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(54) Title: SOLID FORMS OF TENAPANOR AND METHOD OF PREPARATION OF TENAPANOR

(57) Abstract: The invention relates to solid forms of tenapanor, in particular to crystalline form I of tenapanor free base and to solid tenapanor salts, and to methods of preparation thereof. The crystalline form I is the first known crystalline form of tenapanor. The novel solid forms of tenapanor can be used for purification of tenapanor and/or in pharmaceutical compositions. Furthermore, the invention provides a method of preparation of crystalline tenapanor which is economically effective and capable of being scaled up.

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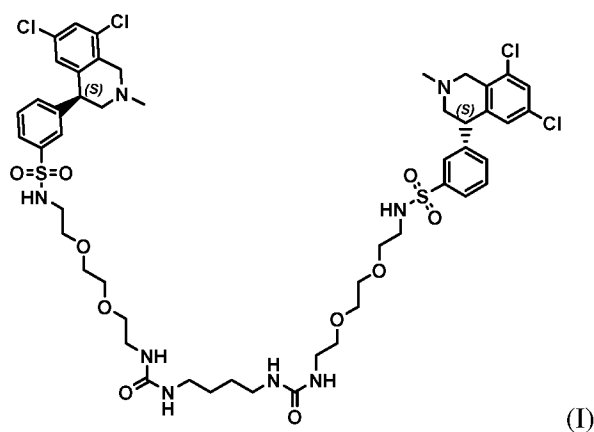
Solid forms of tenapanor and method of preparation of tenapanor

Field of Art

The present invention relates to a stable form of tenapanor free base, stable salts of tenapanor, and use thereof in a dosage form. Furthermore, the present invention relates to methods of production of crystalline tenapanor.

Background Art

Tenapanor, having the chemical name 17-[[[3-[(4S)-6,8-dichloro-1,2,3,4-tetrahydro-2-methyl-4-isoquinoliny]phenyl]sulphonyl]amino]-N-[2-[2-[2-[[[3-[(4S)-6,8-dichloro-1,2,3,4-tetrahydro-2-methyl-4-isoquinoliny]phenyl]sulphonyl]amino]ethoxy]ethoxy]ethyl]-8-oxo-12,15-dioxo-2,7,9-triazaheptadecaneamide, is a selective inhibitor of the sodium protonic NHE3 antiporter. Orally administered tenapanor selectively inhibits the absorption of sodium in the intestine. This leads to an increase of water content in the digestive tract, improved bowel flow and normalization of the frequency of bowel movement and stool consistency. At the same time it exhibits antinociceptive activity and ability to lower serum phosphate levels. Because of these properties, it is clinically tested for the treatment of irritable bowel syndrome, especially when accompanied by constipation, treatment of hyperphosphatemia, especially in patients with dialysis with final stage renal failure, treatment of chronic kidney disease, and prevention of excess sodium in patients with kidney and heart conditions. The tenapanor molecule, which was first described in the international patent application WO 2010/078449, has the following structural formula:

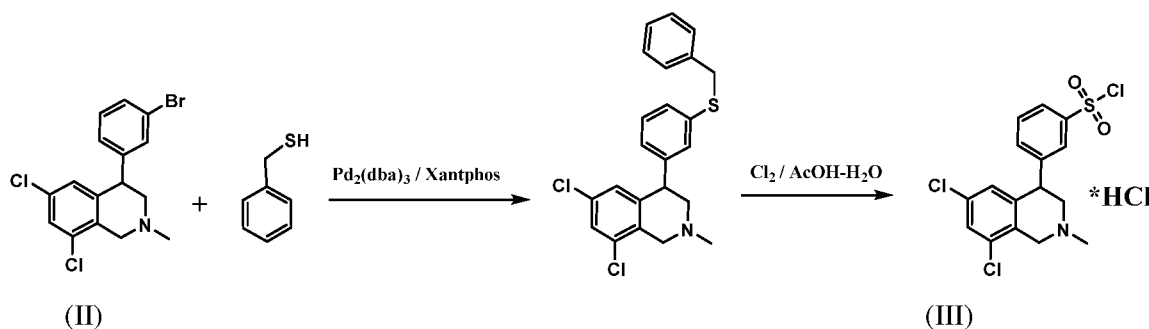


In this document, tenapanor was prepared as bishydrochloride salt. The bishydrochloride salt was prepared only in the form of an amorphous foam, which, after solidification, required grinding for further processing. However, the thus obtained particles are of varying sizes, while a narrow particle size distribution is required for pharmaceutical use in order to ensure uniform behavior. The amorphous foam obtained in the said document is essentially a thickened reaction mixture or a slightly

purified reaction mixture containing, in addition to tenapanor, various impurities. The possibilities to purify the reaction mixtures are limited. Moreover, amorphous foams tend to adsorb solvents, and it is usually difficult to remove (or dry out) the residual solvents from the amorphous foam. This is undesirable for pharmaceutical use. A typical feature of amorphous foams is a large specific surface, resulting in a greater interaction of the substance with the surrounding environment. This significantly increases the risk of decomposition of the substance, for example through air oxygen, moisture or light. The present invention aims at overcoming these problems.

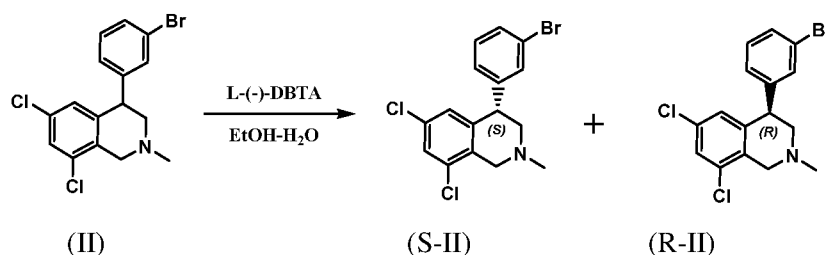
It would be advantageous to provide tenapanor solid forms (tenapanor free base or tenapanor salts) which are precipitated in solid forms, thus allowing to filter off the liquid reaction mixture containing the impurities. This results in a significantly improved purity.

The process used in WO 2010/078449 for the preparation of bishydrochloride salt of tenapanor was based on the preparation of 3-(6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinolin-4-yl)benzene-1-sulfonyl chloride of formula III from 4-(3-bromophenyl)-6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinoline of formula II according to Scheme 1.



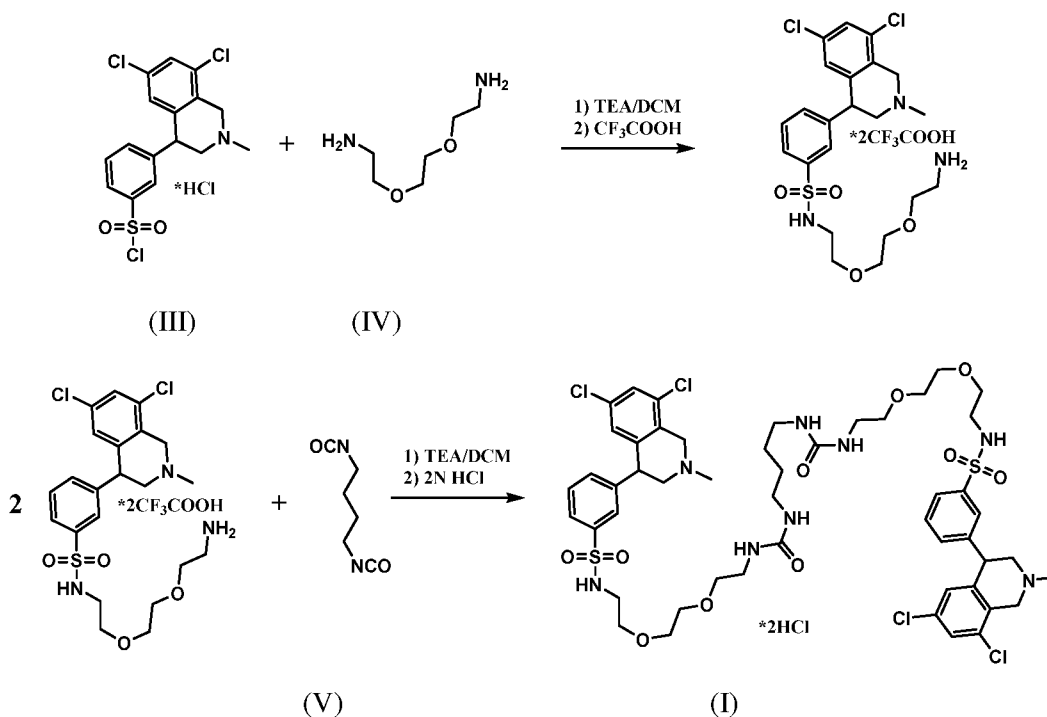
Scheme 1

The said document also discloses resolution of the starting tetrahydroisoquinoline of formula II by L- or D-dibenzoyltartaric acid as shown in Scheme 2.



Scheme 2

WO 2010/078449 discloses further steps of preparation of tenapanor, as shown in Scheme 3.



Scheme 3

Individual synthetic steps described in Scheme 3 result in low yields: 42% for the reaction of the chloride of formula III with 2-(2-(2-aminoethoxy)ethoxy)ethylamine of formula IV, and 59% for the subsequent reaction with 1,4-diisocyanatobutane of formula V. The products of both synthetic steps are isolated by preparative chromatography which is technologically an unsuitable isolation and purification technique. The low yields and the need to use preparative chromatography for the isolation are caused by an abundance of side products and impurities and by the inability of the intermediates as well as of the product to provide a crystalline form.

