(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
18 June 2020 (18.06.2020)

WIPO | PCT

(10) International Publication Number
WO 2020/119836 A1

(51) International Patent Classification:
C07J 9/00 (2006.01) *C07J 51/00* (2006.01)
C07J 41/00 (2006.01)

(21) International Application Number:
PCT/CZ2019/000009

(22) International Filing Date:
12 February 2019 (12.02.2019)

(25) Filing Language: Czech

(26) Publication Language: English

(30) Priority Data:
PV 2018-69 12 December 2018 (12.12.2018) CZ

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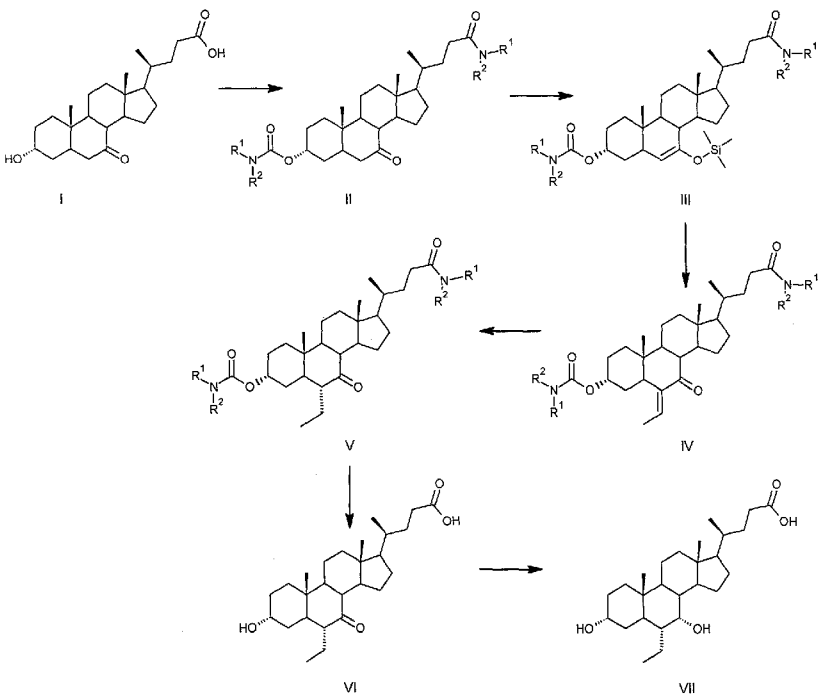
(81) Designated States *(unless otherwise indicated, for every kind of national protection available)*: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DJ, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IR, IS, JO, JP, KE, KG, KH, KN, KP, KR, KW, KZ, LA, LC, LK, LR, LS, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SA, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(84) Designated States *(unless otherwise indicated, for every kind of regional protection available)*: ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, ST, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU, TJ,

(54) Title: PROCESS FOR PREPARING OBETICHOLIC ACID



(57) Abstract: A process for preparing obeticholic acid, wherein the 7-keto-litocholic acid amide of formula (II), where R^1, R^2 are a C_1-C_6 n-alkyl, or together a C_5-C_6 cycle and the protecting group on the hydroxyl in position 3 is a carbamoyl with the same substitution as the amide, is converted to the 7-trimethylsilyl enol ether of formula (III), which is then converted by reaction with an alkylation agent to the 6-ethylidene-derivative of formula (IV), which is hydrogenated to the compound of formula (V) and then simultaneously deprotected in position 3 and hydrolysed to the compound of formula (VI) and then reduced to obeticholic acid of formula (VII).

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[Continued on next page]

TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, KM, ML, MR, NE, SN, TD, TG).

Declarations under Rule 4.17:

— *as to applicant's entitlement to apply for and be granted a patent (Rule 4.17(ii))*

Published:

— *with international search report (Art. 21(3))*

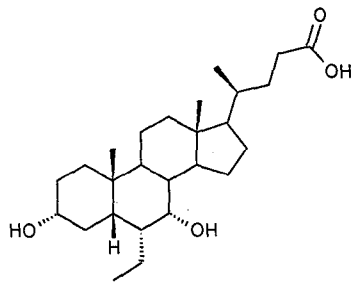
Process for preparing obeticholic acid

Technical Field

The invention relates to a process for preparing obeticholic acid with a reduced number of synthetic steps.

Background Art

Obeticholic acid, i.e. (3α,5β,6α,7α)-6-ethyl-3,7-dihydroxycholanic acid, of structural formula VII

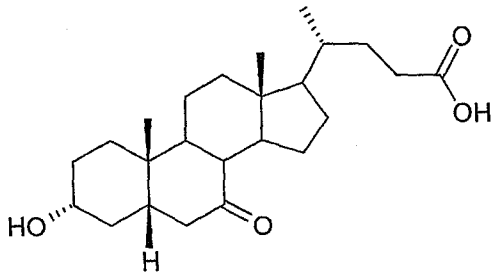


VII

was first described in Yu, D. et al., Steroids 2012, 77(13): 1335, Pellicciari, R. et al., in J Med Chem 2002, 45(17): 3569 and in the international patent application WO 2002/072598 as an agonist of the farnesoid X receptor (FXR).

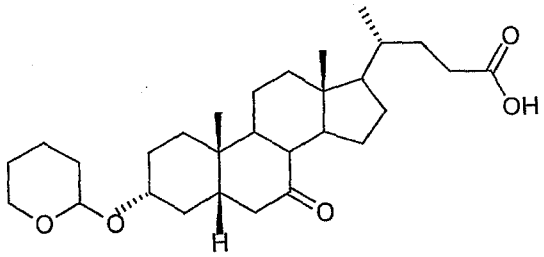
Synthesis of obeticholic acid has been described in literature as well as in numerous patent documents.

The synthesis described in these documents is based on 3-α-hydroxy-7-keto-5β-cholanic acid of formula I

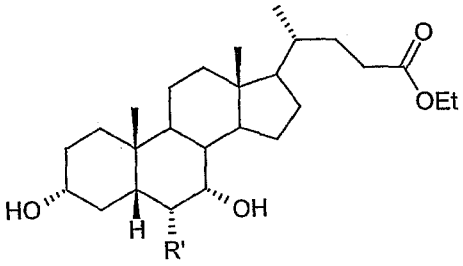


I

and proceeds via a protected derivative



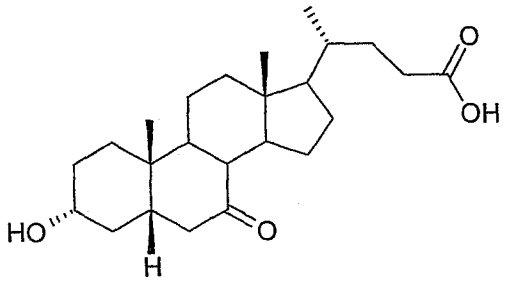
by alkylation, hydrogenation and reduction to the ethyl ester,



which is then hydrolyzed to obeticholic acid. However, this method provides a very low yield of the final compound.

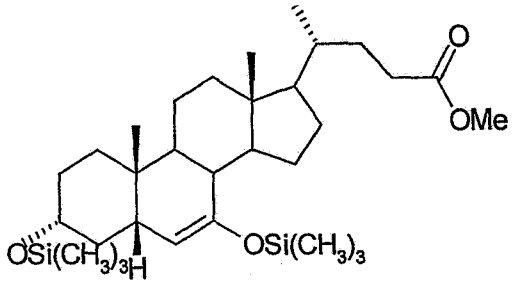
Synthesis of obeticholic acid has been described in other literature as well as in numerous patent documents.

