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- (54) Benævnelse: **ANVENDELSEN AF PREGNAN- OG ANDROSTANSTEROIDER TIL FREMSTILLINGEN AF EN FARMACEUTISK SAMMENSÆTNING TIL BEHANDLINGEN AF CNS-SYGDOMME**
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DESCRIPTION

Field of the invention

[0001] The present disclosure concerns novel steroid compounds that act on the gamma-aminobutyric acid receptor-chloride ionophore (GABA_A-R) complex, and which can be used in the treatment of GABA and GABA-steroid related and/or steroid induced disorders of the central nervous system (CNS).

Background of the invention

[0002] The metabolites of progesterone, desoxycorticosterone, testosterone, androstenedione, cortisone and cortisol known as androstanolones and pregnanolones have been the subject of various studies, at least partially elucidating their role in the neurological signal system in mammals. The nomenclature differs in the field and therefore the IUPAC nomenclature will be used throughout this application. The steroids inducing CNS symptoms and disorders of interest in the present application all share a common feature in comprising a 3 α -hydroxy group, a 5 α or 5 β pregnane steroid body, and a ketone on position 20. Examples of such steroids are given in table 1:

Table 1. Nomenclature of the pregnanolone group

IUPAC - nomenclature	CAS Number
3 α -hydroxy-5 α -pregnan-20-one	516-54-1
3 α -hydroxy-5 β -pregnan-20-one	128-20-1
3 α ,21-dihydroxy-5 α -pregnan-20-one	567-02-2
3 α ,21-dihydroxy-5 β -pregnan-20-one	567-03-3
3 α ,11 β ,17 α ,21-tetrahydroxy-5 β -pregnan-20-one	53-02-1
3 α ,11 β ,17 α ,21-tetrahydroxy-5 α -pregnan-20-one	302-91-0
3 α -17 α ,21-trihydroxy-5 α -pregnan-11,20-dione	547-77-3
3 α -17 α ,21-trihydroxy-5 β -pregnan-11,20-dione	53-05-4

[0003] To the best knowledge of the inventors, all compounds described as novel in the description and examples are previously non-disclosed. Other steroids for the treatment of CNS disorders have however been disclosed, for example in the following documents:

[0004] U.S. 5,232,917 (Bolger et al.) and U.S. 5,925,630; 5,939,545; 6,143,736; 6,277,838, (Upasani et al.) disclose a number of 3 α -hydroxy steroids and some 3 β steroids. These patents concern the agonistic modulation of the GABA-A receptor. In other words, the patents are focused on 3 α -hydroxy-steroids and their benzodiazepine like effect. All steroids that are modulators of the GABA-A receptor have the common feature of one 3 α -hydroxy structure.

[0005] WO 99/45931 (Bäckström & Wang) discloses the antagonistic effect of one steroid, namely 3 β -OH-5 α -pregnan-20-one, but is silent about the steroids described in this application.

[0006] WO 03/059357 (Bäckström et al.) discloses several 3 β -hydroxy steroids and their antagonistic effect on the GABA-A receptor but is silent about the steroids described in this application.

[0007] WO94/27608 discloses steroid derivatives which interact with GABA_A receptor complex.

[0008] The antagonistic effects of 3 β -OH-5 α -pregnan-20-one and other 3 β -OH- 5 α / β pregnan-steroids are disclosed by Wang *et al.* (Wang M.D., Bäckström T. and Landgren S. (2000) The inhibitory effects of allopregnanolone and pregnanolone on the population spike, evoked in the rat hippocampal CA1 stratum pyramidale in vitro, can be blocked selectively by epiallo-pregnanolone, *Acta Physiol Scand* 169, 333-341 and Wang M, He Y, Eisenman LN, Fields C, Zeng CM, Mathews J et al., 3 β -hydroxypregnane steroids are pregnenolone sulfate-like GABA(A) receptor antagonists, *J Neurosci* 2002;22(9):3366-75). In those papers, dose dependent antagonistic effect of 3 β -OH-5 α / β -pregnan-steroids and sulfated-steroids are described. However the compounds in the present disclosure are not mentioned.

[0009] The present disclosure relates to the field of medicinal chemistry and is intended for producing compounds and compositions useful for modulation of the mammal brain excitability via the gamma-aminobutyric acid receptor-chloride ionophore (GABA_A-R) complex and other neurotransmitter systems which are, directly or indirectly, correlated to the GABA_A-R complex. It has been shown that a variety of steroidal molecules are effective in the modulation and stimulation of GABA signaling, displaying a variety of physiologic effects. The steroids comprising the components 3 α -hydroxy-5 α / β -pregnan-20-one have been shown to be specific GABA-A {gamma-aminobutyric acid (A)} receptor enhancers. Due to these properties, these naturally occurring stress- and sex steroids also have adverse effects and cause certain disorders. The adverse effects of the 3 α -hydroxy-pregnan-20-one-steroids are the basis for the negative CNS effects induced by these steroids. Examples of the adverse compounds are 3 α -hydroxy-5 α / β -pregnanolone steroids listed in table 1. Some of these steroids are very potent and have e.g. been shown to have an ability to induce anesthesia at a high pharmacological dose.