Thus present invention thus further aims at providing a method of preparation of tenapanor which would be economically effective, in particular in relation to the expensive starting compound 4-(3-bromophenyl)-6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinoline, and which would also enable industrial scale production, in particular by removing steps which cannot be scaled up effectively or which cannot be scaled up at all. Furthermore, the method of preparation of tenapanor should provide tenapanor in a form which is useful for use in pharmaceutical forms and does not have the disadvantages of an amorphous foam.

Disclosure of the Invention

The present invention provides a crystalline form of tenapanor free base which has a high stability and can be prepared in a simple manner. Unlike the bishydrochloride salt known in the art, the described

form exhibits a high crystallinity and can therefore advantageously be used both for the preparation of high purity tenapanor and into a dosage form. A crystalline form of tenapanor has not yet been prepared in the art.

Object of the present invention is tenapanor free base in crystalline form.

In a preferred embodiment, the tenapanor free base is in crystalline form I which exhibits characteristic reflections in the X-ray powder diffraction spectrum obtained using $\text{CuK}\alpha$ radiation: 11.1; 19.8 and $22.7 \pm 0.2^\circ$ 2-theta. In a particularly preferred embodiment, the crystalline form I exhibits the following further characteristic reflections in the X-ray powder diffraction spectrum obtained using $\text{CuK}\alpha$ radiation: 13.2; 15.5; 19.4; 22.1 and $24.4 \pm 0.2^\circ$ 2-theta.

In a more preferred embodiment, the crystalline form I exhibits characteristic reflections in the X-ray powder diffraction spectrum obtained using $\text{CuK}\alpha$ radiation: 11.1; 11.7; 13.2; 13.5; 15.0; 15.5; 15.9; 17.2; 19.4; 19.8; 20.8; 22.1; 22.7; 23.0; 23.3; 23.7; 24.4 and 24.9.

In a preferred embodiment, the crystalline form I exhibits the differential scanning calorimetry curve with a melting point at the temperature $130.5 \pm 2^\circ\text{C}$.

Preferably, the crystalline form I exhibits characteristic reflections in the X-ray powder diffraction spectrum obtained using $\text{CuK}\alpha$ radiation: 11.1; 19.8 and $22.7 \pm 0.2^\circ$ 2-theta, as well as the differential scanning calorimetry curve with a melting point at the temperature $130.5 \pm 2^\circ\text{C}$. In a particularly preferred embodiment, the crystalline form I exhibits the following further characteristic reflections in the X-ray powder diffraction spectrum obtained using $\text{CuK}\alpha$ radiation: 13.2; 15.5; 19.4; 22.1 and $24.4 \pm 0.2^\circ$ 2-theta.

In a more preferred embodiment, the crystalline form I exhibits characteristic reflections in the X-ray powder diffraction spectrum obtained using $\text{CuK}\alpha$ radiation: 11.1; 11.7; 13.2; 13.5; 15.0; 15.5; 15.9; 17.2; 19.4; 19.8; 20.8; 22.1; 22.7; 23.0; 23.3; 23.7; 24.4 and 24.9, as well as the differential scanning calorimetry curve with a melting point at the temperature $130.5 \pm 2^\circ\text{C}$.

The novel crystalline form of tenapanor according to the present invention can be used in the preparation of pharmaceutical compositions, in particular solid, semi-solid and liquid dosage forms, in particular solid dosage forms, e.g. tablets. Such pharmaceutical compositions may contain at least one excipient selected from the group consisting of fillers (e.g., lactose), binders (e.g., microcrystalline cellulose), disintegrants (e.g., croscarmellose sodium), lubricants (e.g., magnesium stearate),

surfactants. Furthermore, the pharmaceutical compositions may optionally comprise at least one other excipient selected from the group consisting of colorants, solvents, antimicrobial agents, flavorings and olfactory adjusting agents. Tablets may be coated with conventional coatings containing, for example, polyvinyl alcohol or polyethylene glycol.

The pharmaceutical compositions can be prepared in virtually any solid dosage form, e.g. in the form of tablets, capsules, powders, pellets or granules. A preferred dosage form is a tablet or a coated tablet. The tablet or coated tablet may preferably be prepared by mixing tenapanor with at least one pharmaceutically acceptable excipient and subjecting the mixture to tableting, optionally followed by coating.

The content of the active ingredient in the tablet may range from 1 to 100 mg, preferably the tenapanor content in the tablet is within the range of 1 to 50 mg or 10 to 50 mg or 15 to 50 mg or 20 to 50 mg or 30 to 50 mg, or 35 to 50 mg, more preferably the tenapanor content in the tablet is 1, 3, 5, 10, 15, 20, 30, 35, or 50 mg. The composition may be destined for administration once or twice a day.

A method of preparation of the crystalline form of tenapanor free base comprises the following steps:

- a) dissolving and/or suspending tenapanor free base in a solvent; and
- b) crystallization of tenapanor from the solvent;
- c) removal of the solvent.

A particularly suitable solvent for use in the preparation of the tenapanor crystalline form is an organic solvent selected from polar organic solvents having 1 to 6 carbon atoms, mixtures thereof, and mixtures of said polar organic solvents with water. Preferably, the polar organic solvents are selected from C1-C6 alcohols, C3-C6 ketones, C2-C6 nitriles, C2-C6 amides, C2-C6 esters, C2-C6 ethers, C1-C6 sulfoxides and C1-C6 sulfones, optionally substituted by at least one halogen. Examples of suitable alcohols include methanol, ethanol, propane-1-ol, propane-2-ol or trifluoroethanol. Examples of suitable ketones include acetone, butanone and 4-methylpentane-2-one. Examples of suitable nitriles include acetonitrile. Examples of suitable amides include dimethyl formamide and dimethyl acetamide. Examples of suitable ethers include 1,2-dimethoxyethane. Examples of suitable sulfoxides include dimethyl sulphoxide. Examples of suitable sulfones include sulfolane.

Dissolving and/or suspending (dispersion) of tenapanor free base in the solvent may be carried out at a temperature range from 20 °C to the boiling point of the solvent.

Crystallization of the tenapanor free base may be carried out using known crystallization techniques, such as cooling the solution or suspension in the solvent or inoculation. Crystallization by cooling the

solution or suspension in the solvent is preferably carried out by cooling the mixture to the temperature within the range from 0 °C to 30 °C, preferably from 20 °C to 25 °C, and allowing to crystallize.

The solvent may be removed from the thus obtained crystalline form of tenapanor by filtration or centrifugation. Alternatively, the mixture containing the crystals of tenapanor may be lyophilized, spray dried or evaporated. Evaporation may be performed as free evaporation or as evaporation at an elevated temperature, e.g. at a temperature from 40 to 80 °C, and/or under a reduced pressure, e.g., about 10 kPa (100 mbar) or lower.

The preparation of the novel crystalline form of tenapanor may advantageously be used to increase the chemical purity of tenapanor. This form crystallizes well and the crystallization removes impurities from tenapanor. In particular, the novel crystalline form of tenapanor may be used for the preparation of tenapanor with a chemical purity (as determined by HPLC) greater than 96.5%, preferably greater than 97%, or greater than 97.5%, or greater than 98%, or greater than 98.5% , or greater than 98.8%. Most preferably a purity of greater than 99% or greater than 99.5% can be achieved.

The present invention further provides tenapanor salts selected from the group consisting of salt of tenapanor with phosphoric acid, salt of tenapanor with hydrobromic acid, and salt of tenapanor with citric acid. A particularly preferred tenapanor salt is salt of tenapanor with phosphoric acid which shows the highest stability.

The said salts are amorphous. Amorphous forms typically have a very good bioavailability and thus are particularly suitable for use in pharmaceutical forms.

The said tenapanor salts precipitate from the solution in a solid stable form. The impurities can be easily removed by filtering off the liquid phase, further purification can be performed by washing the solid phase.

The tenapanor salts can be prepared by a method comprising a step of dissolving tenapanor to form a tenapanor solution, and a step of adding an acid into the said tenapanor solution. The acid can be added in solid form or in the form of a solution of the acid.

The acid is selected from the group consisting of phosphoric acid, hydrobromic acid, citric acid.

Suitable solvents for dissolving tenapanor and optionally the acid are organic solvents, in particular ketones and ethers, most preferably acetone, tetrahydrofuran.

The reactions of tenapanor with a number of acids were studied, and it was found that tenapanor forms amorphous stable solid salts with phosphoric acid, hydrobromic acid and citric acid. Other tested pharmaceutically acceptable acids did not form salts with tenapanor which would be obtained from the reaction mixture in stable solid form.

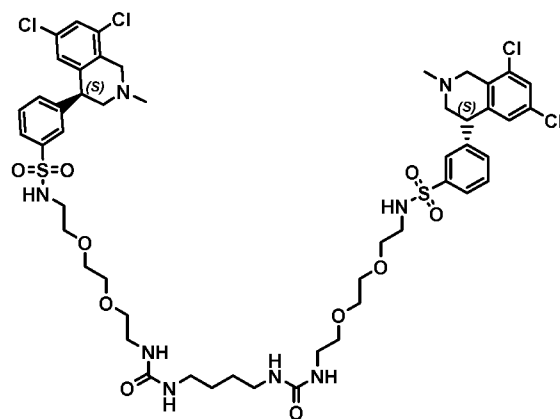
The novel salts of tenapanor according to the present invention can be used in the preparation of pharmaceutical compositions, in particular solid, semi-solid and liquid dosage forms, in particular solid dosage forms, e.g. tablets. Such pharmaceutical compositions may contain at least one excipient selected from the group consisting of fillers (e.g., lactose), binders (e.g., microcrystalline cellulose), disintegrants (e.g., croscarmellose sodium), lubricants (e.g., magnesium stearate), surfactants. Furthermore, the pharmaceutical compositions may optionally comprise at least one other excipient selected from the group consisting of colorants, solvents, antimicrobial agents, flavorings and olfactory adjusting agents. Tablets may be coated with conventional coatings containing, for example, polyvinyl alcohol or polyethylene glycol.