The document WO 2006/122977 (Erregierre S.p.A.) discloses a synthetic method that starts with the same compound of formula I

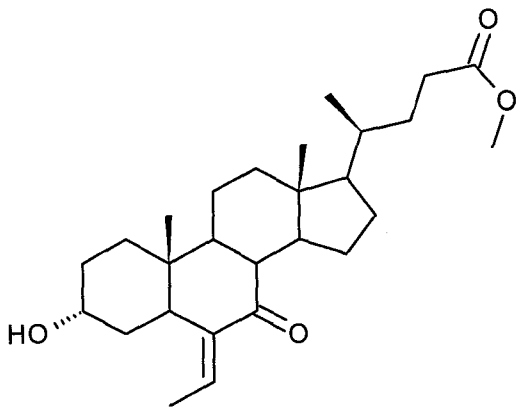


I

and proceeds via the methyl ester and its silyl derivative and silyl enol ether,



from which the methyl ester is obtained by reaction with an aldehyde, catalyzed by a Lewis acid,



which then provides obeticholic acid by hydrolysis, hydrogenation and reduction. The problem of alkylation of the methyl ester within this synthesis is that it partly provides the ethyl derivative in position α with respect to the carboxyl instead of the 6-ethyl derivative unless partial thermal desilylation in position α is conducted in the previous step. The same cannot be monitored well by chromatography or other analytic methods common in interoperational monitoring of technological processes. In addition, the yield is low again, which is also supported by easy hydrolysis of the 3-TMS protecting group during alkylation (quick hydrolysis by Lewis acids). Further, retroaldolization occurs during the alkylation – Lewis acids also catalyze the backward step of the alkylation balance.

Synthesis using the methyl ester as an intermediate is also described in the documents CN 105399793, CN 105646633, CN 105566429, WO 2017/027396, CN 106083971, CN 106279335 or WO 2013/192097.

Synthesis with the use of the benzyl ester as an intermediate is for example described in the documents CN 105481925, CN 104876995, CN 105585603 and CN 106083970.

It would be advantageous to have an efficient synthesis method making it possible to obtain obeticholic acid with high conversion, with a minimal or no share of retroaldolization and deprotection.

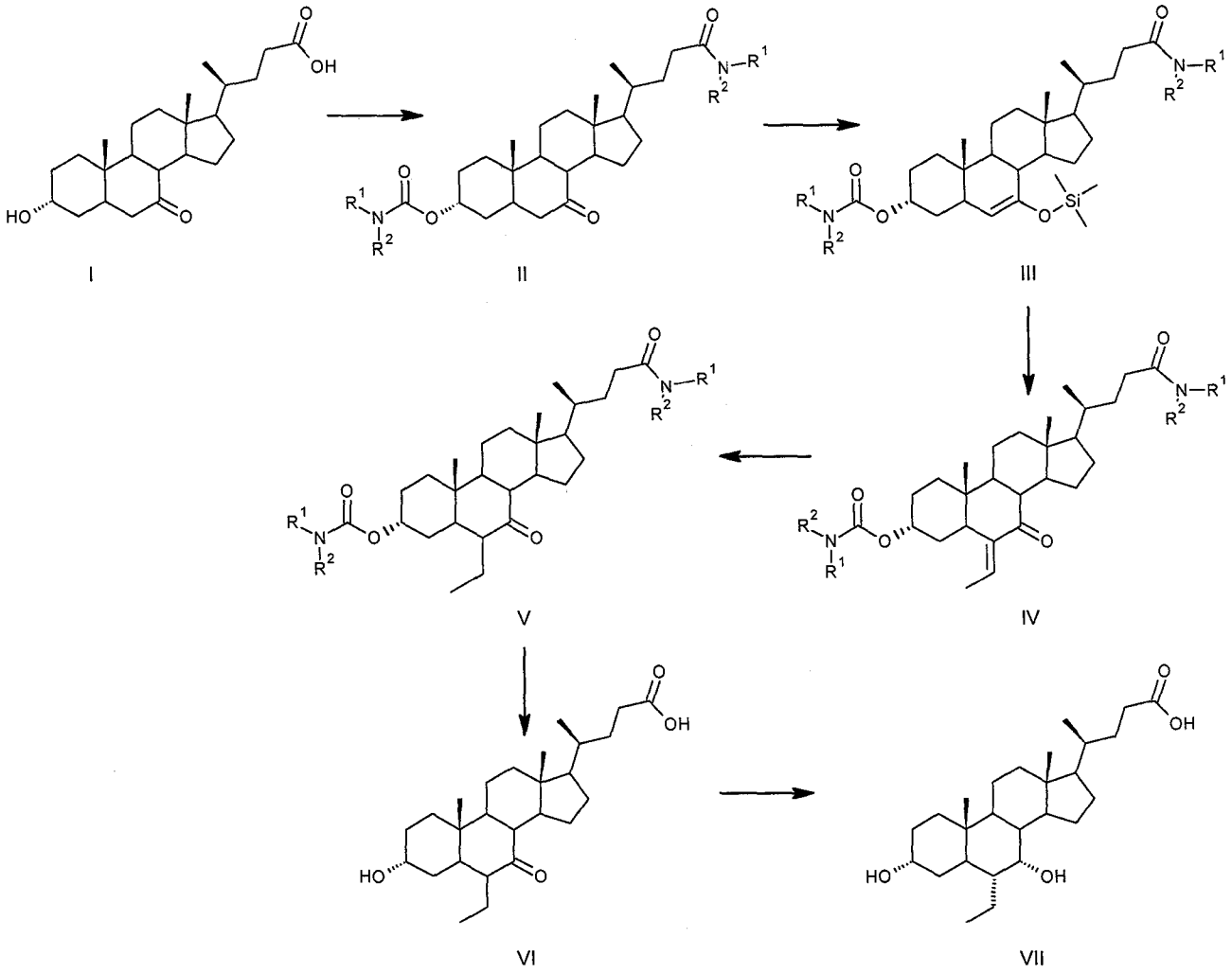
Disclosure of the Invention

It has now been found that the above-mentioned problems will be solved by a process, in which a suitable amide of 7-keto-lithocholic acid is used instead of the ester as the starting compound, said 7-keto-lithocholic acid amide being at the same time protected with a dialkylcarbamoyl group in position 3, wherein the alkyls in the amido and carbamyol groups are the same and the two groups are introduced in one step.

The essence of the present invention consists in conversion of the 7-keto-litocholic acid amide protected in position 3 of formula II, wherein R^1, R^2 are a $C_1 - C_6$ n-alkyl, or together a $C_5 - C_6$ cycle (for example piperidide, pyrrolidide, morpholide), to 7-trimethylsilyl enol ether

of formula III, which is then converted by reaction with an alkylation agent to the 6-ethylidene derivative of formula IV, which is reduced to the compound of formula V and then simultaneously hydrolyzed and deprotected in position 3 to the compound of formula VI and then reduced to obeticholic acid of formula VII.

The amide of formula II is prepared in such a way to protect position 3 by carbamoylation at the same time in one common step (which will shorten the process).



In particular, various steps of the process can be carried out for instance as follows:

Step 1 – The conversion of 7-keto-litocholic acid to the amide with simultaneous protection of the hydroxyl in position 3 can be carried out by reaction of this acid, after suitable activation, with the respective amine. The activated intermediate is prepared by reaction of 7-keto-lithocholic acid with, e.g., DCC-carbonyl diimidazole, hydroxybenzotriazole, or by another method described in the amide preparation literature (Houben-Weyl: Methoden der Organischen Chemie, Bd. E5, G. Thieme, Stuttgart 1985) under the conditions described therein. CDI and a suitable amine can be preferably used because the carbamate protection in position 3 can then be achieved in one step. Thus, one step is saved and the final deprotection is also carried out together with the hydrolysis of the amide.