[0010] As the 3 α -hydroxy-pregnan-steroids are endogenously produced and are metabolites of steroid hormones essential for life, their production cannot easily be interrupted. These steroids are produced in high amounts during several days to weeks during acute and chronic stress, the luteal phase of the menstrual cycle and during pregnancy. They are also produced within the brain. Specific blockers are therefore needed as therapy.

[0011] It has earlier been shown that certain 3 β hydroxy pregnanolone steroids can block the negative brain effect of the aversive stress and sex steroids. A problem with the earlier discovered compounds is that they are easily metabolized in the body at the critical 3 position and that they are difficult to dissolve in water solution.

[0012] The direct mechanism at the receptor site has not yet been fully elucidated, due to the structural complexity of the GABA_A-R complex. The GABA receptor family includes several subunit compositions, of which some are known to be related to specific functions and disorders in the CNS. One aim of the present invention is thus to find new compounds that are useful in the treatment of anomaly in the excitability of GABA receptors or other neurotransmitters related to GABA receptors, in a manner which can be general or specific for some subunit compositions and functions. Disorders that are caused by the action of endogenously produced 3 α -hydroxy-5 α steroids or 3 α -hydroxy-5 β steroids on the GABA-A receptor are well characterized and understood. It is also known that 3 α -hydroxy-5 α / β -steroids can induce tolerance to themselves and other similar substances after exposure, and that abstinence effects occur at withdrawal of the 3 α -hydroxy-5 α / β -steroids. This will be further elucidated in the following:

Diseases caused by 3 α -hydroxy-pregna(e)n-steroids

a) Direct action

[0013] It is established that 3 α -hydroxy-5 α / β -steroids can directly cause inhibition of CNS functions. Examples of disorders and symptoms caused by the direct action of 3 α -hydroxy-5 α / β -steroids are premenstrual dysphoric disorder, premenstrual syndrome, dementia, Alzheimer's disease, sedation, tiredness, chronic fatigue syndrome, memory disturbance, learning disturbance, disturbance of motor function, fractures, clumsiness, increased appetite and food cravings, obesity, negative mood as tension, irritability, depression, decreased hearing and eye sight, worsening of Petit Mal epilepsy, burn out syndrome.

b) Tolerance

[0014] Continuous and long exposure to 3 α -hydroxy-5 α / β -steroids causes a malfunctioning of the GABA-A receptor system. A tolerance develops and this tolerance is the initial step in a process that ultimately leads to stress sensitivity, concentration difficulties, and loss of impulse control and depression. The action of 3 α -hydroxy-5 α / β -steroids have been found to be a factor which reinforces drug dependency. This has been the focus of extensive research.

c) Abstinence

[0015] A continuous but shorter exposure to 3 α -hydroxy-5 α / β -steroids results in a withdrawal effect when the exposure is ended. This phenomenon occurs *i.e.* during menstruation when the production of 3 α -hydroxy-5 α / β -steroids by the corpus luteum of the

ovary is interrupted. This withdrawal phenomenon also occurs after giving birth (*post partum*) when the 3 α -hydroxy-5 α / β -steroid production by the placenta is interrupted. The same phenomenon is also noted when a period of stress is ended. As a response to stress, the adrenals have produced 3 α -hydroxy-5 α / β -steroids. When this production is interrupted, withdrawal symptoms may occur. Examples of conditions that are influenced by this withdrawal/ abstinence phenomenon are partial epilepsy where the patient has an epileptic focus in the cerebral cortex where a worsening occurs at the withdrawal period during menstruation. This phenomenon is called "catamenial epilepsy". Other examples are menstrual related migraine and stress related migraine, mood changes *post partum* and weekend headache. Abstinence is a sign of an earlier developed tolerance.

[0016] In view of the above, it is clear that steroids are important drug candidates. Naturally occurring steroids are however subject to intense metabolism and are therefore usually not suitable for oral administration. Also in other routes of administration the metabolism is high and makes it impossible to use the compounds as medication and treatments as the active parts of the compounds are first destroyed by the metabolism.

[0017] A second problem with steroid compounds is that they are difficult to solute in water solutions and therefore difficult to administer *in vivo*.

[0018] These problems and others are solved by compounds according to the present disclosure.

Summary of the invention

[0019] The present inventors have synthesized new compounds that are protected against metabolism in the 3 position of the steroid. Surprisingly, these compounds also have increased water solubility, due to their modified structural features. The inventors do not wish to be bound to any theory, but it is assumed that the advantageous properties of the novel compounds are due to the presence of a double bond in the steroid core, and the replacement of a keto group with an oxime group at positions 20, 21 or 17 respectively in the mentioned scaffolds of pregnane, pregnene, androstane and androstene series.

[0020] In summary, it is known that 3 α -hydroxy-delta 4-5, 5 α / β -steroids cause CNS disorders through the above mentioned three possible mechanisms: a) direct action, b) tolerance induction, and c) withdrawal effect. The compounds made available through the embodiments of the present invention belong to the pregnane-, pregnene-, androstane- androstene- series, with the addition of suitable functionalities. The compounds can be used alone or as prodrugs and/or in combination with formulations and other compositions in order to enhance and modulate the effects on CNS. Compositions within the scope of this invention include all compositions wherein the compounds of this invention are contained in an amount that is effective to achieve the intended purposes.