The pharmaceutical compositions can be prepared in virtually any solid dosage form, e.g. in the form of tablets, capsules, powders, pellets or granules. A preferred dosage form is a tablet or a coated tablet. The tablet or coated tablet may preferably be prepared by mixing tenapanor salt with at least one pharmaceutically acceptable excipient and subjecting the mixture to tableting, optionally followed by coating.

The content of the active ingredient in the tablet may range from 1 to 100 mg, preferably the tenapanor content in the tablet is within the range of 1 to 50 mg or 10 to 50 mg or 15 to 50 mg or 20 to 50 mg or 30 to 50 mg, or 35 to 50 mg, more preferably the tenapanor content in the tablet is 1, 3, 5, 10, 15, 20, 30, 35, or 50 mg. The composition may be destined for administration once or twice a day.

The present invention further provides an effective method of preparation of tenapanor of formula I. In contrast to the method known in the art, the herein described method has a high yield, a lower number of synthetic steps, a limited number of undesirable side reactions, and an easy isolation and purification of the intermediates and of the final product, tenapanor. These advantages are achieved mainly due to the fact that all synthetic steps yield crystalline compounds. The method of the invention is particularly advantageous with regard to the efficiency of using the expensive chirally pure 4-(3-bromophenyl)-6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinoline. Another advantage is that the process yields tenapanor in crystalline form, for example in crystalline form I.

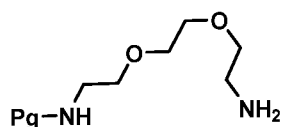
Object of the present invention is a method of preparation of tenapanor of formula I



(I),

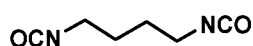
comprising the following steps of:

a) reaction of diamine of formula VIII



(VIII),

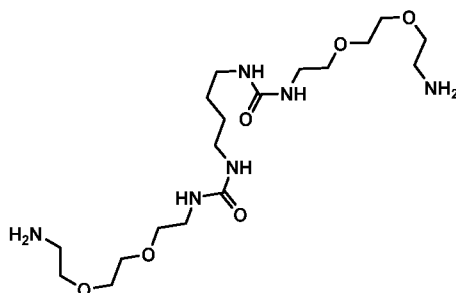
wherein Pg is a protecting group, preferably selected from t-butyloxycarbonyl (BOC), benzyloxycarbonyl (CBZ), acetyl and trifluoroacetyl, with 1,4-diisocyanatobutane of formula V



(V),

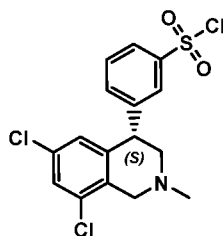
removing the protecting group from the product,

and subsequent reaction of the resulting 1,1'-(butan-1,4-diyl)bis(3-(2-(2-(2-aminoethoxy)ethoxy)ethyl)urea) of formula VII or a salt thereof



(VII)

with (S)-3-(6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinolin-4-yl)benzenesulfonyl chloride of formula III or salts thereof,

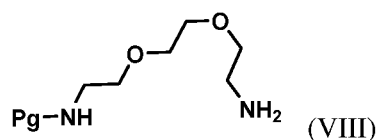


(III)

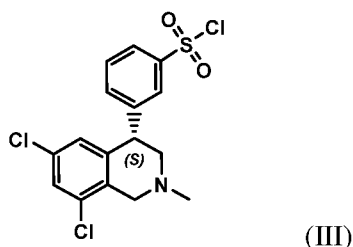
to form tenapanor of formula I,

or

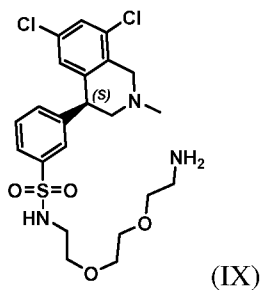
b) reaction of diamine of formula VIII



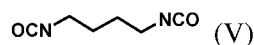
wherein Pg is a protecting group, preferably selected from *t*-butoxycarbonyl (BOC), benzyloxycarbonyl (CBZ), acetyl and trifluoroacetyl, with (*S*)-3-(6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinolin-4-yl)benzenesulfonyl chloride of formula III or a salt thereof,



removing the protecting group from the product, and subsequent reaction of the resulting (*S*)-*N*-(2-(2-(2-aminoethoxy)ethoxy)ethyl)-3-(6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinolin-4-yl)benzenesulfonamide of formula IX or a salt thereof



with 1,4-diisocyanatobutane of formula V



to form tenapanor of formula I.

The common feature of both reaction sequences a) and b) is the use of mono-protected diamine of formula VIII as a starting compound. Preferably, the protected diamine of formula VIII is commercially available *t*-butyl 2-(2-(2-aminoethoxy)ethoxy)ethylcarbamate (Pg = *t*-butoxycarbonyl).

Preferred is the reaction sequence a), because it allows the expensive optically pure starting compound (*S*)-3-(6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinolin-4-yl)benzenesulfonyl chloride to enter the reaction in a later stage of the reaction sequence.

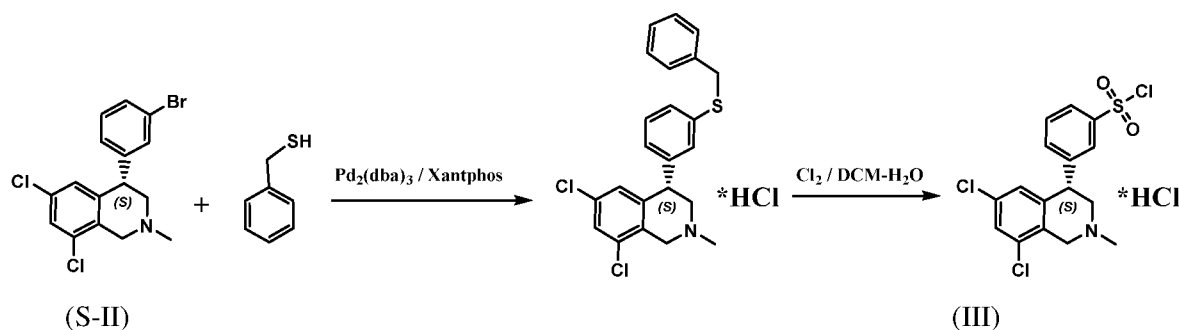
All reactions are preferably carried out in the presence of a base. The base may preferably be selected from alkali metal carbonate or tri(C1-C4)alkylamine wherein the alkyls are the same or different. Particularly preferred bases are triethylamine, N,N-diisopropylethylamine, K_2CO_3 , Na_2CO_3 .

The reactions are preferably performed in dry (anhydrous) polar aprotic solvent. The solvent is more preferably selected from the group comprising dimethylsulfoxide, dimethylformamide, dimethylacetamide, acetonitrile, N-methylpyrrolidone, dichloromethane. Most preferably, the solvent is N-methylpyrrolidone or dichloromethane.

The salts of intermediates are preferably salts with inorganic or organic acids, more preferably salts with hydrogen halides, most preferably hydrochlorides.

Tenapanor, the final product, is preferably re-crystalized from the system acetone/methanol.

In a preferred embodiment, (*S*)-3-(6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinolin-4-yl)benzene-1-sulfonyl chloride of formula III, optionally in the form of salt, particularly hydrochloride, is prepared by C-S coupling of (*S*)-4-(3-bromophenyl)-6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinoline of formula II with benzylmercaptane and subsequent oxidation by chlorine.



The step of C-S coupling is preferably carried out in dioxane with catalysis by palladium salt or complex in the presence of a base. The step of oxidation by chlorine is preferably performed in the system dichloromethane-water.

Chlorine for oxidation can be introduced into the reaction mixture in a gaseous state or in the form of a chlorinating agent such as NCS, cyanuric chloride, PCl_5 , $POCl_3$, sodium hypochlorite, calcium hypochlorite.

Brief Description of Drawings

Fig. 1: X-ray powder diffraction spectrum (XRPD spectrum) of the crystalline form I.

Fig. 2: X-ray powder diffraction spectrum (XRPD spectrum) of the salt of tenapanor with phosphoric acid.

Fig. 3: X-ray powder diffraction spectrum (XRPD spectrum) of the salt of tenapanor with HBr.

List of abbreviations

AcOH	acetic acid
DBTA	dibenzoyl tartaric acid
DCM	dichloromethane
DIPEA	<i>N,N</i> -diisopropylethylamine
DMF	dimethylformamide
DMS	dimethylsulfoxide
EtOH	ethanol
NCS	<i>N</i> -chlorosuccinimide
NMP	1-methyl-2-pyrrolidone
Pd ₂ (dba) ₃	tris(dibenzylideneacetone)dipalladium(0)
TEA	triethylamine

Examples of Carrying Out the Invention

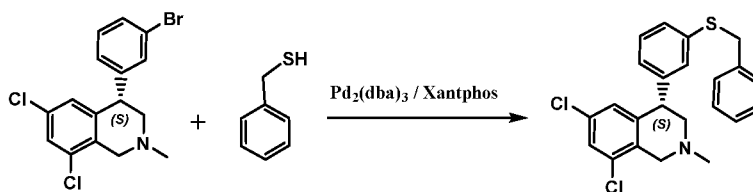
The following exemplary embodiments serve only to illustrate and explain the invention and are by no means intended to limit the scope of protection which is defined solely by the teachings of the claims.

Comparative Example

Tenapanor free base in the form of an amorphous solid foam was prepared by the procedure disclosed in patent application WO 2010/078449, Example 202. The chemical purity of the tenapanor prepared by this procedure was 96.5% (HPLC). The structure of tenapanor was verified by MS and ¹H and ¹³C NMR spectra.