Step 2 –The preparation of 7-silyl enol ether is conducted via a lithio or sodio enolate by means of an alkali amide, while the basicity of the secondary amine used for preparing the amide must be lower than the basicity (pKa) of LDA and higher than pKa of the ketolithocholic acid 7-enolate itself. The absolute value of pKA depends on the environment (solvent used), advantageously ethers, especially THF. This criterion is met by, e.g., HMDS, diphenylamine, ditolylamine, dinaphthylamine, hexamethyldisilazane, *N*-methylaniline, *N*-methyltoluidine, *N*-isopropylaniline, *N*-tertbutylaniline and other secondary aromatic and silylated amines, preferably HMDS and diphenylamine.

Step 3 – The 6-ethylation can be conducted with common alkylation agents: ethyl bromide, ethyl iodide, ethyl triflate, ethyl mesylate, ethyl tosylate, diethyl sulphate, preferably EtJ and Et triflate with an auxiliary base, e.g. a tertiary amine, preferably Hünig's base. The working methods are known to experimenters skilled in the art.

An alternative comprises two-stage alkylation (aldolization with acetaldehyde and its derivatives) and subsequent reduction, which provides higher yields, does not work with alkylation agents, which are carcinogenic, but adds one step (reduction in the synthetic process). Acetaldehyde or its cyclic derivatives (metaldehyde, paraldehyde) in DCM or ethers are used, preferably in DCM, being catalyzed with strong Lewis acids (BF_3 etherate, $TiCl_4$ and $SnCl_4$), preferably $TiCl_4$ in DCM. The process is conducted at low temperatures, in the range of -80 to -20°C, preferably -60 to -40°C, to prevent retroaldolization. The aldolization is fast, and at the same time slow dehydration to the 6-ethylidene derivative proceeds, which is accelerated by addition of ACN, catalysis with J_2, HCl, TMSCl, $NaHSO_3$, $ZnCl_2$ and other weak Lewis acids, preferably with a combination of ACN and $ZnCl_2$.

The reaction is stopped and back retroaldolization is blocked by complexation of $TiCl_4$ by means of Et_3N or DMF, preferably Et_3N. Titanium salts soluble in the organic phase are removed by precipitation either as TiO_2 or $Ti^{4+}\cdot 2DMF$ or by extraction with α-hydroxy acids into the aqueous phase (tartaric, hydroxyacetic, lactic, citric and other acids, best tartaric acid).

Step 4 – The hydrogenation is conducted in an autoclave on a Pd catalyst (5-10% Pd/C, with or without Cu modification) under the pressure of 1-10 atmospheres, best 5% Pd/C and 3 atmospheres at RT to 100°C, in a solution of methanol, HCl or HAC, best at 40 to 60°C in HAC. After completed hydrogenation the catalyst is filtered off and returned to recycling and the solution is evaporated or neutralized and extracted, after dilution with water, to the organic phase.

Step 5 – The hydrolysis of the amide and deprotection in position 3 is conducted by alkaline hydrolysis (KOH, NaOH, KO*t*-Bu, Cs_2CO_3, KOEt and NaOEt). The hydrolysis is carried out in higher boiling alcohols miscible with water under reflux (n-butanol, amyl alcohol, hexanol, cyclohexanol, ethylene glycol, propylene glycol, ethyl cellosolve, best glycols, KOH, 120°C). After completion of the hydrolysis, it is carefully neutralized and acidified to the free acid and extracted with an organic solvent (DCM, toluene, MTBE, EtOAc). The final product (6-ethyl-7-ketoacid) can be purified by crystallization in the form of salts with amines (diethylamine, diisopropylamine, dibutylamine, *tert*-butylamine, octylamine, *tert*-octylamine,

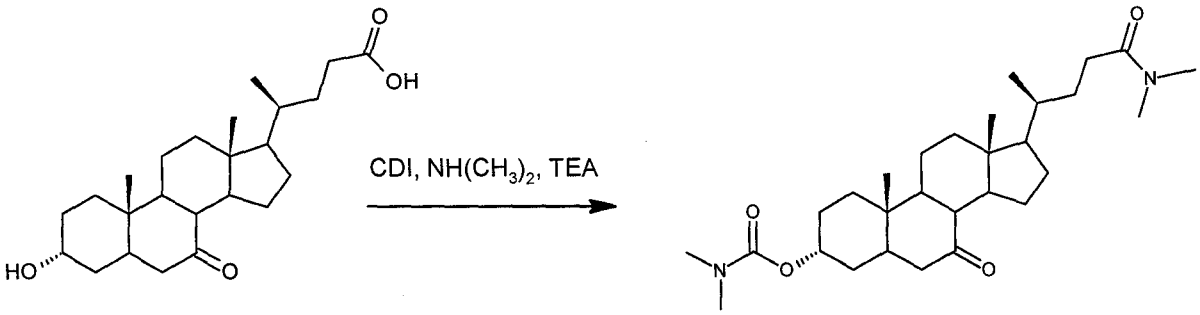
cyclohexylamine, dicyclohexylamine, morpholine, benzylamine, dibenzylamine, *N*-benzylmethylamine, (S)-α-methylbenzylamine, (R)-α-methylbenzylamine, preferably *t*-octylamine, (S)-α-methylbenzylamine).

Step 6 – The process of final reduction with $NaBH_4$ is identical with the basic patent (US7138390). The final product can be crystallized via a *t*-octylamminium salt in an environment of methanol or isopropanol.

The invention is clarified in more detail in the working examples below, which, however, do not restrict its scope.

Examples

Example 1: – Preparation of the 3-dimethylcarbamoyl-7-keto-lithocholic acid dimethylamide

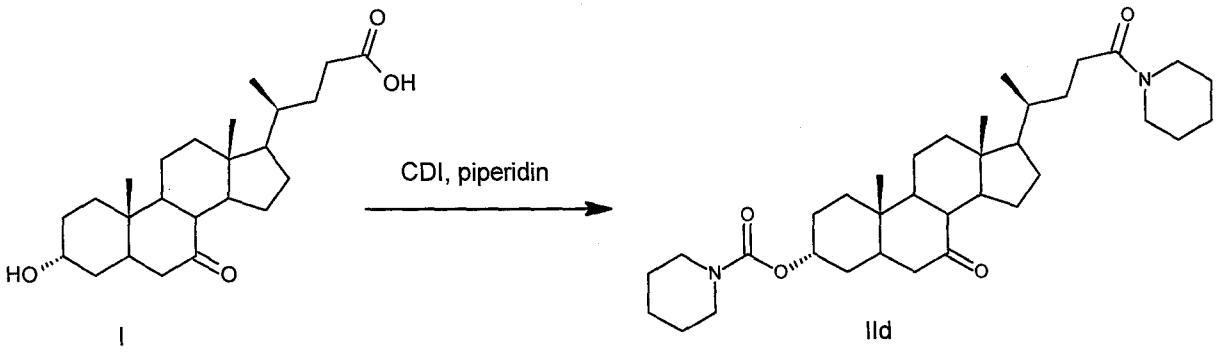


I IIc

In a 500 ml flask annealed *in vacuo* and purged with nitrogen, 16.6 g (102.4 mmol, 2 equiv.) of 1,1'-carbonyldiimidazole was slowly added to 20 g (51.2 mmol, 1 equiv.) of 7-keto-litocholic acid (I) in 120 ml of dichloromethane. The reaction was conducted in an inert atmosphere (N_2), CO_2 being vigorously generated during the reaction. After 1 hour (monitoring with LC/MS) 12.5 g of dried dimethylamine hydrochloride (153.6 mmol, 3 equiv.) was added to the reaction mixture, rinsed with a small amount of dichloromethane, and 21.4 ml of triethylamine (153.6 mmol, 3 equiv.) was added dropwise. The reaction mixture was stirred overnight (16 hours; the reaction half-life is ca. 2 hours). The reaction with dimethylamine can be accelerated by heating under reflux.