[0021] To the best knowledge of the present inventors, this is the first time steroid compounds with increased resistance against metabolism and increased water solubility are disclosed. In addition, these substances are suggested for the manufacture of pharmaceuticals for the treatment of many specific steroid related or steroid induced CNS disorders and for use in methods of treatment, according to the attached claims which are incorporated herein by reference.

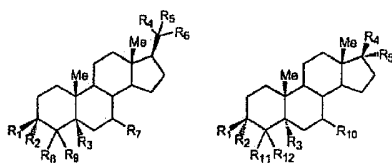
Detailed description of the invention

[0022] The scope of the invention is defined by the appended claims.

[0023] The disclosure concerns new compounds, with protection against metabolism and with increased water solubility, and methods of producing such compounds with antagonistic and blocking effects of the 3 α -hydroxy-pregna(e)n-steroid induced CNS disorders. The present invention is based on the discovery that steroids represented by the formulae I or II have an effect as modulators of the GABA receptor signaling as agonists, antagonists or inverse agonists.

[0024] The inventors have shown that the presence of a tertiary alcohol moiety in position 3 prolongs the half-life of a steroidal compound within the body through preventing metabolic oxidations or degradation in the body. The presence of a hydrogen-bond acceptor/donator group bonded to the D ring of a steroidal molecule influences the ability of the steroid to modulate the GABA receptor signaling.

[0025] The present disclosure relates to novel steroids represented by the formulae I or II, and pharmaceutically acceptable, salts thereof.



I

II

wherein

R₁ is chosen among an ethynyl, ethenyl, ethyl, or other saturated or unsaturated alkyl groups; hydroxyl, in its free form or combined with carboxylic acid residues, sugars, alkyl groups to form esters or ether or glycosylated compounds; fluorine or other halogens; a proton;

R₂ is chosen among an ethynyl, ethenyl, ethyl, or other saturated or unsaturated alkyl groups; hydroxyl, in its free form or combined with carboxylic acid residues, sugars, alkyl groups to form esters or ether or glycosylated compounds; fluorine or other halogens; a proton;

R₃ is 5 α - or 5 β -H

R₄ is a nitro, hydroxyl, free or bonded with an ester, ether, sugar; and

R₅ is a proton.

[0026] According to an embodiment of the disclosure, the compounds are steroids represented by the above formulae I or II, and pharmaceutically acceptable salts or solvates thereof, wherein

R₄, R₅ is = O or N as oxime =NOH, or a homo- or heterocycle;

R₆ is a methyl, an alkyl group or -CH₂OR where R is H, a carboxylic acid residue, an alkyl group or a sugar; -CH₂X where X is fluorine or another halogen;

R₇, R₁₀ is OH, CH₃ or H at the seven position.

R₈, R₉ or R₁₁, R₁₂ represent two Me- groups, or Me- and H, respectively, or two -H.

[0027] According to an embodiment R₇, R₁₀ is OH or CH₃ at the seven position.

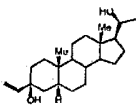
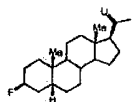
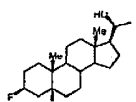
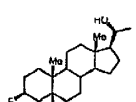
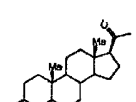
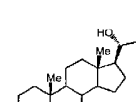
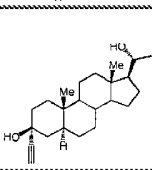
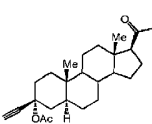
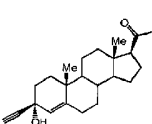
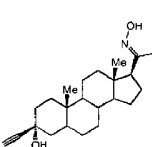
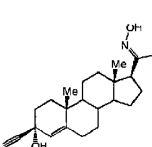
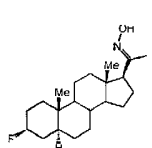
[0028] According to an embodiment of the disclosure, the compounds are steroids represented by the above formula II, or a pharmaceutically acceptable salt thereof, wherein R₁ is an ethynyl group or hydroxyl; R₂ is an ethynyl group or hydroxyl; R₃ is 5 α - or 5 β -H; R₄ and R₅ together is O or N as oxime =NOH; R₁₀ is H; and R₁₁ = R₁₂ = H.

[0029] The compounds of formula I and II may exist as optical isomers; consequently the disclosure includes all individual isomers that may be separated by common chromatographic techniques as well as other known separation methods known by those skilled in the art.

[0030] Tables 2 and 3 show examples of the structure of a series of compounds according to the disclosure, where the 3-hydroxy position of the pregnane, pregnene, androstane or androstene steroid structure is protected against metabolism in 3-position with an ethynyl, ethenyl or acetate structure added to the steroid molecule or the hydroxyl group replaced with a fluorine atom.