Step A

Preparation of (*S*)-4-(3-(benzylthio)phenyl)-6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinoline

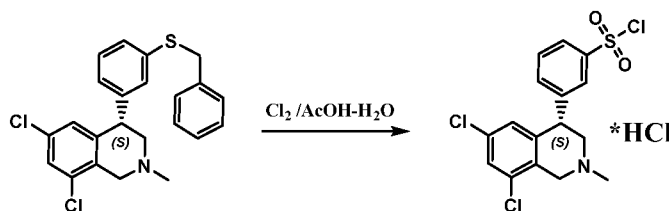


Potassium carbonate (9.30 g) and anhydrous xylene (500 ml) were added to the reaction vessel. Benzyl mercaptane (25 g) was added dropwise to the stirred mixture under ice-cooling. The resulting mixture was stirred at 25 °C for 1h.

(S)-4-(3-bromophenyl)-6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinoline 50 g in anhydrous xylene (500 ml), $\text{Pd}_2(\text{dba})_3$ (3 g) and Xantphos (3 g). The resulting solution was stirred at 25 °C for 30 minutes and then added to a solution of benzyl mercaptane. The resulting reaction mixture was maintained at 140 °C for 16 h. The mixture was then concentrated and the residue was subjected to preparative chromatography on silica gel with the mobile phase ethyl acetate / petroleum ether (1:100-1:50). 20 g of product are obtained as a yellow oil (36% yield).

Step B

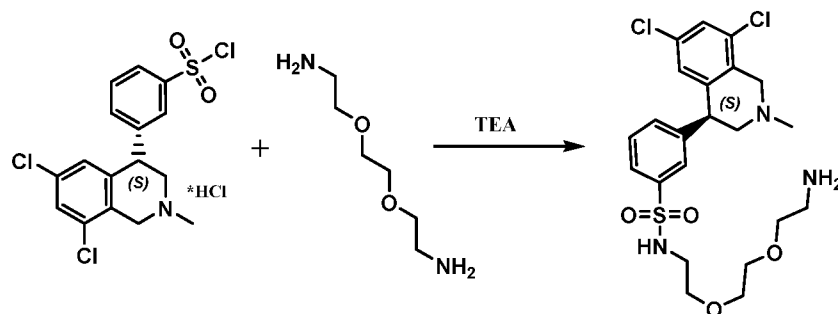
Preparation of (S)-3-(6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinolin-4-yl)benzenesulfonyl chloride hydrochloride



(S)-4-(3-(benzylthio)phenyl)-6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinoline (16 g) was dissolved in the reaction vessel in acetic acid/water (160 mL:16 mL) mixture. The mixture was cooled in an ice bath and then gaseous Cl_2 was introduced into the well stirred mixture. After disappearance of the starting material, the reaction mixture was purged with nitrogen and concentrated in vacuo. A product (10 g, 66.6%) was obtained as a colorless substance.

Step C

Preparation of (S)-N-(2-(2-(2-aminoethoxy)ethoxy)ethyl)-3-(6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinolin-4-yl)benzenesulfonamide



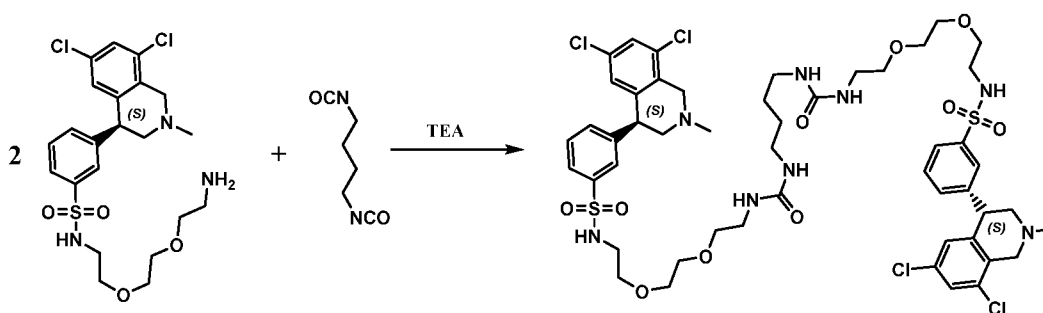
2-(2-(2-Aminoethoxy)ethoxy)ethylamine HCl (30 g; 0.2 mol) and triethylamine (5.2 g; 52 mmol) were dissolved in dichloromethane (500 ml) and the mixture was chilled in an ice bath. (S)-3-(6,8-Dichloro-2-methyl-1,2,3,4-tetrahydroisoquinolin-4-yl)benzenesulfonyl chloride hydrochloride (10 g; 26 mmol) was added in parts during 40 minutes to the chilled reaction mixture. The ice bath was removed and the reaction mixture was stirred at laboratory temperature for additional 30 minutes.

The dichloromethane solution was extracted three times by brine (2x 250 ml), dried over sodium sulphate, and concentrated in vacuo. The residue was purified using preparative chromatography on silica gel with dichloromethane-methanol mobile phase.

Yield 7.2 g. HRMS 502.1247 $[M+H]^+$, $C_{22}H_{29}Cl_2N_3O_4S$.

Step D

Preparation of 17-[[[3-[(4S)-6,8-dichloro-1,2,3,4-tetrahydro-2-methyl-4-isoquinolinyl]phenyl]sulphonyl]amino]-N-[2-[2-[2-[[[3-[(4S)-6,8-dichloro-1,2,3,4-tetrahydro-2-methyl-4-isoquinolinyl]phenyl]sulphonyl]amino]ethoxy]ethoxy]ethyl]-8-oxo-12,15-dioxo-2,7,9-triazaheptadecaneamide (tenapanor free base)



(S)-N-(2-(2-(2-aminoethoxy)ethoxy)ethyl)-3-(6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinolin-4-yl)benzenesulfonamide (5g; 10 mmol) prepared in step A was dissolved in dichloromethane (50 ml). Triethylamine (1.5 g; 14.9 mmol) and 1,4-diisocyanatobutane (0.48 g; 3.4 mmol) were added to the solution. The reaction mixture was cooled using ice and stirred overnight. The resulting fine suspension was filtered off, the filtrate was concentrated and the obtained product was purified by preparative chromatography on silica gel with dichloromethane-methanol mixture as a mobile phase.

Yield: 2 g of tenapanor in the form of amorphous solid foam. HPLC purity 96.5 %.

HRMS 1143.3186 $[M+H]^+$, $C_{50}H_{66}Cl_4N_8O_{10}S_2$. 1H NMR (500MHz, DMSO, ppm): 7.69-7.66 (m, 6H), 7.54-7.50 (m, 6H), 6.89 (bs, 2H), 5.9 (t, 2H), 5.79 (t, 2H), 4.4 (dd, 2H), 3.7 (dd, 4H), 3.44-3.44 (m, 8H), 3.35 (dd, 8H), 3.12 (dd, 4H), 2.96-2.64 (m, 12H), 2.37 (s, 6H), 1.31 (bs, 4H).

Step E

Preparation of bishydrochloride salt of tenapanor

Tenapanor free base (1 g; 0.85 mmol) prepared in step B was dissolved in a mixture of methanol (10 ml) and 4M aqueous HCl (0.5 ml; 2 mmol) under mild reflux. The solution was concentrated on rotary vacuum evaporator, and the title product was obtained in the yield of 1 g of amorphous solid foam.

Example 1

Preparation of tenapanor, crystalline form I

Tenapanor free base (200 mg, 0.17 mmol), prepared as in step D of the comparative example, was dissolved in 0.4 ml acetonitrile under mild reflux. The clear solution was cooled at the rate of 1 °C/min with stirring to laboratory temperature (i.e., range from 22 °C to 26 °C) and then stirred for additional 2 hours at this temperature. The resulting crystals were isolated by filtration on sintered glass filter and dried for 6 hours in a vacuum oven at 40 °C. Crystallization yield was 170 mg of crystalline form I of tenapanor. HPLC showed a purity of 99.5%.

X-ray powder diffraction spectrum of the crystalline form I is shown on Fig. 1. A complete list of reflections (XRPD peaks) having a relative intensity above 14% is shown in Table 1.

Table 1: Diffraction peaks of the crystalline form of tenapanor

Pos. [°2θ]	d [Å]	Rel. Int. [%]
11.07	7.986	84.9
11.70	7.555	18.9
13.16	6.721	42.9
13.45	6.579	36.1
14.95	5.920	14.9
15.47	5.724	17.3
15.90	5.571	12.3
17.16	5.164	14.1
19.38	4.577	75.0
19.80	4.481	75.5
20.77	4.274	45.9
22.08	4.022	63.8
22.67	3.920	100.0
23.03	3.859	28.9
23.28	3.817	36.0
23.71	3.749	32.5
24.40	3.646	42.7
24.85	3.580	25.5

DSC analysis showed the melting temperature of tenapanor crystalline form I 130.5 ± 2 °C.

Testing of the crystalline form stability: Tenapanor base crystalline form I was suspended in a solvent in such a ratio that the amount of tenapanor significantly exceeds its solubility in the said solvent under laboratory conditions. The suspension obtained was vigorously stirred for 10 days at laboratory temperature. The solvents used were 4-methylpentan-2-one; cyclopentyl methyl ether; ethyl acetate; toluene; dichloromethane; acetone; water or a mixture of propan-2-ol: water (9: 1).

In any of the mentioned solvents, the crystal form did not change under the given conditions and no new crystalline form appeared in detectable amounts.