After 16 hours, the reaction mixture was stirred with 150 ml of distilled water (5 min), the phases were separated. The organic layer was then shaken with 150 ml of 1M HCl, 150 ml of H_2O, 150 ml of saturated $NaHCO_3$, 150 ml H_2O and 150 ml of brine. The clear solution was dried with Na_2SO_4, filtered and the filtrate was concentrated using a rotary vacuum evaporator (hereinafter RVE only), which provided a white amorphous product (IIc). The crude reaction yield was almost quantitative. Crystallization of the intermediate (IIc) from 60 ml of cyclohexane and 6 ml of toluene. Yield 20.5 g (82%) - white crystalline substance. HRMS (ESI+) calculated for $C_{29}H_{49}N_2O_4$ $[M+H]^+$ 489.3692, found 489.3688.

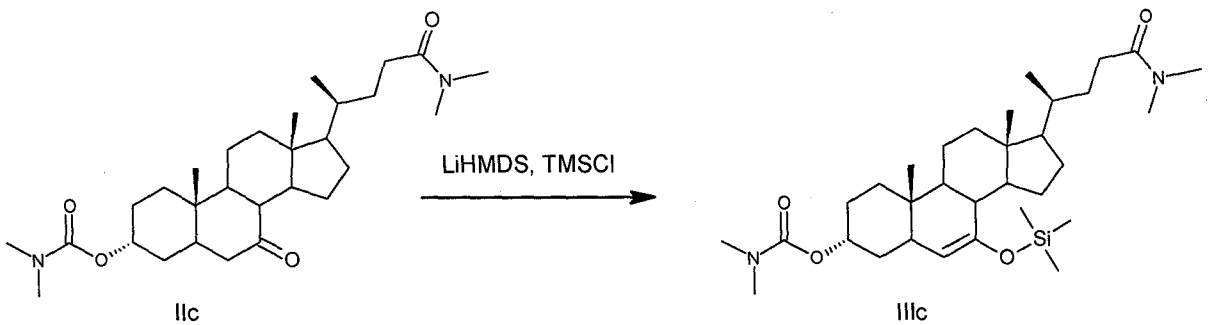
Example 2 – Preparation of 7-keto-lithocholic acid 3-*N,N'*-pentamethylenocarbamoyl piperidide



I IId

In a 500 ml flask annealed *in vacuo* and purged with nitrogen, 16.6 g (102.4 mmol, 2 equiv.) of 1,1'-carbonyldiimidazole was slowly added to 20 g (51.2 mmol, 1 equiv.) of the 7-keto-litocholic acid (I) in 120 ml of dichloromethane. The reaction was conducted under an inert atmosphere (N_2), CO_2 being vigorously generated during the reaction. After 1 hour (LC/MS check) 15.2 ml (153.6 mmol, 3 equiv.) of piperidine was added to the reaction mixture, and rinsed with a small amount of dichlormethane. The reaction mixture was shaken with 3x 150 ml of distilled water after 1 hour, and the phases were separated. The organic phase was shaken with 150 ml of brine; the resulting clear solution was dried with Na_2SO_4, filtered, and the filtrate was concentrated using a rotatory vacuum evaporator (hereinafter RVE only), affording a white amorphous solid (IId). The crude yield of the reaction was almost quantitative. Crystallization of the intermediate (IId) from 60 ml of cyclohexane and 6 ml of toluene. Yield 26.8 g (92%) – white crystalline solid. HRMS (ESI+) calculated for $C_{29}H_{49}N_2O_4$ $[M+H]^+$ 568.424, found 568.431.

Example 3 – Preparation of the TMS-enol ether



IIc IIIc

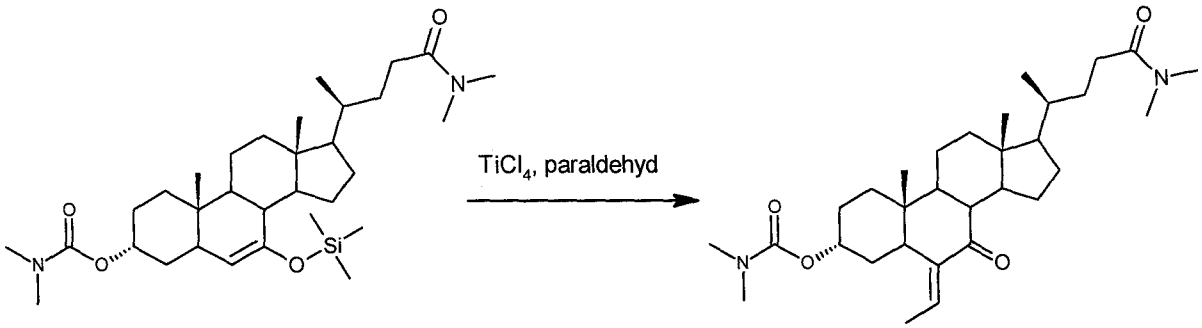
In a 500 ml flask annealed *in vacuo* and purged with nitrogen, 100 ml of dry THF was charged through a septum together with 20.59 ml of hexamethyl disilazane (98.2 mmol, 2.4 equiv.). The mixture was cooled down to -70°C, and at this temperature, 39.3 ml (98.2 mmol 2.4 equiv.) of *n*-BuLi was added dropwise (reaction mixture temperature < -60°C) in an inert atmosphere. After completion of the addition of butyllithium, the stirring and cooling continued for 30 minutes. A solution of 20 g (40.92 mmol, 1 equiv.) of the 7-keto-litocholic acid amide (IIc) in 100 ml of dry THF was added dropwise to lithium bis(trimethylsilyl) amide

in THF at the temperature of -80°C. Then, 12.47 ml of trimethyl silyl chloride (98.2 mmol, 2.4 equiv.) was added dropwise to the mixture and the stirring and cooling continued for 60 minutes.

200 ml of water was poured to the mixture, the mixture was removed from the bath and stirred for 10 minutes. The layers were separated, the aqueous layer was shaken with 3x 50 ml of toluene. The combined organic fractions were shaken with 150 ml of a solution of $NaHCO_3$ (saturated) : H_2O (1:1), 150 ml of H_2O and 150 ml of brine. Dried with Na_2SO_4, the desiccant was filtered off and the filtrate was concentrated using an RVE at T_{max} = 30°C to obtain an oily evaporation product. The crude reaction yield was almost quantitative. The product (IIIc) is a white amorphous substance. Crystallization of the intermediate from 65 ml of cyclohexane. Yield 21.1 g (92%) - white crystalline substance. HRMS (ESI+) calculated for $C_{32}H_{57}N_2O_4Si$ $[M+H]^+$ 561.4088, found 561.4094.

The preparation of the lithioenolate can also be carried out with the commercially available lithium bis(trimethylsilyl) amide.

Example 4 – Alkylation of the TMS-enol ether



IIIc IVb

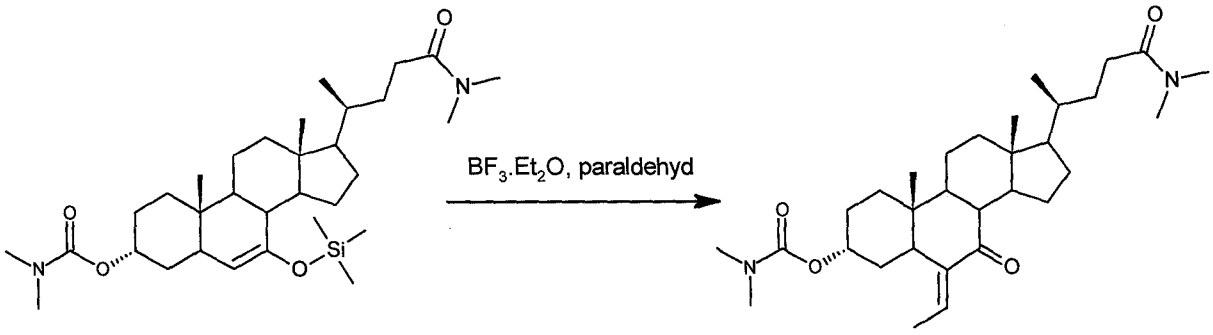
A 1L sulfonation flask with a mechanical stirrer was annealed *in vacuo* and purged with nitrogen. After cooling off the flask, 120 ml of dried dichloromethane (dried using an A4 molecular sieve) and 1.75 ml (13.2 mmol, 0.37 equiv.) of paraldehyde were charged in a nitrogen atmosphere. The solution was cooled down to -60°C. Then, 19.55 ml of $TiCl_4$ (178.3 mmol, 5 equiv.) was added dropwise and a solution of 20 g (35.66 mmol, 1 equiv.) of the TMS-enol ether (IIIc) in 120 ml of dichloromethane (R.m. T maintained at -60 to -55°C) was added dropwise to the slightly yellowish suspension. The primarily produced suspension got dissolved, providing an orange to brown solution.

