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(54) **Title**: USE OF ESTROGEN DERIVATIVES FOR THE MANUFACTURE OF PHARMACEUTICAL PRODUCTS USEFUL FOR THE PROPHYLAXIS AND/OR TREATMENT OF PSYCHIATRIC DISEASES

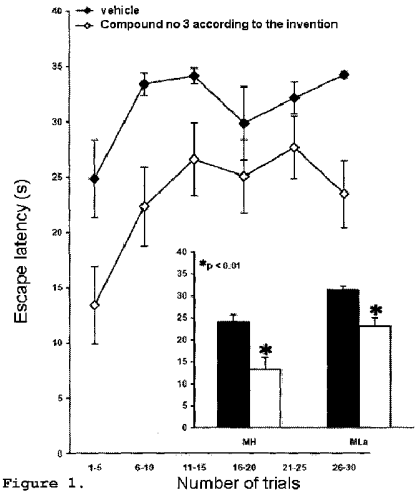
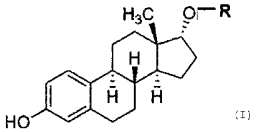


Figure 1.



(57) **Abstract**: The object of the invention is the use of the derivatives of 17-alpha-estradiol of formula (I) where R means • ethyl, higher alkyl, cycloalkyl, aryl and heterocycle as well as hydroxyl, alkyl groups containing amino and carboxy groups; or • CO-O-R', where R' means alkyl, cycloalkyl, aryl and heterocycle; or • CO-(R")-0-Q, where R" means alkyl, Q means alkyl, cycloalkyl, aryl and heterocycle, for the treatment and/or prevention of psychiatric diseases. According to the invention 17-alpha-estradiol derivatives can be used primarily as an antidepressant, more preferably fast- acting antidepressant. According to the invention derivatives of 17-alpha-estradiol of formula (I) can be used to treat and/or prevent post-partum depression, peri-menopausal depression, anxiety, schizophrenia, memory disturbance associated with depression, bipolar depression, Alzheimer-disease or post-traumatic stress disease.

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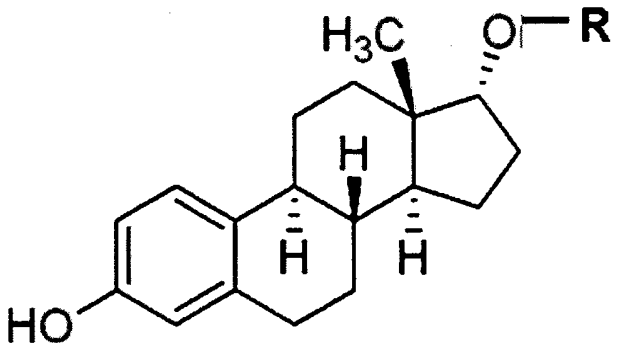
USE OF ESTROGEN DERIVATIVES FOR THE MANUFACTURE OF PHARMACEUTICAL PRODUCTS USEFUL FOR THE PROPHYLAXIS AND/OR TREATMENT OF PSYCHIATRIC DISEASES

The object of our invention is using 17-alpha-estradiol derivatives for the treatment and/or prophylaxis of psychiatric diseases. Our invention relates specifically to the use of 17-alpha-estradiol derivatives for the treatment and/or prophylaxis of depression as well as post-partum depression, peri-menopausal depression, anxiety, schizophrenia and memory disturbance associated with depression together with the treatment and/or prophylaxis of bipolar depression, Alzheimer-disease and post-traumatic stress disease. The object of our invention is also the procedure for the manufacture of pharmaceutical products containing 17-alpha-estradiol derivatives as active agents for the treatment and/or prophylaxis of psychiatric diseases mentioned above, and such pharmaceutical products.

The 17-alpha-estradiol derivatives according to the present invention have the formula (I):



(I),

where

R means

- ethyl, higher alkyl, cycloalkyl, aryl and heterocycle as well as hydroxyl, alkyl groups containing amino and carboxy groups; or
- CO-O-R', where R' means alkyl, cycloalkyl, aryl and heterocycle; or
- CO-(R")-O-Q, where R" means alkyl, Q means alkyl, cycloalkyl, aryl and heterocycle.