Example 2

Alternative method of preparation of crystalline form I

Tenapanor free base (200 mg, 0.17 mmol), prepared as in step D of the comparative example, was suspended in 0.5 ml of acetone. The suspension was stirred under mild reflux for 30 minutes. The resulting suspension was cooled to laboratory temperature with stirring and then stirred for additional 2 hours. The resulting crystals were filtered off and dried for 6 hours in a vacuum oven at 40 °C. Crystallization yield was 100 mg of crystalline form I of tenapanor. HPLC showed a purity of 98.9%. RTG and DSC analysis confirmed that the obtained form I was identical to the form I obtained in Example 1.

Example 3

Pharmaceutical composition

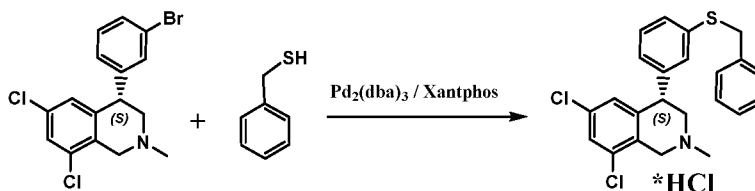
<u>Substance</u>	<u>Amount – core /mg/</u>
Tenapanor, form I	50.0
Lactose Monohydrate	160.0
Microcrystalline Cellulose	50
Sodium Crosscarmellose	37.3
Magnesium Stearate	2.7

The individual substances were added into a homogenizer: tenapanor (crystalline form I), lactose, microcrystalline cellulose and croscarmellose sodium. The mixture was homogenized for 15 minutes at 20 rpm. Finally, magnesium stearate was added and the mixture was homogenized for another 3 minutes at 20 rpm. The tableting mixture was compressed on a rotary tableting machine to make approximately 300-mg-tablet cores.

Examples 4 to 9 illustrate the inventive method of preparation of crystalline tenapanor.

Example 4

Preparation of (S)-4-(3-(benzylthio)phenyl)-6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinoline



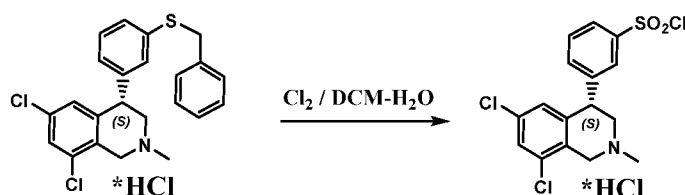
DIPEA (9.6 mL) and anhydrous dioxane (100 mL) were added to a reaction vessel. Benzyl mercaptan (8.1 mL) was added dropwise to the stirred mixture under ice-cooling. The resulting mixture was stirred at 25 °C for 1h.

In a second reaction vessel, (S)-4-(3-bromophenyl)-6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinoline (21.2 g) in anhydrous dioxane (140 mL), Pd₂(dba)₃ (835 mg) and Xantphos (835 mg) were mixed. The resulting solution was stirred at 25 °C for 30 minutes and then added to the solution of benzyl mercaptan. The resulting reaction mixture was maintained at gentle reflux for 3 hours.

After cooling, the suspension obtained was filtered through a thin layer of celite. HCl was added to the filtrate. The precipitated hydrochloride was isolated by filtration, washed well and dried. 21 g of pinkish product were obtained (81.6% yield).

Example 5

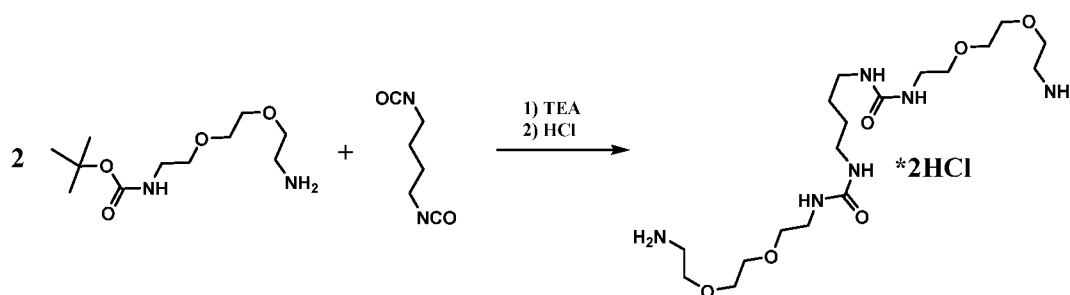
Preparation of (S)-3-(6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinolin-4-yl)benzenesulfonyl chloride hydrochloride



(S)-4-(3-(benzylthio)phenyl)-6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinoline hydrochloride (11.1 g) was stirred in DCM/2M HCl (70 mL:6 mL) mixture in a reaction vessel. The mixture was cooled in an ice bath and then gaseous Cl₂ was introduced into the vigorously stirred mixture. After disappearance of the starting material, the resulting suspension was bubbled through by nitrogen and the product was filtered off and washed with DCM. 9.2 g of white product was obtained (82.7% yield).

Example 6

Preparation of 1,1'-(butan-1,4-diyl)bis(3-(2-(2-(2-aminoethoxy)ethoxy)ethyl)urea)*2HCl



In the reaction vessel, t-butyl 2-(2-(2-aminoethoxy)ethoxy)ethylcarbamate (21.8 g) was stirred in DCM. The mixture was cooled in an ice bath under an inert atmosphere. To the cooled solution was

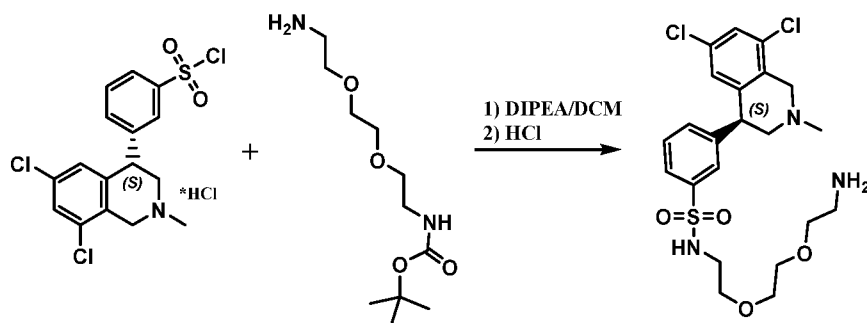
added 1,4-diisocyanatobutane (6.14 g) and TEA (0.1 mL). The cooling bath was removed and the reaction mixture was further stirred for 2 h.

35% HCl was added to the reaction mixture and the mixture was stirred under gentle reflux overnight. After cooling, the precipitated product was filtered off and washed with DCM.

The product was recrystallized from propan-2-ol. 22.3 g of white product was obtained (80% yield).

Example 7

Preparation of (S)-N-(2-(2-(2-aminoethoxy)ethoxy)ethyl)-3-(6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinolin-4-yl)benzenesulfonamide



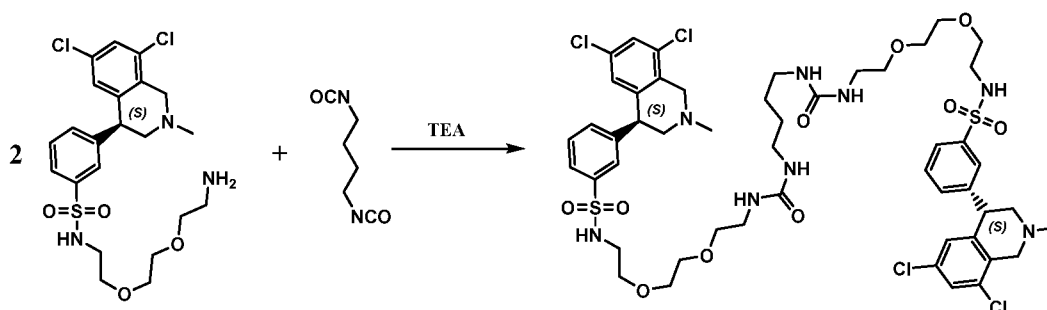
(S)-3-(6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinolin-4-yl)benzenesulfonyl chloride hydrochloride (11.7 g) prepared in Example 2 was stirred in dichloromethane (100 ml) and the suspension was cooled in an ice bath. To the cooled suspension was added a solution of t-butyl 2-(2-(2-aminoethoxy)ethoxy)ethylcarbamate (6.8 g) and DIPEA (14 ml) in DCM (50 ml). The resulting solution was stirred for 2 hours in an ice bath. The reaction mixture was extracted twice with water. Concentrated HCl (15 mL) was added to the dichloromethane solution and the mixture heated at gentle reflux for 2 h.

The precipitated product, after cooling, was extracted into water. The aqueous phase was separated and basified with Na_2CO_3 . The product as the free base was extracted into DCM and the dichloromethane solution was dried over sodium sulfate and concentrated in vacuo. 12.9 g of product were obtained.

Yield 93.4%. HRMS 502.1247 $[\text{M}+\text{H}]^+$, $\text{C}_{22}\text{H}_{29}\text{Cl}_2\text{N}_3\text{O}_4\text{S}$.