After 10 min from the addition of the TMS-enol ether (IIIc), the reaction was stopped by adding a solution of 39.76 ml (285.27 mmol, 8 equiv.) of triethylamine in 200 ml of acetonitrile.

The temperature of the reaction mixture was adjusted to the room temperature within 15 min and then 1.93 ml of distilled water (106.97 mmol, 3 equiv.) and 486 mg (3.57 mmol, 10 mol %) of zinc chloride were added. In 30 min after the addition of $ZnCl_2$, the product got completely dehydrated to (IVb). 300 ml of MTBE and 300 ml of water were poured to the

mixture, stirred, the phases were separated and the organic layer was stirred with 14.12 ml (106.97 mmol, 3 equiv.) of triethanolamine for 10 min. 200 ml of water were then added to the mixture, and stirred for 10 min. The phases were separated, the organic layer was stirred with 4x 350 ml of 20% tartaric acid (4x 20 min). Then, the organic phase was shaken with 300 ml of water, 300 ml of a saturated solution of $NaHCO_3$, 300 ml of water and 300 ml of brine. Dried with Na_2SO_4. The desiccant was filtered off and the filtrate was concentrated in an RVE. Crude reaction yield 15.2 g (83%) of (IVb). The crude intermediate was purified by stirring with Al_2O_3 and then with active charcoal. Crystalization was made from the mixture diisopropylether : cyclohexane (10 : 1) in the ratio of 3 ml of solvent per 1 g of the substance. HRMS (ESI+) calculated for $C_{31}H_{51}N_2O_4$ $[M+H]^+$ 515.3849, found 515.3855.

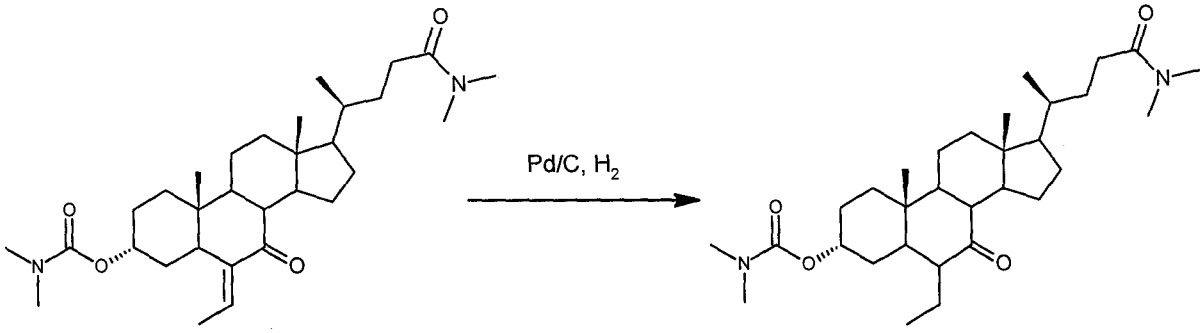
Example 5 – Alkylation of the TMS-enol ether



IIIc IVb

120 ml of dry DCM and 1.75 ml (13.2 mmol, 0.37 equiv.) of paraldehyde were charged in an annealed 1L sulfonation flask in a nitrogen atmosphere. The solution was cooled down to -60°C. Then, 47.3 ml of $BF_3{\cdot}Et_2O$ (≥46.5 BF_3 basis, 178.3 mmol, 5 equiv.) was added and subsequently a solution of 20 g (35.66 mmol, 1 equiv.) of the TMS-enol ether (IIIc) in 120 ml of DCM was added dropwise. After stirring for 30 min at T = -60°C, the temperature of the mixture was adjusted to the room temperature and the mixture was stirred for another 1 hour. The mixture was cooled down to 0°C, diluted with 150 ml of distilled water and extracted with toluene (3x 100 ml). The combined organic fractions were shaken with 150 ml of water, 150 ml of a saturated solution of $NaHCO_3$, 150 ml of water and 150 ml of brine. Dried with Na_2SO_4, the desiccant was filtered off and the filtrate was concentrated in an RVE. The yield was 15 g (82%) of crude (IVb). The crude intermediate was purified by stirring with Al_2O_3 and then with active charcoal. Crystallization was made from the mixture diisopropylether : cyclohexane (10 : 1) in the ratio of 3 ml of solvent per 1 g of the substance. HRMS (ESI+) calculated for $C_{31}H_{51}N_2O_4$ $[M+H]^+$ 515.3849, found 515.3855.

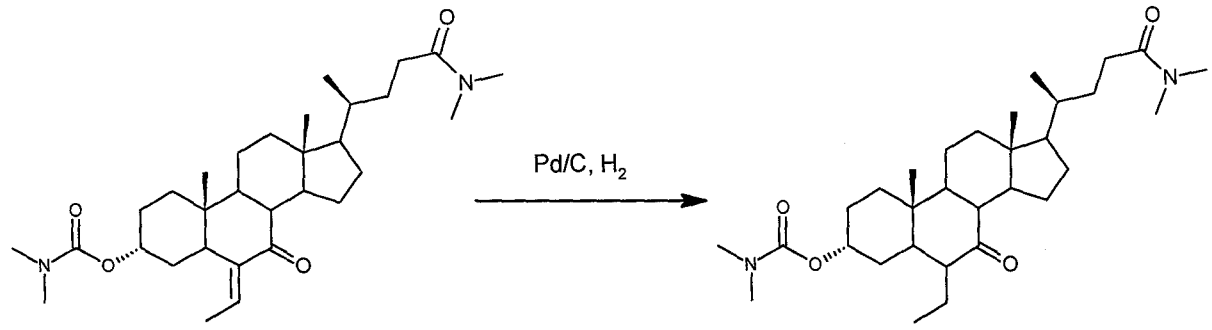
Example 6 – Hydrogenation of the 6-ethylidene amide (IVb)



IVb V

A solution of 20 g (38.85 mmol, 1 equiv.) of (IVb) in 180 ml of MeOH and 40 ml H_2O was charged in an autoclave, 3.4 ml of 35% HCl (38.85 mmol, 1 equiv.) and 2 g of Pd/C (5% by weight) were added. The autoclave was pressurized 3 times and purged with N_2 (10 atm), 3x H_2 (10 atm). At the pressure of 10 atm, the mixture was hydrogenated for 3 hours at T = 40°C. The catalyst was filtered off on a paper filter, rinsed with 20 ml of MeOH. The filtrate was heated at T = 50°C for 1 hour until the "undesired" β-epimer disappeared. MeOH was evaporated from the mixture in an RVE and then 100 ml of MTBE and 100 ml of H_2O were added to the mixture and the mixture was extracted. The aqueous phase was shaken with 2x 50 ml of MTBE, the combined organic phases were washed with 100 ml of water, 100 ml of brine, dried with Na_2SO_4, the desiccant was filtered off and the filtrate was concentrated in an RVE. Reaction yield 14.5 g (72%) of (V). HRMS (ESI+) calculated for $C_{31}H_{53}N_2O_4$ $[M+H]^+$ 517.3994, found 517.4000.