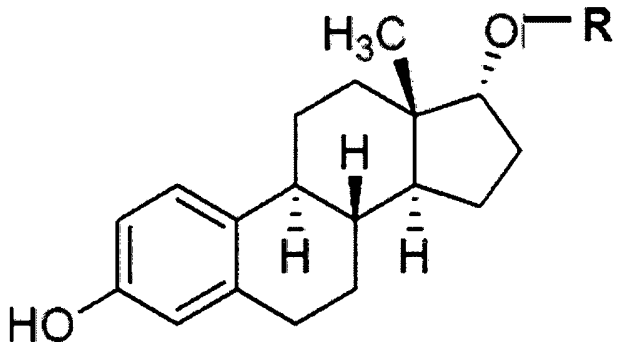
It is known, that 17-alpha-estradiol has neuroprotective effects [Dykeus JA: Development of 17-α-estradiol as a neuroprotective therapeutic agent. Ann NY Acad Sci 1052:116-135 (2005)], specific example was given, that 17-alpha-estradiol has neuroprotective effects on SK-N-SH cells [Green PS et al.: J Neurosci 17(2):511-515 (1997)]. It has also been described that 17-alpha-estradiol develops favorable effects on Alzheimer-disease treatment [Onio H et al.: Estrogen and non-feminizing estrogen for Alzheimer-disease. Endocrin Journal 50(4):361-367 (2003)].

A treatment method for estrogen deficiency of the central nervous system was described in the US patent specification nr. 6 245 756, whereby patients requiring treatment are administered steroid compound in an efficient amount which has selective neurotropic transcriptional effect in contrary to characteristics of the systemic effect of estrogen compounds.

The objective of our invention is using 17-alpha-estradiol derivatives in the indications of psychiatric diseases.

We solved this objective by the synthesis and use of new type 17-alpha-estradiol derivatives.

The object of our invention is a process for the preparation of pharmaceutical products in such a way where the 17-alpha-estradiol (I) formula's one or more derivatives which were produced in a known manner



(I),

where

R means

- ethyl, higher alkyl, cycloalkyl, aryl and heterocycle as well as hydroxyl, alkyl groups containing amino and carboxy groups; or
- CO-O-R', where R' means alkyl, cycloalkyl, aryl and heterocycle; or
- CO-(R")-O-Q, where R" means alkyl, Q means alkyl, cycloalkyl, aryl and heterocycle.

are mixed with inert pharmaceutical carriers and/or excipients and form them into pharmaceutical preparations that are capable of treating and/or preventing psychiatric diseases.

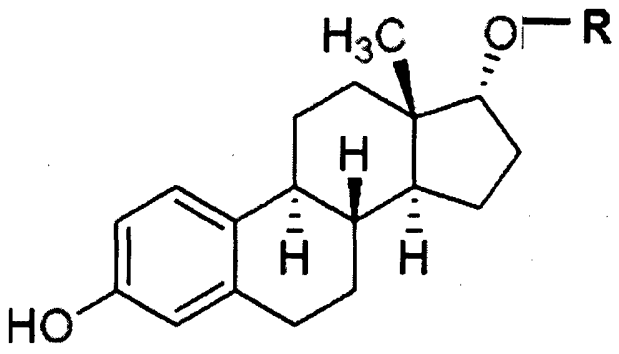
According to a preferred embodiment of our invention pharmaceutical preparations are produced with antidepressant effect, especially preferred with fast-acting antidepressant effect.

As a special embodiment of the indication mentioned above we produce pharmaceutical preparations that are capable of the

treatment and/or prophylaxis of post-partum depression, or peri-menopausal depression.

According to a further embodiment of our invention pharmaceutical preparations are produced that are capable of the treatment and/or prophylaxis of anxiety, schizophrenia, memory disturbance associated with depression, bipolar depression, Alzheimer-disease and post-traumatic stress disease.

The further object of our invention is the use of 17-alpha-estradiol (I) formula's one or more derivatives



(I),

where

R means

- ethyl, higher alkyl, cycloalkyl, aryl and heterocycle as well as hydroxyl, alkyl groups containing amino and carboxy groups; or
- CO-O-R', where R' means alkyl, cycloalkyl, aryl and heterocycle; or
- CO-(R")-O-Q, where R" means alkyl, Q means alkyl, cycloalkyl, aryl and heterocycle.

for the treatment and/or prophylaxis of psychiatric diseases.

A preferable embodiment of the above aspect of our invention is the use 17-alpha-estradiol (I) formula's derivatives as antidepressants, especially preferable fast-acting antidepressants.

A special embodiment of the use mentioned above is using 17-alpha-estradiol (I) formula's one or more derivatives for the treatment and/or prophylaxis of post-partum depression, or peri-menopausal depression.

According to the above aspect of the invention, 17-alpha-estradiol (I) formula's derivatives can be used furthermore also for the treatment and/or prophylaxis of anxiety, schizophrenia, memory disturbance associated with depression, bipolar depression, Alzheimer-disease or post-traumatic stress disease.

A further object of our invention is also the pharmaceutical product that is capable of the treatment and/or prophylaxis of psychiatric diseases, which contains 17-alpha-estradiol (I) formula's one or more derivatives as the active agent and inert solid or liquid pharmaceutically acceptable carriers and/or excipients.