Example 8

Preparation of 17-[[[3-[(4S)-6,8-dichloro-1,2,3,4-tetrahydro-2-methyl-4-isoquinolinyl]phenyl]sulfonyl]amino]-N-[2-[2-[2-[[[3-[(4S)-6,8-dichloro-1,2,3,4-tetrahydro-2-methyl-4-isoquinolinyl]phenyl]sulfonyl]amino]ethoxy]ethoxy]ethyl]-8-oxo-12,15-dioxo-2,7,9-triazaheptadecanamide (tenapanor free base)

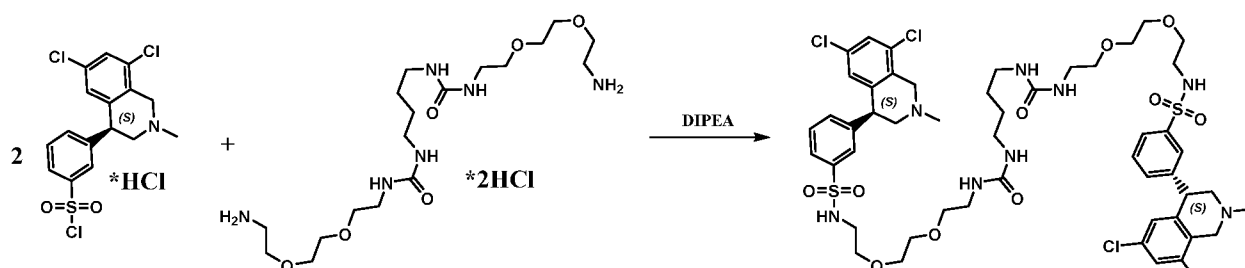


(S)-N-(2-(2-(2-aminoethoxy)ethoxy)ethyl)-3-(6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinolin-4-yl)benzenesulfonamide (12.9 g) prepared in Example 4 was dissolved in dichloromethane (150 ml). To the solution was added triethylamine (0.3 ml) and 1,4-diisocyanatobutane (1.7 g). The reaction mixture was stirred at 25 °C for 2 h. The resulting reaction mixture was extracted with water and aqueous Na₂CO₃. The dichloromethane solution of the product was dried over sodium sulfate and concentrated to a solid foam. Yield 13.9 g. The crude product was taken up in acetone (100 ml) and then recrystallized from methanol (80 ml). 7.3 g of white crystalline product was obtained. Yield 49.8%.

HRMS 1143.3186 [M+H]⁺, C₅₀H₆₆Cl₄N₈O₁₀S₂. ¹H NMR (500MHz, DMSO, ppm): 7.69-7.66 (m, 6H), 7.54-7.50 (m, 6H), 6.89 (bs, 2H), 5.9 (t, 2H), 5.79 (t, 2H), 4.4 (dd, 2H), 3.7 (dd, 4H), 3.44-3.44 (m, 8H), 3.35 (dd, 8H), 3.12 (dd, 4H), 2.96-2.64 (m, 12H), 2.37 (s, 6H), 1.31 (bs, 4H)

Example 9

Preparation of 17-[[[3-[(4S)-6,8-dichloro-1,2,3,4-tetrahydro-2-methyl-4-isoquinolinyl]phenyl]sulfonyl]amino]-N-[2-[2-[2-[[[3-[(4S)-6,8-dichloro-1,2,3,4-tetrahydro-2-methyl-4-isoquinolinyl]phenyl]sulfonyl]amino]ethoxy]ethoxy]ethyl]-8-oxo-12,15-dioxo-2,7,9-triazaheptadecanamide (tenapanor free base)



(S)-3-(6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinolin-4-yl)benzenesulfonyl chloride hydrochloride (0.81 g) prepared in Example 2 and 1,1'-(butane-1,4-diyl)bis(3-(2-(2-(2-aminoethoxy)ethoxy)ethyl)urea) dihydrochloride prepared according to Example 3 (0.48 g) were stirred in anhydrous NMP (10 ml). To the suspension was added DIPEA (2 mL) and the resulting solution was stirred at 60 °C for 1.5 h. Water (10 mL) was added dropwise to the reaction mixture and the mixture was cooled to 5 °C. The precipitated product was isolated and stirred in acetone at 5 °C overnight. The beige product was filtered off (0.67 g) and recrystallized from methanol (12 ml). 0.53 g of a colorless crystalline product was obtained.

Yield 78.7 %. HRMS 502.1247 $[M+H]^+$, $C_{22}H_{29}Cl_2N_3O_4S$. DSC analysis showed the melting temperature of 130.5 °C.

Example 10

Tenapanor (1.48 g, 1.3 mmol) is dissolved in 10 ml of tetrahydrofuran (THF). From the thus prepared solution, 1 ml is taken and phosphoric acid (0.4 mmol) is added. The mixture is stirred at room temperature for 24 hours. Salt of tenapanor with phosphoric acid precipitated from the solution in solid stable form, the salt was filtered off, washed with THF and dried by stream of inert gas. XRPD confirmed amorphousness of the product.

Example 11

Tenapanor (1.48 g, 1.3 mmol) is dissolved in 10 ml of tetrahydrofuran (THF). From the thus prepared solution, 1 ml is taken and hydrobromic acid (0.4 mmol) is added. The mixture is stirred at room temperature for 24 hours. Salt of tenapanor with hydrobromic acid precipitated from the solution in solid stable form, the salt was filtered off, washed with THF and dried by stream of inert gas. XRPD confirmed amorphousness of the product.

Example 12

Tenapanor (1.48 g, 1.3 mmol) is dissolved in 10 ml of acetone. From the thus prepared solution, 1 ml is taken and phosphoric acid (0.4 mmol) is added. The mixture is stirred at room temperature for 24 hours. Salt of tenapanor with phosphoric acid precipitated from the solution in solid stable form, the salt was filtered off, washed with acetone and dried by stream of inert gas. XRPD confirmed amorphousness of the product.

Example 13

Tenapanor (1.48 g, 1.3 mmol) is dissolved in 10 ml of acetone. From the thus prepared solution, 1 ml is taken and citric acid (0.4 mmol) is added. The mixture is stirred at room temperature for 24 hours. Salt of tenapanor with citric acid precipitated from the solution in solid stable form, the salt was filtered off, washed with acetone and dried by stream of inert gas. XRPD confirmed amorphousness of the product.

Other pharmaceutically acceptable acids were tested by the procedures shown in Examples 10-13, but did not yield salts which would precipitate in amorphous stable solid form from the solution. The tested acids were: methanesulfonic acid, benzenesulfonic acid, oxalic acid, maleinic acid, tartaric acid, fumaric acid, trichloroacetic acid.

Example 14

Tenapanor (500 mg, 0.44 mmol) is dissolved in 20 ml of THF at 45 °C. To this clear solution, a solution of phosphoric acid in THF (50 µl/5 ml) is added dropwise during 10 minutes. The resulting suspension is stirred at room temperature for 30 minutes. The precipitated salt of tenapanor with phosphoric acid (79 %) is filtered off, washed with 3 ml of THF and dried by stream of inert gas. Yield: 430 mg of colourless salt of tenapanor with phosphoric acid. XRPD showed amorphousness of the product.

Example 15

Tenapanor (500 mg, 0.44 mmol) is dissolved in 20 ml of THF at 45 °C. To this clear solution, hydrobromic acid (48%; 100 µl) is added dropwise during 10 minutes. A fine precipitate forms already during the dropwise addition of HBr, and the suspension is stirred at room temperature for 30 minutes. The precipitated salt of tenapanor with HBr is filtered off, washed with 3 ml of THF and dried by stream of inert gas. Yield: 397 mg (69 %) of colourless salt of tenapanor with HBr (1:2). XRPD showed amorphousness of the product.

Analytical methods

XRPD measurement parameters: Diffractograms were measured on X'PERT PRO MPD PANalytical diffractometer, using CuK α irradiation ($\lambda = 1.542 \text{ \AA}$), excitation voltage: 45 kV, anode current: 40 mA, measuring range: 2 - 40 °2 θ , step size: 0.01 °2 θ , measured on a flat powder sample which was applied to a Si plate. For the primary optics, programmable divergence apertures with a 10 mm sample illuminated surface, Soller apertures of 0.02 rads, and an anti-dispersion screen ¼° were used. An X'Celerator detector with maximum opening of the detection slot, Soller's apertures of 0.02 rad, and a 5.0 mm anti-denture aperture were used to set the secondary optics.

Nuclear magnetic resonance (NMR) spectra were measured on a Bruker Avance 500 instrument. The ^1H spectra were measured at 500.13 MHz, ^{13}C at 125.8 MHz. The sample was measured in a deuterated solvent, typically at 25 °C (unless otherwise stated for a specific analysis). The chemical shift δ is expressed in ppm, the interaction constants J are given in Hz. The spectra were referenced to the solvent residual signal.

Differential scanning calorimetry (DSC) curves were measured on Perkin Elmer's DSC Pyris 1 instrument. Sample loading into a standard Al crucible (40 µL) was from 2 to 4 mg and the heating rate was 10 °C/min. The temperature program used is composed of 1 stabilization minute at 0 °C and then heating to 300 °C with a heating rate of 10 °C/min. 4.0 N $_2$ was used as a carrier gas at a flow rate of 20 ml/min.

Reaction course and product quality controls were performed using HPLC on an XSelect C18 column, 2.5 μm , 4.6x100 mm using a gradient method and a mobile phase of acetonitrile-buffer (10 mM $(\text{NH}_4)_2\text{HPO}_4$ - pH 7.8). Mobile phase flow 1.2 ml/min.