Example 7 – Hydrogenation of the 6-ethylidene amide (IVb)

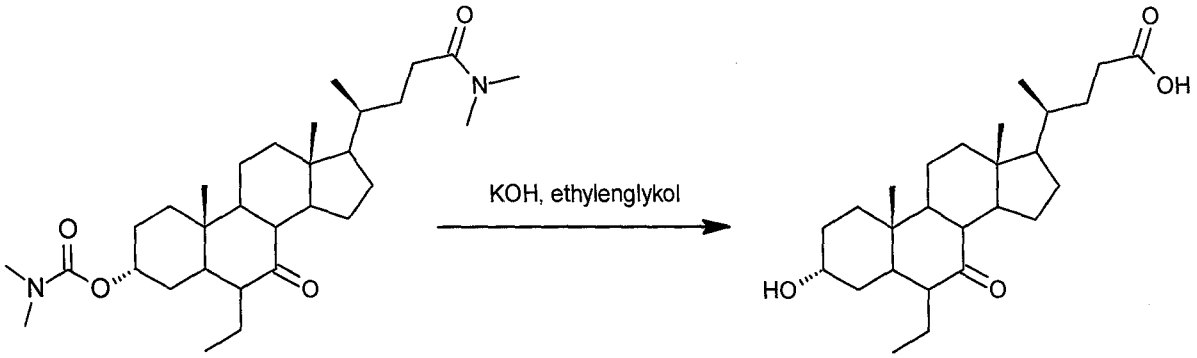


IVb V

A solution of 20 g (38.85 mmol, 1 equiv.) of (IVb) in 140 ml of 80% AcOH was charged in an autoclave together with 2 g of 5% Pd/C (10% by weight). The autoclave was pressurized 3 times and purged with N_2 (3 atm), 3x H_2 (3 atm). At the pressure of 3 atm, the mixture was hydrogenated at T = 50°C for 2 hours. The catalyst was filtered off on a paper filter, rinsed with 20 ml of 80% AcOH. The filtrate was diluted with water, neutralized with a 2M solution of NaOH to pH 5-6, then extracted 3 times with 70 ml of MTBE. The combined organic phases were washed with 100 ml of water, 2x 100 ml of a 2M solution of NaOH, 100 ml of water and

100 ml of brine. Dried with Na_2SO_4, the desiccant was filtered off and the filtrate was concentrated in an RVE. Reaction yield 15.9 g (79%) of (V). HRMS (ESI+) calculated for $C_{31}H_{53}N_2O_4$ $[M+H]^+$ 517.4000, found 517.4004.

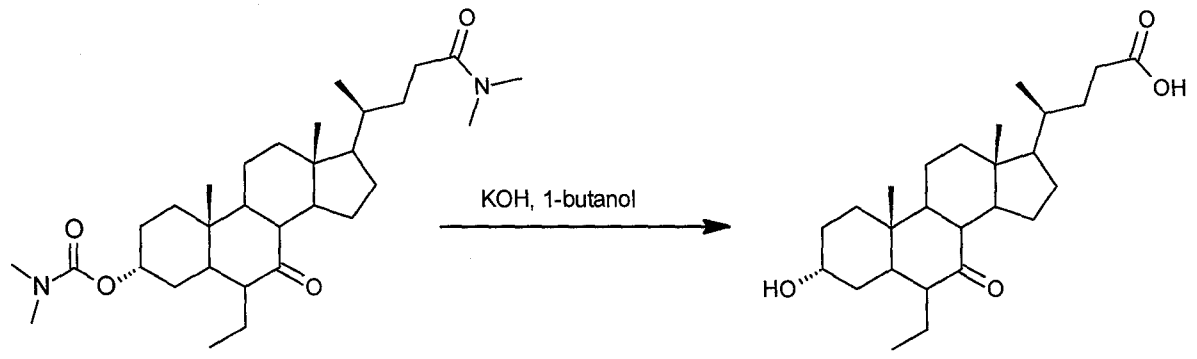
Example 8 – Hydrolysis of the amidic groups of (V)



V VI

20 g (38.7 mmol, 1 equiv.) of (V) in 200 ml of ethylene glycol was dissolved in a 500ml flask and then 13 g (232.2 mmol, 6 equiv.) of KOH was added. The mixture was heated under reflux at 120°C for 5 hours, then cooled down to r.t. and neutralized with 2M HCl to pH = 7. The reaction mixture was extracted with 3x 60 ml of EtOAc. The combined organic fractions were washed with 2x 100 ml of water, 100 ml of brine and dried with Na_2SO_4. The desiccant was filtered off and the filtrate was concentrated using an RVE. Reaction yield 14.1 g (87%) of crude (VI). Crystallization of the intermediate via the amminium salt: 10 ml MeOH, 1.02 equiv. of (S)-α-methylbenzylamine for 1 g of (VI); 10 ml of H_2O added under reflux. Then, the mixture was refluxed for 30 minutes, and left to cool down freely. The product after isolation was converted back to the free acid via the sodium salt with NaOH, then extracted, using 1M HCl, into EtOAc. The solvent was concentrated to the ratio 3 g of EtOAc : 1 g of (VI); after cooling down, a white crystalline substance was isolated. HRMS (ESI+) calculated for $C_{26}H_{43}O_4$ $[M+H]^+$ 419.3156, found 419.3161.

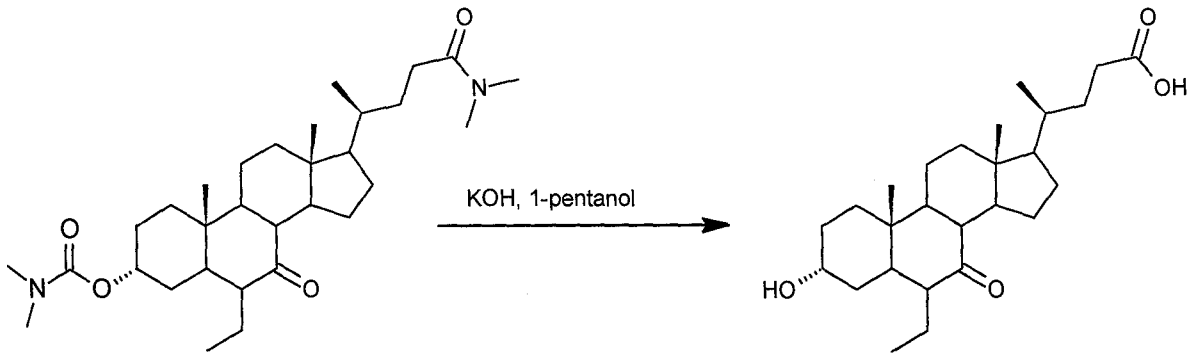
Example 9 – Hydrolysis of the amidic groups of (V)



V VI

20 g (38.7 mmol, 1 equiv.) of (V) was dissolved in 150 ml of 1-butanol in a 500ml flask and then 13 g (232.2 mmol, 6 equiv.) of KOH was added. The mixture was refluxed under a condenser for 10 hours, then cooled down to r.t.. 330 ml of distilled water and 200 ml of cyclohexane were added, shaken, phases separated and the aqueous layer was additionally shaken with 2x 100 ml of cyclohexane. Then the aqueous layer was neutralized with 2M HCl to pH = 2-3. The reaction mixture was extracted with 1x 150 ml and 2x 50 ml of EtOAc. The combined organic fractions were washed with 2x 100 ml of water, 100 ml of brine and dried with Na_2SO_4. The desiccant was filtered off and the filtrate was concentrated using an RVE. Reaction yield 13.5 g (83%) of crude (VI). Crystallization of the intermediate via the amminium salt: 10 ml MeOH, 1.02 equiv. of (S)-α-methylbenzylamine for 1 g of (VI), 10 ml of H_2O added under reflux. Then, the mixture was refluxed for 30 minutes, and left to cool down freely. The product after isolation was converted back to the free acid via the sodium salt with NaOH, and then extracted, using 1M HCl, into EtOAc. The solvent was concentrated to the ratio of 3 g of EtOAc : 1 g of (VI); after cooling down, a white crystalline substance was isolated. HRMS (ESI+) calculated for $C_{26}H_{43}O_4$ $[M+H]^+$ 419.3156, found 419.3162.