Table 2: New Compounds with metabolism protection based on formula I

UC2005		3 α -ethynyl, 3 β -hydroxyl, 5 β -pregnan-20-one
UC2007		3 α -ethynyl, 5 β -pregnan-3 β ,20(R)-diol

UC2009		3β-ethenyl, 5α-pregnan-3α, 20(R)-diol
UC2012		3β-fluor, 5β-pregnan-20-one
UC2013		3β-fluor, 5β-pregnan-20-(R)-ol
UC2014		3β-fluor, 5β-pregnan-20-(S)-ol
UC2016		3β-fluor, 5α-pregnan-20-one
UC2017		3α-fluor, 5α-pregnan-20-(R)-ol
UC2018		3α-ethynyl, 5α-pregnan-3β,20(R)-diol
UC2019		3β-ethynyl, 3α-acetyl, 5α-pregnan-20-one
UC2024		3β-ethynyl, 3α-hydroxyl, Δ4-pregnen-20-one
UC2026		3β-ethynyl, 3α-hydroxyl, 5α-pregnan-20-one oxide
UC2029		3β-ethynyl, 3α-hydroxyl, Δ4-pregnen-20-one oxime
UC2030		3α-fluor, 5α-pregnan-20-one oxime

UC2032		3-dimethyl, Δ5-pregnen-3β,20(R)-diol
UC2034		3β-fluor, 5α-pregnan-20-one oxime

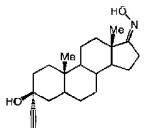
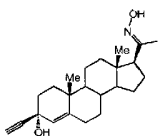
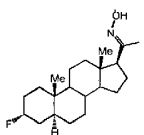
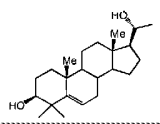
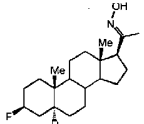
Table 3: New Compounds with metabolism protection based on formula II

UC2021		3β-ethynyl, 3α-hydroxyl, androstan-17-one
UC2025		3β-ethynyl, 3α-hydroxyl, androstan-17-one oxime
UC2027		3α-ethynyl, 3β-hydroxyl, androstan-17-one oxime

[0031] In Table 4 are shown examples of structures of a series of compounds where the water solubility is increased relative to the 5-saturated compound (UC2024) and where the water solubility is increased by adding an oxime group relative to a keto group or simple hydroxy group (UC2027, UC2029).

Table 4. Examples of new compounds with increased water solubility

UC2013		3β-fluor, 5β-pregnan-20-(R)-ol
UC2014		3β-fluor, 5β-pregnan-20-(S)-ol
UC2017		3α-fluor, 5α-pregnan-20-(R)-ol
UC2018		3α-ethynyl, 5α-pregnan-3β,20(R)-diol
UC2024		3β-ethynyl, 3α-hydroxyl, Δ4-pregnen-20-one

UC2027		3α-ethynyl, 3β-hydroxyl, androstan-17-oxime
UC2029		3β-ethynyl, 3α-hydroxyl, Δ4-pregnen-20-oxime
UC2030		3α-fluor, 5α-pregnan-20-oxime
UC2032		3-dimethyl, Δ5-pregnen-3β,20(R)-diol
UC2034		3β-fluor, 5α-pregnan-20-one oxime

HPLC retention times [min] for compounds according to the invention

HPLC conditions:

[0032] HPLC System (Waters) Column Symmetry® C₁₈ 3.5 μm 4.6 × 75 mm (Waters); T: 45 °C; flow 1.0 mL/min; isocratic eluent conditions 40:60 v/v H₂O:MeOH. Injection volume: 100 μL.

[0033] Solvents used for eluent are of HPLC grade, water filtered through a Millipore apparatus; all solvents were filtered through a 0,45 μm Millipore filter and degassed by N₂ stream prior use.

[0034] Since the analyses are carried in reverse phase, shorter retention times correspond to higher hydrophilicity.

Table 5

Reference substances:	retention time [min]
3beta-hydroxy-5beta-pregnan-20-one	24.4
3beta-hydroxy-5alpha-pregnan-20-one	30.0

Table 6

Compounds:	retention time [min]
UC 2021	7.8
UC 2024	14.0
UC 2025	13.9
UC 2026	16.9

[0035] As seen above in Tables 5 and 6 the retention times for the inventive compounds are shorter than for the reference

compounds, indicating that the former have higher hydrophilicity than the latter.

[0036] The synthesis, the separation and the identification of the compounds in this invention can be performed using method steps known as such by those skilled in the art.

[0037] The present inventors have surprisingly found that the reaction of the ethynyl Grignard reagent with 3, 20/17 diketone steroids is in the most cases selective for the position 3 and no protection/deprotection for the other ketone functionality was required. Both α and β isomers are formed, which can be separated by chromatographic methods and recrystallization.

[0038] The starting materials for the compounds described in the present invention are the corresponding steroids with a 3-hydroxyl substituent and a keto group in positions 20 or 17. They can be converted into the respective diones by oxidation with IBX reagent. The reaction proceeds smoothly and with complete conversion. Other suitable steroids can be employed as starting material when required.

[0039] The reactions are carried out in a suitable solvent such as methanol, ethanol, water, THF, diethyl ether, dichloromethane or other solvents or mixture of solvents that one skilled in the art can recognize as suitable.