CLAIMS

1. A crystalline form of tenapanor free base.
2. The crystalline form of tenapanor free base according to claim 1, wherein the crystalline form exhibits characteristic reflections in the X-ray powder diffraction spectrum obtained using CuK α radiation: 11.1; 19.8 and $22.7 \pm 0.2^\circ$ 2-theta.
3. The crystalline form of tenapanor free base according to claim 2, wherein the crystalline form exhibits further characteristic reflections in the X-ray powder diffraction spectrum obtained using CuK α radiation: 13.2; 15.5; 19.4; 22.1 and $24.4 \pm 0.2^\circ$ 2-theta.
4. The crystalline form of tenapanor free base according to any one of claims 1 to 3, exhibits the differential scanning calorimetry curve with a melting point at the temperature $130.5 \pm 2^\circ\text{C}$.
5. Use of the crystalline form of tenapanor free base according to any one of claims 1 to 4 for the preparation of tenapanor with a chemical purity, as determined by HPLC, greater than 96.5%, preferably greater than 97%, or greater than 97.5%, or greater than 98%, or greater than 98.5%, or greater than 98.8%.
6. A method of preparation of the crystalline form of tenapanor free base according to any one of claims 1 to 4, **characterized in that** it comprises the following steps:
 - a) dissolving and/or suspending tenapanor free base in a solvent; and
 - b) crystallization of tenapanor free base from the solvent;
 - c) removal of the solvent.
7. The method according to claim 6, **characterized in that** the solvent is selected from polar organic solvents having 1 to 6 carbon atoms, mixtures thereof, and mixtures of said polar organic solvents with water; more preferably, the polar organic solvents are selected from C1-C6 alcohols, C3-C6 ketones, C2-C6 nitriles, C2-C6 amides, C2-C6 esters, C2-C6 ethers, C1-C6 sulfoxides, C1-C6 sulfones and mixtures thereof, wherein the alcohols, nitriles, amides, esters, ethers, sulfoxides or sulfones are optionally substituted by at least one halogen,
and/or
in that the step of crystallization of tenapanor free base from the solvent is performed by cooling the solution or suspension in the solvent to the temperature within the range from 0°C to 30°C , preferably from 20°C to 25°C , and allowing to crystallize,
and/or

in that the solvent is removed from the crystalline form of tenapanor free base by filtration, centrifugation, lyophilization, spray drying or solvent evaporation.

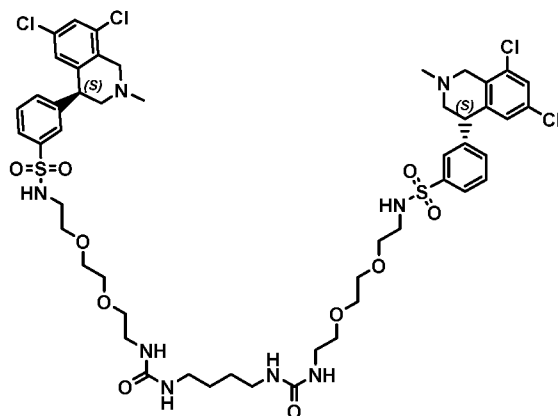
8. Tenapanor salt selected from the group consisting of salt of tenapanor with phosphoric acid, salt of tenapanor with hydrobromic acid, and salt of tenapanor with citric acid, preferably the tenapanor salt is salt of tenapanor with phosphoric acid.

9. A method of preparation of the tenapanor salt according to claim 8, **characterized in that it contains** a step of dissolving tenapanor to form a tenapanor solution, and a step of adding an acid selected from the group consisting of phosphoric acid, hydrobromic acid and citric acid to the said tenapanor solution.

10. A pharmaceutical composition containing tenapanor, **characterized in that** it contains the crystalline form of tenapanor free base according to any one of claims 1 to 4 and/or at least one tenapanor salt according to claim 8, and at least one pharmaceutically acceptable excipient, preferably selected from the group consisting of fillers, binders, disintegrants, lubricants, surfactants, more preferably selected from the group consisting of lactose, microcrystalline cellulose, sodium croscarmellose, magnesium stearate.

11. The crystalline form of tenapanor free base according to any one of claims 1 to 4 and/or the tenapanor salt according to claim 8 for use in the treatment of a condition selected from: irritable bowel syndrome, especially when accompanied by constipation; hyperphosphatemia, especially in patients with dialysis with final stage renal failure; chronic kidney disease; or for use in the prevention of excess sodium in patients with kidney and heart conditions.

12. A method of preparation of tenapanor of formula I

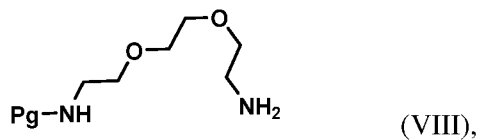


(I),

characterized in that it comprises the steps of:

a) - reaction of diamine of formula VIII

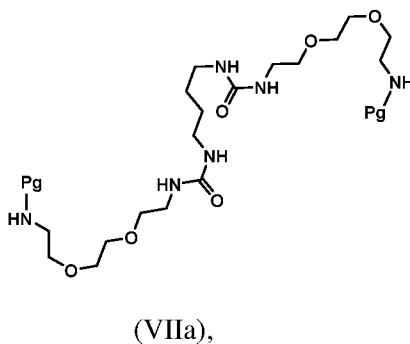
24



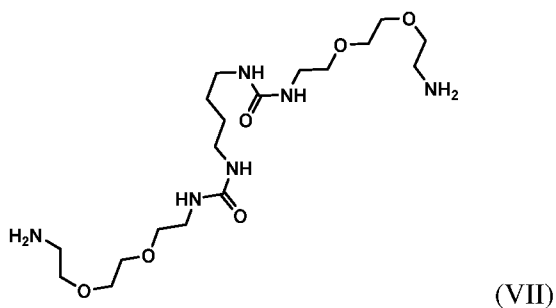
wherein Pg is a protecting group, preferably selected from t-butyloxycarbonyl (BOC), benzyloxycarbonyl (CBZ), acetyl and trifluoroacetyl, with 1,4-diisocyanatobutane of formula V



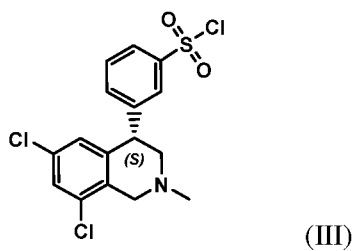
to form the protected diamine of formula VIIa



- removing the protecting groups from the diamine of formula VIIa,
- and subsequent reaction of the resulting 1,1'-(butan-1,4-diyl)bis(3-(2-(2-(2-aminoethoxy)ethoxy)ethyl)urea) of formula VII or a salt thereof



with (S)-3-(6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinolin-4-yl)benzenesulfonyl chloride of formula III or salts thereof,

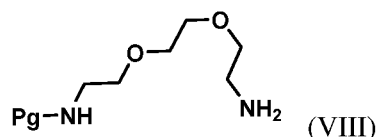


to form tenapanor of formula I,

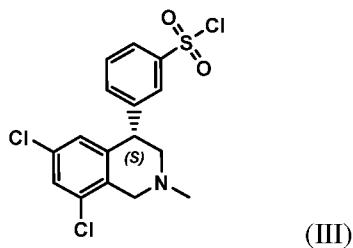
or

- b) - reaction of diamine of formula VIII

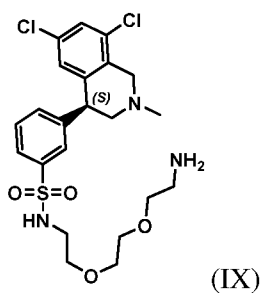
25



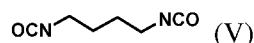
wherein Pg is a protecting group, preferably selected from t-butyloxycarbonyl (BOC), benzyloxycarbonyl (CBZ), acetyl and trifluoroacetyl, with (*S*)-3-(6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinolin-4-yl)benzenesulfonyl chloride of formula III or a salt thereof,



- removing the protecting group from the product,
- and subsequent reaction of the resulting (*S*)-*N*-(2-(2-(2-aminoethoxy)ethoxy)ethyl)-3-(6,8-dichloro-2-methyl-1,2,3,4-tetrahydroisoquinolin-4-yl)benzenesulfonamide of formula IX or a salt thereof



with 1,4-diisocyanatobutane of formula V

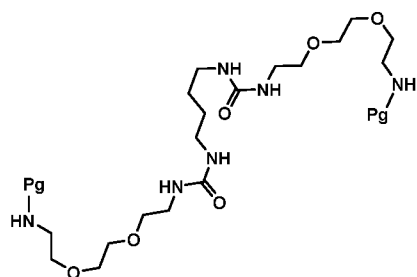


to form tenapanor of formula I.

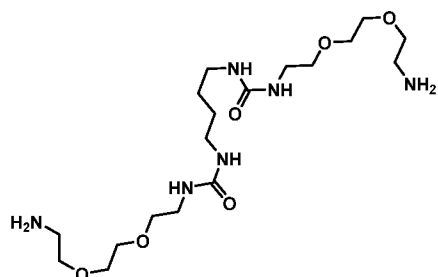
13. The method according to claim 12, wherein the salts of the intermediates are salts with inorganic or organic acids, preferably salts with hydrogen halides, most preferably hydrochlorides.

14. The method according to claim 12 or 13, wherein the final product tenapanor is recrystallized from acetone/methanol system.

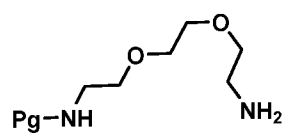
15. Use of an intermediate selected from the group consisting of:



(VIIa)



(VII)



(VIII),

and salts of these compounds,

wherein Pg is a protecting group, preferably selected from t-butyloxycarbonyl (BOC), benzyloxycarbonyl (CBZ), acetyl and trifluoroacetyl, for preparation of tenapanor.