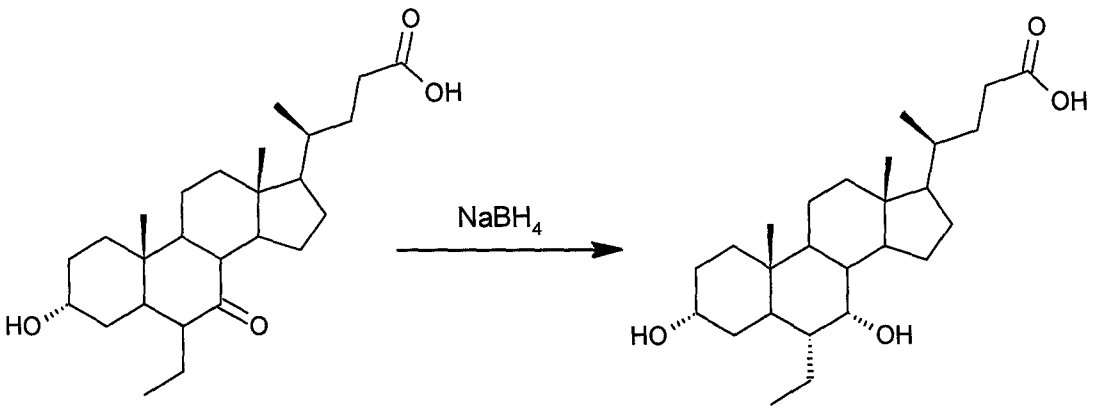
Example 10 – Hydrolysis of the amidic groups of (V)



V VI

20 g (38.7 mmol, 1 equiv.) of (V) was dissolved in 150 ml of 1-pentanol in a 500 ml flask and then 13 g (232.2 mmol, 6 equiv.) of KOH was added. The mixture was refluxed under a condenser for 3 hours, then cooled down to r.t., diluted with 50 ml of heptane and 50 ml of water. A heptane layer, product layer and aqueous layer were formed. The oily product was diluted with 150 ml of EtOAc, washed with 2x 100 ml of water, 100 ml of brine, dried with Na_2SO_4. The desiccant was filtered off and the filtrate was concentrated using an RVE. Reaction yield 14.6 g (90%) of crude (VI). Crystallization of the intermediate via the amminium salt: 10 ml MeOH, 1.02 equiv. of (S)-α-methylbenzylamine for 1 g of (VI); 10 ml of H_2O added under reflux. Then, the mixture was refluxed for 30 minutes, and left to cool down freely. The product after isolation was converted back to the free acid via the sodium salt with NaOH, then extracted, using 1M HCl, into EtOAc. The solvent was concentrated to the ratio 3 g of EtOAc : 1 g of (VI); after cooling down, a white crystalline substance was isolated. HRMS (ESI+) calculated for $C_{26}H_{43}O_4$ $[M+H]^+$ 419.3156, found 419.3159.

Example 11 – Reduction of the 7-keto group of (VI) with $NaBH_4$

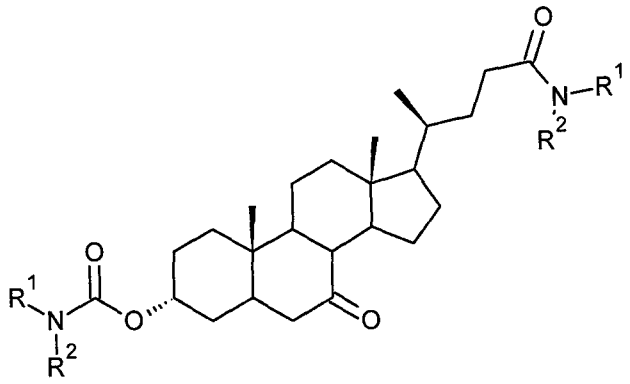


VI VII

20 g (47.78 mmol, 1 equiv.) of (VI) was dissolved in 200 ml of a 50% solution of ethanol with water in a 500 ml round flask. 3.82 g (95.55 mmol, 2 equiv.) of NaOH was added. Then, 7.23 g (191.11 mmol, 4 equiv.) of $NaBH_4$ was added in parts. The mixture was refluxed at the temperature of 80°C for 3 hours. Then it was cooled down to r.t., acidified with 2 M HCl to pH = 2-3, the temperature of the reaction mixture being kept below 30°C all the time. Then 100 ml of *i*-PrOAc was added, shaken, phases separated; the aqueous layer was extracted with 2x 50 ml of *i*-PrOAC. The combined organic extracts were washed with 4x 50 ml of distilled water and dried with Na_2SO_4. After filtration, 3x 50 ml of MaOH was gradually added during evaporation of the filtrate in order to azeotropically remove excess *i*-PrOH in an RVE (T = 40°C). Subsequently, the mixture was additionally diluted with methanol such that 60 ml of MeOH come to theoretical 20.1 g. Purification of the final product was made by crystallization in the form of a salt with 1,1,3,3-tetramethylbutylamine. 3 ml of MeOH, 1.02 equiv. of 1,1,3,3-tetramethylbutylamine (*tert*-octylamine) for 1 g of (VII); 10 ml of H_2O added under reflux. Then, the mixture was refluxed for 30 minutes, and left to cool down freely. The product after isolation was converted back to the free acid via the sodium salt with NaOH, then extracted into *i*-PrOAc using 1M HCl. The organic phase was washed with 3x 100 ml of water, 1x 100 ml of brine, and dried with Na_2SO_4. The resulting filtrate was concentrated in an RVE to produce a dense oily evaporation residue, which was used for preparation of an amorphous product. HRMS (ESI+) calculated for $C_{26}H_{45}O_4$ $[M+H]^+$ 421.3312 found 421.3316.

Claims:

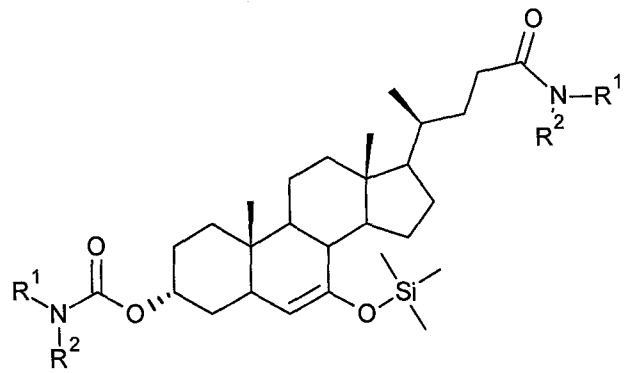
1. A process for preparing obeticholic acid, characterized in that the 7-keto-litocholic acid amide, protected with dialkylcarbamoyl group in position 3, of formula II



II

,

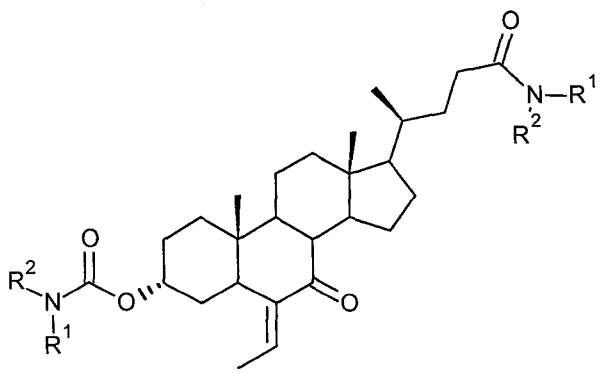
wherein R^1, R^2 are a $C_1 - C_6$ n-alkyl, or together a $C_5 - C_6$ cycle and wherein the substituents on the amidic and carbamoyl nitrogens are identical, is converted to the 7-trimethylsilyl enol ether of formula III