[0040] The reactants are chosen in order to avoid, when possible, use of reactants, such as heavy metals, which are toxic even in traces or are difficult to be completely removed in the workup procedure.

[0041] Reactions involving air or moisture sensitive reagents or products are carried out under inert atmosphere, such as nitrogen or argon gas, in the presence of dry solvents. Diethyl ether and tetrahydrofuran are dried over Na in the presence of benzophenone. Syringes purged with inert gas were used for the transfer of reagents and dry solvents. Optimized reaction conditions, such as time and temperature, were determined by monitoring the formation of products and the loss of starting material using a suitable chromatographic technique such as TLC or GC/MS.

[0042] Purifications are carried out by using chromatographic techniques such as flash silica chromatography or preparative high performance liquid chromatography (HPLC) by using a HPLC apparatus. Those skilled in the art can recognize that alternative purification methods can be employed, and laboratory chromatographic techniques can be adapted to industrial scale by using chromatographic columns for scaled preparations.

[0043] Identification of the products is carried out by using suitable analytical techniques such as $^1\text{H-NMR}$, $^{13}\text{C-NMR}$, mass spectrometry, IR spectroscopy and any other assay that one skilled in the art would recognize as suitable for structural identification and purity determination of the compounds in the present invention.

[0044] The compounds according to the invention have *inter alia* the advantage of being both protected against metabolism, and more easily water soluble. The method of synthesis has the advantage of consisting of steps known as such, and is comparatively easy - once disclosed-to use.

[0045] The following examples of steroids in the present disclosure are provided. The examples are illustrative, but not limiting, of the methods and compositions of the present invention. One skilled in the art will recognize that similar reagents, solvents, conditions and parameters can be used in the reactions, depending on the substrate. NMR data were recorded using a Bruker 400 MHz spectrometer.

EXAMPLES

Examples based on formula I

Reference Example 1: UC2016 - 3 β -fluor, 5 α -pregnan-20-one

[0046] 3 α -OH 5 α -pregnan-20-one (3 mmol), was dissolved in 20 mL dry dichloromethane under N_2 atmosphere. DAST (700 mg, 4.33 mmol) was added slowly dropwise at room temperature (rt) and the resulting yellowish solution was left stirring at rt for 1 h. Reaction was followed by TLC. The solution was quenched by slow addition of a 5 % NaHCO_3 solution (60 mL). The water phase

was extracted with dichloromethane (3 x 20 mL), the organic phases collected dried over MgSO₄ and the solvent removed under reduced pressure, yielding a yellowish oil which was purified by silica flash column chromatography (pentane : ethyl acetate 9 : 1). Products are, in the order: elimination of H₂O at 2,3 positions (yield: 67 %); fluorination at 3-OH with inversion of configuration 30 % (UC2016); fluorination at 3-OH with retention of configuration (3 % - traces). ¹H NMR (400 MHz, CDCl₃-d₆): δ 4.57-4.37 (dm, 1H); 2.51 (t, 1H); 2.12 (m, 1H); 2.11 (s, 3H); 2.02-1.99 (m, 2H); 0.83 (s, 3H); 0.67 (m, 1H); 0.61 (s, 3H).

Reference Example 2: UC2018 - 3α-ethynyl, 5α-pregnan-3β,20(R')-diol

[0047] 3α-ethynyl, 3β-hydroxy, 5α-pregnan-20-one (0.3 mmol) was dissolved in a solution of 2 mL dichloromethane and 5 mL MeOH at rt, in a flask with outlet to air. NaBH₄ (2.1 equiv.) was added in one portion and the suspension left stirring for 3 h at rt. The colorless solution was evaporated in vacuum, yielding a white residue which was extracted with 20 + 20 mL H₂O diethyl ether. The aqueous phase was extracted with 30 mL dichloromethane: diethyl ether 1:1, the organic phases collected, dried over MgSO₄, and the solvents removed under vacuum. The white solid was purified by silica flash column chromatography (1:4 diethyl ether : dichloromethane), Quantitative overall yield. The product with (R)-configuration at 20-C is the main product (90 %), as determined by NMR measurements.

¹H NMR (400 MHz, CDCl₃-d₆): δ 3.72 (m, 1H); 2.49 (s, 1H); 2.02 (m, 1H); 1.86 (2m, 2H); 1.12 (d, 3H); 0.80 (s, 3H); 0.74 (s, 3H).

Reference Example 3: UC2019 - 3β-ethynyl, 5α-pregnan-20-one 3α-acetate

[0048] 3β-ethynyl, 3α-hydroxy, 5α-pregnan-20-one (0.25 mmol) and pyridine (2 equiv.) were dissolved in dry dichloromethane, followed by the dropwise addition of acetic anhydride (4 equiv.) at rt under nitrogen atmosphere.

[0049] The mixture was left stirring at 40 °C under three days. The dark mixture was quenched by the addition of 50 mL HCl 10%, then washed with a NaHCO₃ 10% aqueous solution (2 x 30 mL) until pH = 7. The organic phase was collected, dried over MgSO₄, and concentrated. The yellowish residue was purified by silica flash column chromatography (1:4 diethyl ether: dichloromethane) to afford the ester in 87 % yield.