1 / 2

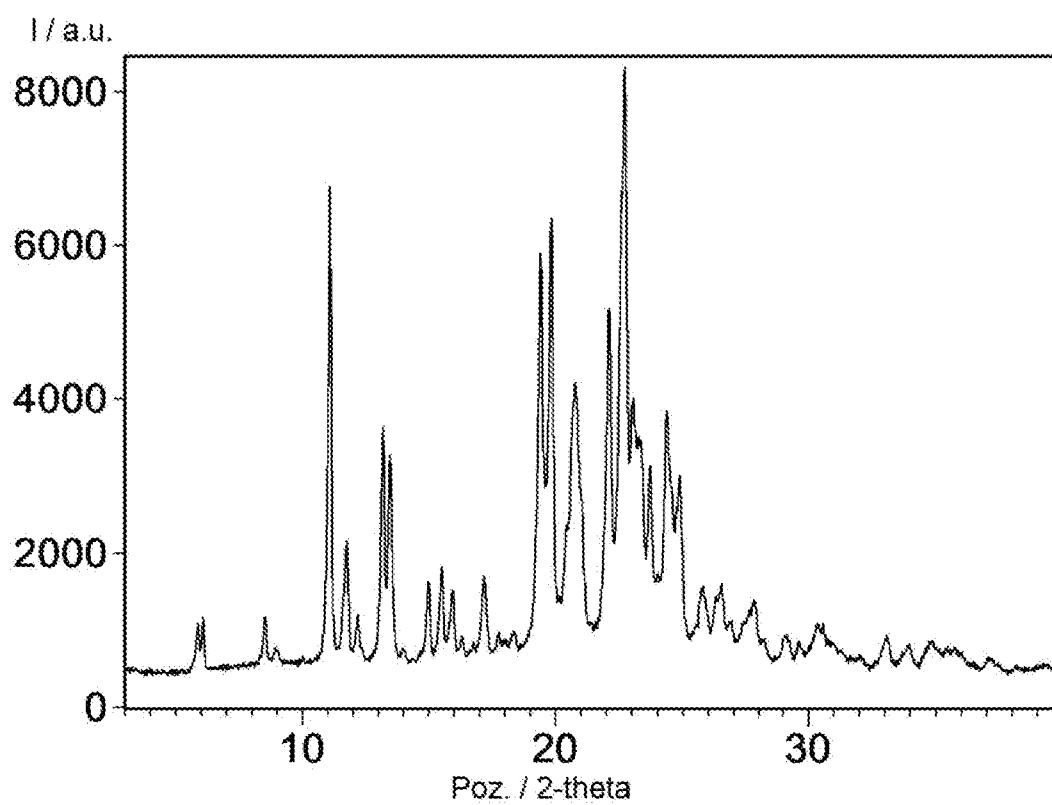


Fig. 1

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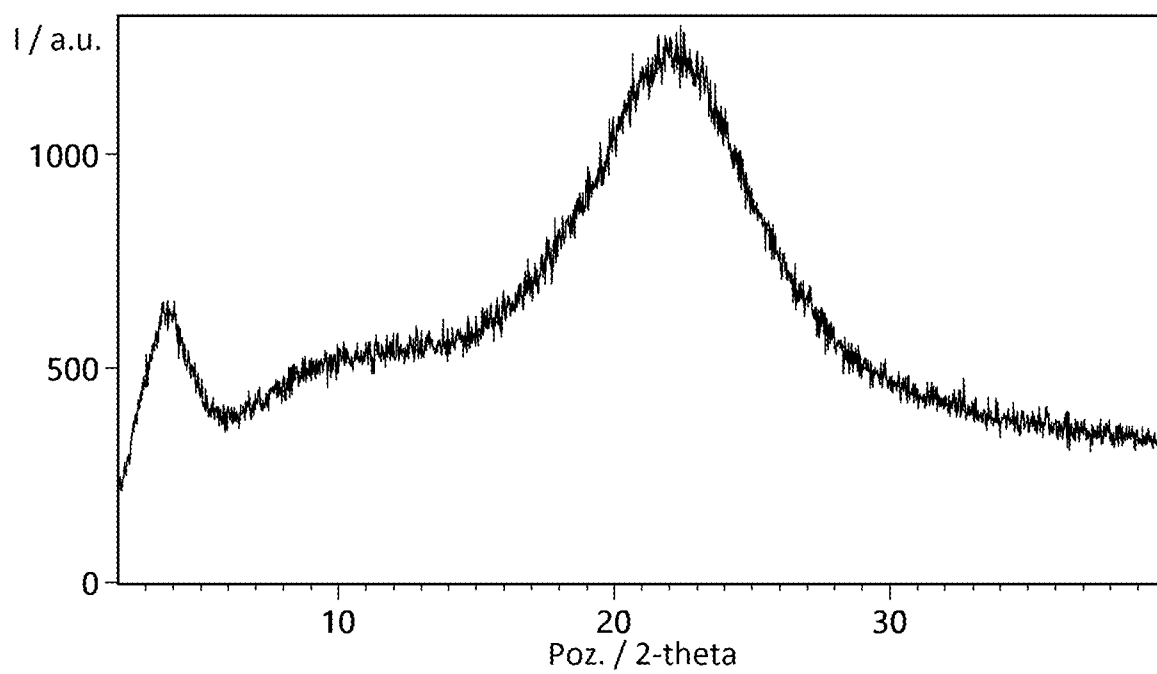


Fig. 2

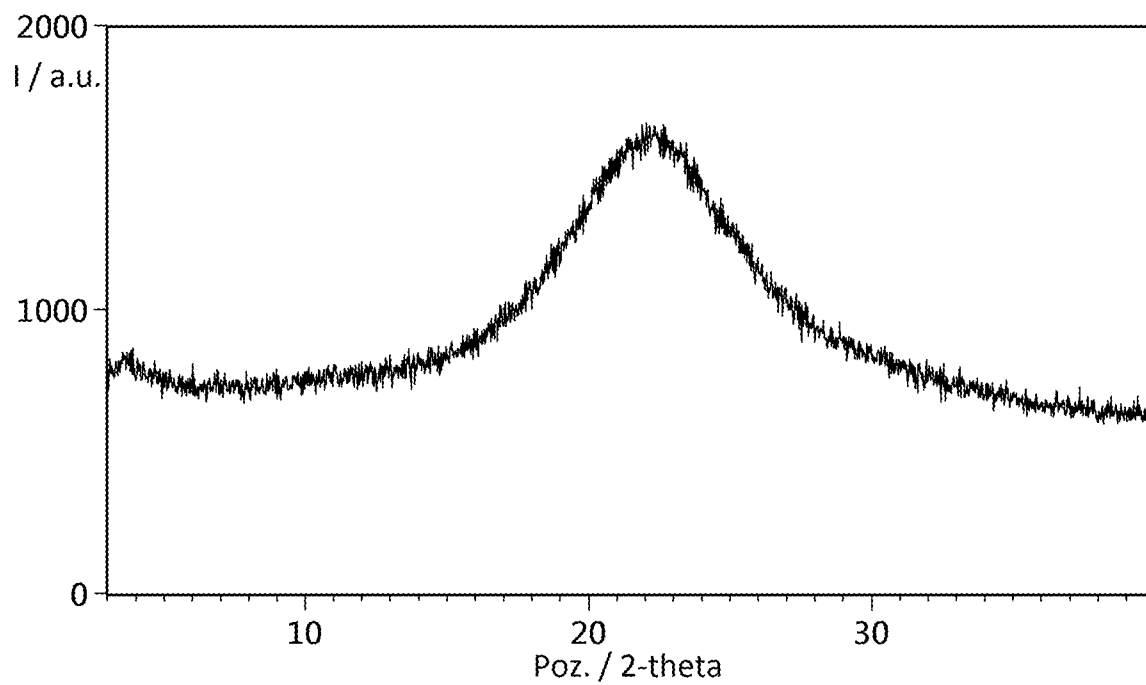


Fig. 3

INTERNATIONAL SEARCH REPORT

International application No
PCT/CZ2018/050055A. CLASSIFICATION OF SUBJECT MATTER
INV. C07D217/16 A61P1/04
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)
C07D A61P

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

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 2010/078449 A2 (ARDELYX INC [US]; CHARMOT DOMINIQUE [US]; JACOBS JEFFREY W [US]; LEADB) 8 July 2010 (2010-07-08) cited in the application example 202 claims 71-131 ----- -/--	1-7,10, 11



Further documents are listed in the continuation of Box C.



See patent family annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"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

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"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

9 January 2019

Date of mailing of the international search report

26/03/2019

Name and mailing address of the ISA/

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Authorized officer

Sarakinis, Georgios

INTERNATIONAL SEARCH REPORT

International application No

PCT/CZ2018/050055

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X,P	STEN O. NILSSON LILL ET AL: "Elucidating an Amorphous Form Stabilization Mechanism for Tenapanor Hydrochloride: Crystal Structure Analysis Using X-ray Diffraction, NMR Crystallography, and Molecular Modeling", MOLECULAR PHARMACEUTICS, vol. 15, no. 4, 2 April 2018 (2018-04-02), pages 1476-1487, XP055539475, US ISSN: 1543-8384, DOI: 10.1021/acs.molpharmaceut.7b01047 ANHY crystal form preparation, page 1479, left column, second paragraph & Sten O Nilsson Lill ET AL: "Elucidating an Amorphous Form Stabilization Mechanism for Tenapanor Hydrochloride: Crystal Structure Analysis Using X-ray Diffraction, NMR Crystallography, and Molecular Modeling", Molecular pharmaceuticals, 12 March 2018 (2018-03-12), pages 1476-1487, XP055539474, United States DOI: 10.1021/acs.molpharmaceut.7b01047 Retrieved from the Internet: URL:http://pubs.acs.org/doi/suppl/10.1021/acs.molpharmaceut.7b01047/suppl_file/mp7b01047_si_001.pdf Figure S28 pn page S31 -----	1-7,10, 11

INTERNATIONAL SEARCH REPORT

International application No.
PCT/CZ2018/050055

Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:
2. ☐ Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:
3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

see additional sheet

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fees, this Authority did not invite payment of additional fees.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☒ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

1-7(completely); 10, 11(partially)

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.
- ☐ The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.
- ☐ No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. claims: 1-7(completely); 10, 11(partially)

Crystalline form of Tenapanor, method of preparation of crystalline form and uses thereof

2. claims: 8, 9(completely); 10, 11(partially)

Salts of Tenapanor, methods of preparation thereof and uses thereof

3. claims: 12-15

A method of preparation of Tenapanor; intermediates

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No

PCT/CZ2018/050055

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