III

,

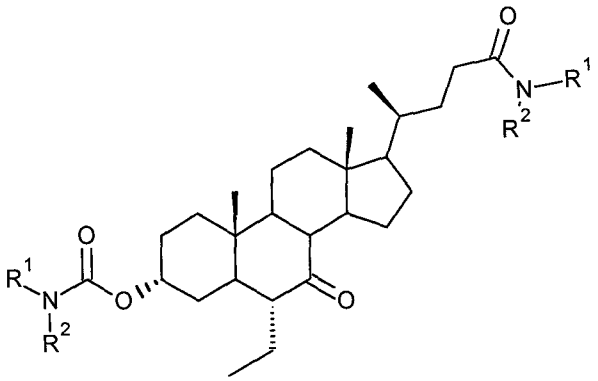
which is then converted by reaction with an alkylation agent to the 6-ethylidene-derivative of formula IV



IV

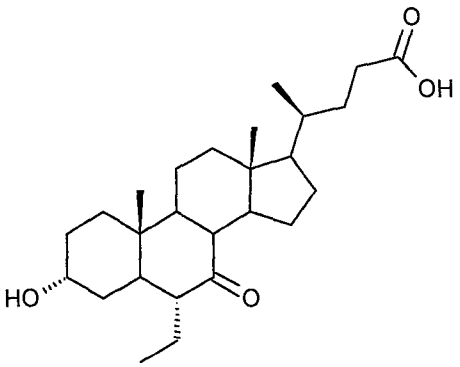
,

which is reduced to the compound of formula V



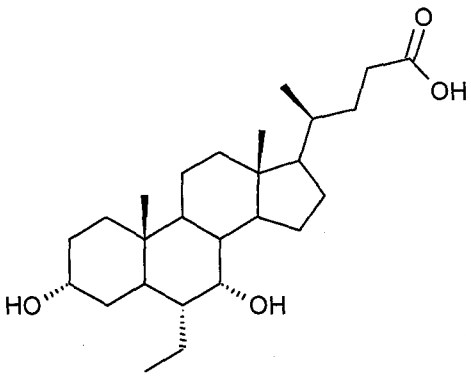
V

and then simultaneously deprotected in position 3 to the compound of formula VI



VI

and reduced to obeticholic acid of formula VII



VII

2. The process according to claim 1, characterized in that a base with suitable basicity expressed as pKa is used for the preparation of the lithium enolate in the conversion of the amide of formula II to 7-silyl enol ether III, e-g. lithium diphenylamide, lithium

bis-ortho-, bis-meta-, bis-para-tolylamide, lithium N,N-dimethyl-, N,N-diethyl-, N,N-di-*tert*-butyl-anilide, lithium hexamethyl disilazide.

3. The process according to claim 2, characterized in that lithium hexamethyl disilazide is used as the base for the preparation of the lithium enolate.

4. The process according to claims 2 or 3, characterized in that 7-silyl enol ether is purified by crystallization from cyclohexane.

5. The process according to claim 1, characterized in that paraldehyde is used as the alkylation agent for the alkylation of silyl enol ether III.

6. The process according to claim 1, characterized in that acetaldehyde is used as the alkylation agent for the alkylation of silyl enol ether III.

7. The process according to claim 5 or 6, wherein the alkylation catalyst is a Lewis acid.

8. The process according to claim 7, wherein the Lewis acid is $TiCl_4$, BF_3 etherate, $ZrCl_4$, $SnCl_4$, $AlCl_3$, or a group of rare earth triflates.

9. The process according to claim 8, wherein the catalyst is $TiCl_4$.

10. The process according to claim 9, wherein a complex of the catalyst with acetaldehyde or paraldehyde is first prepared at a temperature of 0 to -80°C.

11. The process according to claim 10, wherein the temperature range of preparation of the catalytic complex is -40 to -70°C.

12. The process according to claim 11, wherein the temperature of preparation of the catalytic complex is in the range of -50 to -60°C.

13. The process according to claim 1, characterized in that the 6-(1-hydroxyethyl) derivative obtained from 7-silyl enol ether III is dehydrated to the ethylidene derivative IV, wherein quantitative dehydration without retroaldolization is achieved by means of $ZnCl_2$.

14. The process according to claim 13, characterized in that the dehydration is conducted in an acetonitrile environment.

15. The process according to claim 1, characterized in that deprotection is carried out by simultaneous hydrolysis of the dialkylamide and dialkylcarbamoyl groups of the intermediate V in a basic environment.

16. The process according to claim 15, wherein the base used is from the group of Na methylate, Na ethylate, potassium hydroxide, K ethylate, and K *tert*-butylate.

17. The process according to claim 16, wherein the base used is KOH in ethylene glycol, or propylene glycol, or ethyl cellosolve.

18. The process according to claim 16, wherein the base used is KOH in *n*-butanol or amyl alcohol.

19. The process according to any one of claims 16-18, wherein the media temperature is between 50 and 140°C or at the reflux of the solvent.

20. The process according to claim 1, characterized in that the intermediate VI is purified by crystallization as an amminium salt with amines, which are especially diethylamine, diisopropylamine, dibutylamine, *tert*-butylamine, *n*-octylamine, *tert*-octylamine, cyklohexylamine, dicyclohexylamine, morpholine, benzylamine, dibenzylamine, *N*-benzylmethylamine, (S)-α-methylbenzylamine, or (R)-α-methylbenzylamine).

21. The process according to claim 20, wherein the amminium salt of intermediate VI is with *tert*-octylamine, (S)-α-methylbenzylamine, or (R)-α-methylbenzylamine.

22. The process according to claim 21, wherein the amminium salt is with (R)-α-methylbenzylamine.

23. The process according to claim 1, characterized in that obeticholic acid VII is purified by crystallization as an amminium salt with *tert*-octylamine (1,1,3,3-tetramethylbutylamine).

24. The compound II, wherein R^1, R^2 are methyl.

25. The compound V, wherein R^1, R^2 are methyl.

INTERNATIONAL SEARCH REPORT

International application No
PCT/CZ2019/000009

A. CLASSIFICATION OF SUBJECT MATTER
INV. C07J9/00 C07J41/00 C07J51/00
ADD.

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
C07J

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
EPO-Internal, CHEM ABS Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	CN 104 558 086 B (KANGMEI BEIJING DRUG RES INST CO LTD ET AL.) 5 October 2016 (2016-10-05) page 3 -----	1-25
A	CN 107 383 139 A (HANGZHOU HEZE PHARMACEUTICAL TECH CO LTD) 24 November 2017 (2017-11-24) examples 8, 17 -----	1-25
A	A BARRETT ET AL: "The Deoxygenation of N,N-DialkylaminothiocarbonyloxyalkanES", JOURNAL OF THE CHEMICAL SOCIETY, PERKIN TRANSACTIONS I, 1 January 1981 (1981-01-01), pages 1510-1515, XP55593573, DOI: doi.org/10.1039/P19810001510 page 1511; table 1; compounds 3c, 3g, 3h -----	1-25

☐ Further documents are listed in the continuation of Box C.

☒ See patent family annex.

* Special categories of cited documents :

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"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

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"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
4 June 2019	19/06/2019

Name and mailing address of the ISA/	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Watchorn, Peter

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/CZ2019/000009

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
CN 104558086 B	05-10-2016	NONE	
CN 107383139 A	24-11-2017	NONE	