¹H NMR (400 MHz, CDCl₃-d₆): δ 2.60 (s, 1H); 2.51 (t, 1H); 2.43 (m, 1H); 2.11 (s, 3H); 2.03 (s, 3H); 0.82 (s, 3H); 0.60 (s, 3H).

Reference Example 4: UC2024 - 3β-ethynyl, 3α-hydroxyl, Δ-4-pregnen-20-one

[0050] Progesterone (1 mmol) was dissolved in 25 mL dry THF at rt under nitrogen. Ethynyl magnesium bromide (1.1 equiv.) was added dropwise at rt under stirring and the solution was left stirring overnight at rt under nitrogen. The yellowish solution was then quenched with saturated NH₄Cl(aq) and the aqueous phase extracted with dichloromethane (3 x 30 mL). The collected organic phases were evaporated under reduced pressure, the resulting yellow oil dissolved in dichloromethane, washed with brine and dried over MgSO₄. The solution was reduced under vacuum, and the residue purified by silica flash column chromatography (1:4 diethylether : dichloromethane), typical yields 30 %.

¹H NMR (400 MHz, CDCl₃-d₆): δ 5.32 (s, 1H); 2.51 (m, 2H); 2.14 (m, 2H); 2.11 (s, 3H); 1.05 (s, 3H); 0.64 (s, 3H).

Reference Example 5: UC2026 - 3β-ethynyl, 3α-hydroxyl, 5α-pregnan-20-one oxime

3β-ethynyl, 3α-hydroxyl, 5α-pregnan-20-one

[0051] 3,20-5α-pregnandione (1.580 g, 5.0 mmol) was dissolved in 50 mL dry THF at rt under nitrogen. Ethynyl magnesium bromide (1.1 equiv) was added dropwise at rt under stirring and the solution was left stirring overnight at rt under nitrogen flow. The yellowish solution was then quenched with saturated NH₄Cl(aq) and the aqueous phase extracted with dichloromethane (3 x 30 mL). The collected organic phases were evaporated under reduced pressure, the resulting yellow oil dissolved in dichloromethane, washed with brine and dried over MgSO₄. The solution was reduced under vacuum, and the residue purified by silica flash column chromatography (1:4 diethylether : dichloromethane), typical yields 13%. Eventual traces of byproducts can be eliminated by further recrystallization from diethylether.

¹H NMR (400 MHz, CDCl₃-d₆): δ 2.51 (t, 1H); 2.43 (s, 3H); 2.14 (m, 1H); 2.11 (s, 3H); 0.81(s, 1H); 0.60 (s, 3H).

3α-ethynyl, 3β-hydroxyl, 5α-pregnan-20-one

[0052] Was obtained as a byproduct from the above described reaction and separated by silica flash column chromatography. Typical yield 72 %.

¹H NMR (400 MHz, CDCl₃-d₆): δ 2.52 (t, 1H); 2.47 (s, 1H); 2.11 (s, 3H); 0.80 (s, 3H), 0.60 (s, 3H).

3β-ethynyl, 3α-hydroxyl, 5α-pregnan-20-one oxime

[0053] 3β-ethynyl, 3α-hydroxyl, 5α-pregnan-20-one (10 mmol) is dissolved in dichloromethane 5 mL and ethanol 50 mL at rt and air atmosphere, in a 250 mL round bottom flask. 4 equiv. of NH₂OH chlorhydrate and 4 equiv. of sodium acetate are dissolved in 5 mL H₂O and then added to the steroid solution. 20 mL of ethanol are added and the mixture put on reflux overnight. The mixture is then cooled and the solvent removed under reduced pressure. The white residue is then treated with 50 mL H₂O and 50 mL dichloromethane, the aqueous phase extracted with 3 x 30 mL dichloromethane. The collected organic phases are then dried over MgSO₄, filtrated and the solvent removed under reduced pressure. The final residue is purified by silica flash column chromatography dichloromethane : diethyl ether 4:1, typical yields 95-100%.

¹ H NMR (400 MHz, CDCl₃-d₆ δ 2.43 (s, 1H); 2.22 (t, 1H); 2.05 (m, 1H); 1.88 (s, 3H); 1.86 (m, 1H); 0.81 (s, 3H), 0.62 (s, 3H).

Reference Example 6: UC2029 - 3β-ethynyl, 3α-hydroxyl, Δ4-pregnen-20-one oxime

[0054] 3β-ethynyl, 3α-hydroxyl, Δ4-pregnen-20-one (10 mmol) is dissolved in dichloromethane 5 mL and ethanol 50 mL at rt and air atmosphere, in a 250 mL round bottom flask. 4 equiv. of NH₂OH chlorhydrate and 4 equiv. of sodium acetate are dissolved in 5 mL H₂O and then added to the steroid solution. 20 mL of ethanol are added and the mixture put on reflux overnight. The mixture is then cooled and the solvent removed under reduced pressure. The white residue is then treated with 50 mL H₂O and 50 mL dichloromethane, the aqueous phase extracted with 3 x 30 mL dichloromethane. The collected organic phases are then dried over MgSO₄, filtrated and the solvent removed under reduced pressure. The final residue is purified by silica flash column chromatography dichloromethane : diethyl ether 4:1, typical yields 85 %.

