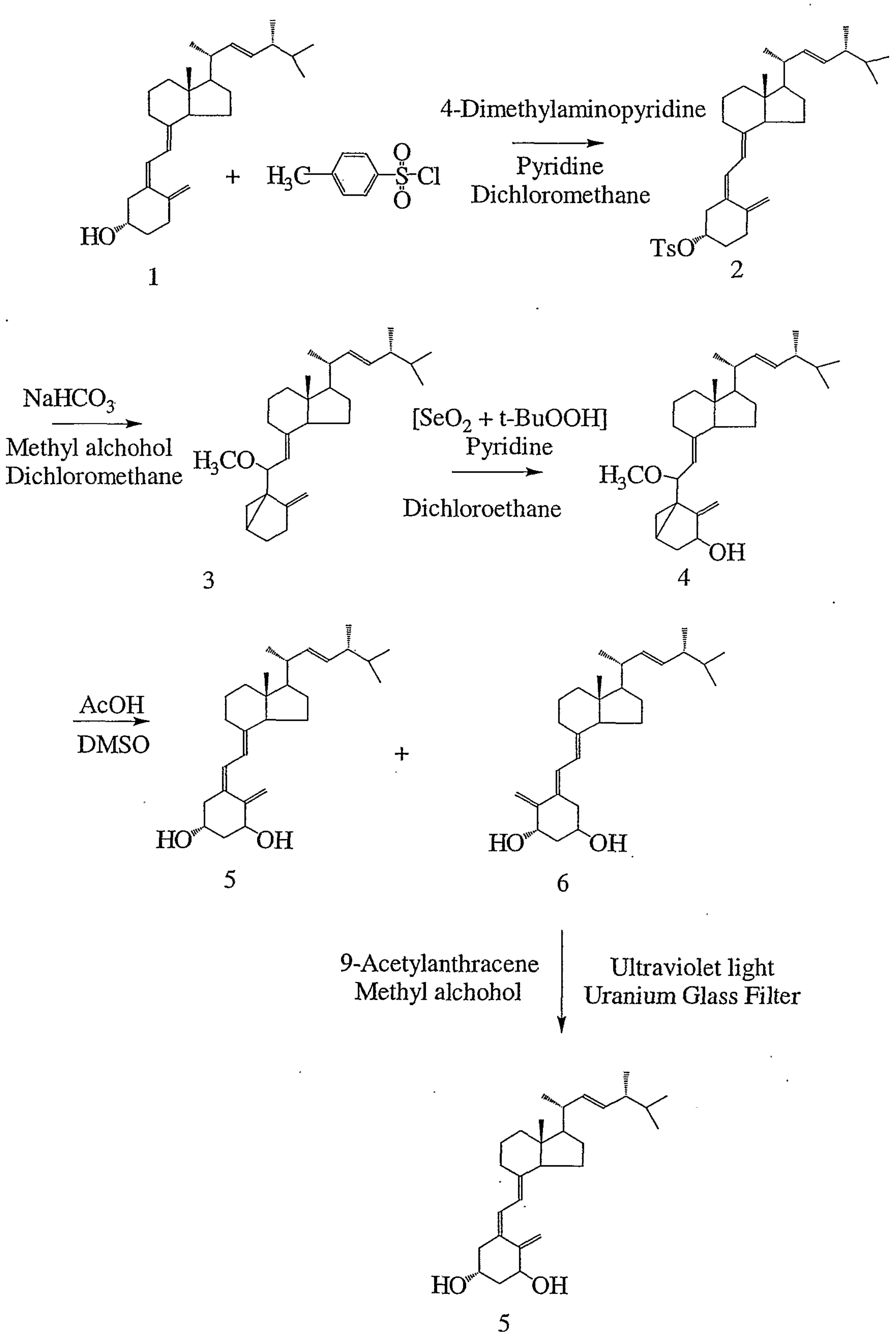

HO
H$_3$C
S
Cl
O
O
4-Dimethylaminopyridine
Pyridine
Dichloromethane
TsO
1
2
NaHCO$_3$
Methyl alchohol
Dichloromethane
H$_3$CO
[SeO$_2$ + t-BuOOH]
Pyridine
Dichloroethane
H$_3$CO
OH
3
4
AcOH
DMSO
HO
OH
HO
OH
5
6
9-Acetylanthracene
Methyl alchohol
Ultraviolet light
Uranium Glass Filter
HO
OH
5

FIG. 2A

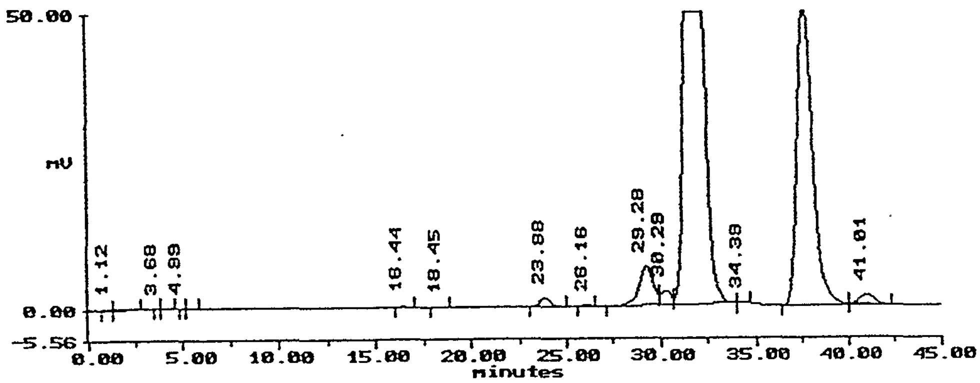
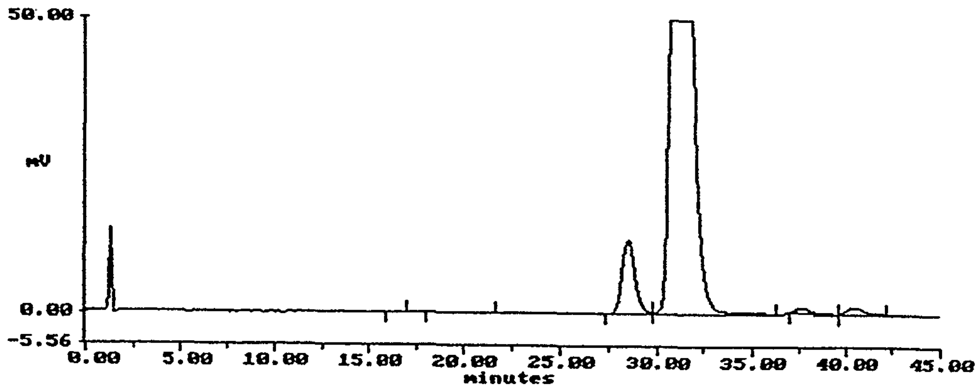

50.00
mV
0.00
-5.56
1.12
3.68
4.89
16.44
18.45
23.88
26.16
29.28
30.28
34.38
41.01
0.00
5.00
10.00
15.00
20.00
25.00
30.00
35.00
40.00
45.00
minutes

FIG. 2B


50.00
mV
0.00
-5.56
0.00
5.00
10.00
15.00
20.00
25.00
30.00
35.00
40.00
45.00
minutes