¹H NMR (400 MHz, CDCl₃-d₆): δ 5.32 (s, 1H); 2.51 (s, 1H); 2.19 (m, 2H); 2.06 (m, 1H); 1.88 (s, 3H); 2.03 (s, 3H); 1.05 (s, 3H); 0.65 (s, 3H).

Reference Example 7: UC2030 - 3α-fluor, 5α-pregnan-20-one oxime

3α-fluor, 5α-pregnan-20-one

[0055] 3α-OH 5α-pregnan-20-one (3 mmol), was dissolved in 20 mL dry dichloromethane under N₂ atmosphere. DAST (700 mg, 4.33 mmol) was added slowly dropwise at -78 °C and the resulting yellowish solution was left stirring at rt for 1 h. Reaction was followed by TLC. The solution was quenched by careful addition of a 5 % NaHCO₃ solution (60 mL). The water phase was extracted with dichloromethane (3 x 20 mL), the organic phases collected dried over MgSO₄ and the solvent removed under reduced pressure, yielding a yellowish oil which was purified by silica flash column chromatography (pentane : ethyl acetate 9 : 1). Products are, in the order: elimination of H₂O at 2,3 positions (yield: 67 %); fluorination at 3-OH with inversion of configuration 30 %; fluorination at 3-OH with retention of configuration (3 % - traces).

¹H NMR (400 MHz, CDCl₃-d₆): δ 4.87-4.75 (d, 1H); 2.53 (t, 1H); 2.11 (s, 3H); 2.00 (m, 1H); 0.95 (m, 1H); 0.80 (m, 1H); 0.78 (s, 3H); 0.60 (s, 3H).

3α-fluor, 5α-pregnan-20-one oxime

[0056] 3α-fluor, 5α-pregnan-20-one (10 mmol) is dissolved in dichloromethane 5 mL and ethanol 50 mL at rt and air atmosphere,

in a 250 mL round bottom flask. 4 equiv. of NH_2OH chlorhydrate and 4 equiv. of sodium acetate are dissolved in 5 mL H_2O and then added to the steroid solution. 20 mL of ethanol are added and the mixture put on reflux overnight. The mixture is then cooled and the solvent removed under reduced pressure. The white residue is then treated with 50 mL H_2O and 50 mL dichloromethane, the aqueous phase extracted with 3 x 30 mL dichloromethane. The collected organic phases are then dried over MgSO_4 , filtrated and the solvent removed under reduced pressure. The final residue is purified by silica flash column chromatography (dichloromethane : diethyl ether 4:1). Quantitative yield.

^1H NMR (400 MHz, CDCl_3 - d_6): δ 4.90-4.78 (d, 1H); 2.26 (t, 1H); 2.10 (m, 1H); 1.90 (s, 3H); 0.98 (m, 1H); 0.82 (s, 3H); 0.65 (s, 3H).

Reference Example 8: UC2034 - 3 β -fluor, 5 α -pregnan-20-one oxime

[0057] was obtained by using the same synthetic protocol as Example 7 for UC2030 starting from the corresponding 3 β -fluor, 5 α -pregnan-20-one isomer.

Reference Example 9: UC2032 - 3-dimethyl, Δ^5 -pregnen-3 β ,20(R)-diol

3-dimethyl, Δ^5 -pregnen-3,20-dione

[0058] 525 mg progesterone are dissolved in 10 mL dry toluene at rt. 3.4 mL (2 equiv.) of a 1.0 M solution of sodium t-buthylate in dry toluene is added dropwise to the progesterone solution, under stirring and N_2 atmosphere. The yellowish solution is let stirring in 20 min. 2 equiv. of MeI are then added dropwise to the mixture, which is stirred overnight at rt under N_2 .

[0059] The mixture is quenched with 10 mL water and 10 mL dichloromethane, the aqueous phase extracted with 2 x 30 mL dichloromethane. The organic phases are collected, dried over MgSO_4 , the solvent removed under vacuum yielding a yellowish residue, which is purified by silica flash column chromatography (1:4 diethyl ether : dichloromethane). A further purification of the desired fraction is performed by silica flash column chromatography (1:9 ethyl acetate : pentane). Yield: 25 %.

^1H NMR (400 MHz, CDCl_3 - d_6): δ 5.56 (m, 1H); 2.54 (m, 3H); 2.13 (s, 3H); 1.23 (s, 6H); 0.86 (s, 3H); 0.64 (s, 3H).

3-dimethyl, Δ^5 -pregnen-3 β ,20(R)-diol

[0060] 91 mg of 3-dimethyl, Δ^5 -pregnen-3,20-dione are dissolved in 3.0 mL dichloromethane and 15 mL MeOH at rt, in a flask with outlet to air. NaBH_4 (2.1 equiv.) is added in one portion and the suspension left stirring for 6 h at rt. The colorless solution was evaporated in vacuum, yielding a white residue which was extracted with 20 + 20 mL H_2O diethyl ether. The aqueous phase was extracted with 30 mL dichloromethane: diethyl ether 1:1, the organic phases collected, dried over MgSO_4 , and the solvents removed under vacuum. The white solid was purified by silica flash column chromatography (1:4 diethyl ether : dichloromethane), 95 % yield.

^1H NMR (400 MHz, CDCl_3 - d_6): δ 5.60 (m, 1H); 3.75 (m, 1H); 3.26 (m, 1H); 2.09-2.13 (m, 2H); 1.18 (s, 6H); 1.21 (s, 3H); 1.10 (s, 3H); 0.80 (s, 3H).

Examples based on formula II

Reference Example 10: UC2021 - 3 β -ethynyl, 3 α -hydroxyl, androstan-17-one

3 β -ethynyl, 3 α -hydroxyl, androstan-17-one

[0061] 3, 17 androstandione (5.0 mmol) was dissolved in 50 mL dry THF at rt under nitrogen. Ethynyl magnesium bromide (1.1

equiv) was added dropwise at rt under stirring and the solution was left stirring overnight at rt under nitrogen flow.

[0062] The solution was then quenched with saturated $\text{NH}_4\text{Cl}(\text{aq})$ and the aqueous phase extracted with dichloromethane (3 x 30 mL). The collected organic phases were evaporated under reduced pressure, the resulting yellow oil dissolved in dichloromethane, washed with brine and dried over MgSO_4 . The solution was reduced under vacuum, and the residue purified by silica flash column chromatography (1:4 diethylether : dichloromethane), typical yields 8 %. Eventual traces of byproducts can be eliminated by further recrystallization from diethylether.

^1H NMR (400 MHz, $\text{CDCl}_3\text{-d}_6$): δ 2.43 (s, 1H); 2.42 (m, 1H); 2.10-2.04 (m, 2H); 1.02 (m, 1H); 0.86 (s, 3H); 0.83 (s, 3H).

3 α -ethynyl, 3 β -hydroxyl, androstane-17-on

[0063] Was obtained as a byproduct from the above described reaction and separated by preparative HPLC chromatography. Typical yield 65%.

^1H NMR (400 MHz, $\text{CDCl}_3\text{-d}_6$): δ 2.47 (s, 1H); 0.86 (s, 3H), 0.83 (s, 3H).

Reference Example 11: UC2025 - 36-ethynyl, 3 α -hydroxyl, androstan-17-one oxime

[0064] 3 β -ethynyl, 3 α -hydroxyl, androstan-17-one (10 mmol) is dissolved in dichloromethane 5 mL and ethanol 50 mL at rt and air atmosphere, in a 250 mL round bottom flask. 4 equiv. of NH_2OH chlorhydrate and 4 equiv. of sodium acetate are dissolved in 5 mL H_2O and then added to the steroid solution. 20 mL of ethanol are added and the mixture put on reflux overnight. The mixture is then cooled and the solvent removed under reduced pressure. The white residue is then treated with 50 mL H_2O and 50 mL dichloromethane, the aqueous phase extracted with 3 x 30 mL dichloromethane. The collected organic phases are then dried over MgSO_4 , filtrated and the solvent removed under reduced pressure. The final residue is purified by silica flash column chromatography dichloromethane : diethyl ether 4:1, typical yields 95-100 % (quantitative).

^1H NMR (400 MHz, $\text{CDCl}_3\text{-d}_6$): δ 2.56-2.41 (m, 2H); 2.43 (s, 1H); 1.87 (m, 2H); 1.00 (m, 1H); 0.80 (m, 1H); 0.90 (s, 3H), 0.83 (s, 3H).

Example 12: UC2027- 3 α -ethynyl, 3 β -hydroxyl, androstan-17-one oxime (according to the invention)

[0065] The title compound is obtained with the same procedure described for UC2025, starting from the corresponding 3 α -ethynyl, 3 β -hydroxyl, androstan-17-one, which is obtained as a by-product from the reaction described for the synthesis of UC2021.

^1H NMR (400 MHz, $\text{CDCl}_3\text{-d}_6$): δ 2.51-2.47 (m, 2H); 2.48 (s, 1H); 1.00 (m, 1H); 0.80 (m, 1H); 0.90 (s, 3H), 0.83 (s, 3H).

REFERENCES CITED IN THE DESCRIPTION

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- **WANG M HE YEISENMAN LNFIELD S CZENG CMMATHEWS J et al.**3 β -hydroxypregnane steroids are pregnenolone sulfate-like GABA(A) receptor antagonistsJ Neurosci, 2002, vol. 22, 93366-75 [0008]

Patentkrav

1. Forbindelse som er 3 α -ethynyl, 3 β -hydroxy, androstan-17-on-oxim eller et farmaceutisk acceptabelt salt deraf.

5 **2.** Farmaceutisk sammensætning omfattende en farmaceutisk effektiv mængde af en forbindelse ifølge krav 1 og en farmaceutisk acceptabel bærer.

3. Forbindelse ifølge krav 1 til anvendelse i lindring, forebyggelse eller behandling af en CNS-sygdom.