According to our invention's preferred aspect the pharmaceutical preparation has antidepressant effects. According to another preferred aspect of the pharmaceutical preparation of the invention, it has fast-acting antidepressant effects.

The aforesaid aspect of the invention is also referring to pharmaceutical preparations for post-partum depression, or peri-menopausal depression.

The pharmaceutical preparations of the invention are also especially suitable for the treatment and/or prophylaxis of anxiety, schizophrenia, memory disturbance associated with depression, bipolar depression, Alzheimer-disease and post-traumatic stress disease.

The object of our invention is also the process for the treatment and/or prophylaxis of psychiatric diseases in such a way where a pharmaceutically suitable amount of preparation containing 17-alpha-estradiol (I) formula's one or more derivatives are administered for patients in need of treatment.

The daily dose of the derivative of 17-alpha-estradiol of (I) formula is 0,2-300 µg/kg, preferably 0,5-100 µg/kg, especially preferably 5-50 µg/kg.

According to all the foresaid aspects of the invention, 17-alpha-estradiol derivatives could be their's pharmaceutically acceptable salts as well.

Brief description of the drawing:
in Figure 1. the result of the experiment of the effect of the analysis with compound of example 3 on learned helplessness depression animal model is shown.

The term „higher alkyl group" means straight or branched chain of alkyl groups with 3-20 carbon atoms (e.g. isopropyl-group, butyl-group, hexyl-group, etc.).

The term „cycloalkyl group" means cyclic groups with 3-8 carbon atoms (eg. cyclopropyl-, cyclobutyl-, cyclohexyl-group etc.).

The term „aryl-group" refers to monocyclic or bicyclic aromatic hydrocarbon-groups (eg. phenyl-, naphthyl-group etc.).

The term „heterocyclic-group" means 3-7 parts, preferably 5 or 6 parts of aromatic groups containing one or more oxygen-, nitrogen- and/or sulfur atoms (eg. pyridyl-, pyrimidyl-, pyrrolyl-, oxazolyl-group etc.).

The 17-alpha-estradiol derivatives according to the present invention have better biological availability and/or absorption characteristics than the given characteristics of 17-alpha-estradiol, therefore these compounds might have more favorable pharmaceutical characteristics. These derivatives are most likely acting as prodrugs of 17-alpha-estradiol. These derivatives are probably better active agents than 17-alpha-estradiol because their water solubility is improved by derivation, for example they absorb better from the stomach. Therefore those derivatives are considered preferred by the invention which have better water solubility than 17-alpha-estradiol. For a pharmaceutical researcher those functional groups and moieties that can be used to improve the water solubility of a molecular-sceleton are obvious.

Solubilizing means can also be used to achieve water solubility, such as cyclodextrins. For example, regarding to the poorly soluble prednisolone beta-cyclodextrin was described as a solubilizing agent, where prednisolone was absorbed in a controlled way and fully where tablets contained hydroxypropyl methylcellulose as a matrix [Rao VM,

Haslam JL, Stella VJ: J Pharm Sci. 2001 Jul; 90(7):807-816.].

It was not to be foreseen that the 17-alpha-estradiol derivatives according to the present invention show antidepressant effect. It is known that depression occurs twice as often in women than men [Kessler RC et al.: Sex and depression in the National Comorbidity Survey. I: Lifetime prevalence, chronicity and recurrence. J Affect Disord 29:85-96 (1993)], and many elucidates the difference by the 17-beta-estradiol presence in women [Steiner M et al.: Hormones and mood: from menarche to menopause and beyond. J Affect Disord 74:67-83 (2003)]. Therefore numerous preclinical and clinical research corroborates that 17-beta-estradiol does not have antidepressant effect [Morrison MF et al.: Lack of efficacy of estradiol for depression in postmenopausal women: a randomized, controlled trial. Biol Psychiatry 55:406-412 (2004); Resnick SM et al.: Effects of combination estrogen plus progestin hormone treatment on cognition and affect. J Clin Endocrinol Metab 91:1802-1810 (2006); LaPlant Q et al.: Role of nuclear factor kappaB in ovarian hormone-mediated stress hypersensitivity in female mice. Biol Psychiatry 65:874-880 (2009); Autry AE et al.: Gender-specific impact of brain-derived neurotrophic factor signaling on stress-induced depression-like behavior. Biol Psychiatry 66:84-90 (2009)].

The 17-alpha-estradiol derivatives according to the present invention can be used prefereble as antidepressants, especially preferred fast-acting antidepressants. The advantage of the latter usage is the preparation's very rapid effect. One has to take the antidepressants known by the state of the art for 3-4 weeks to develop favorable effects, while the pharmaceutical preparation according to

the present invention develops favorable effects in just a few hours. Therefore the 17-alpha-estradiol derivatives according to the present invention are capable of ambulatory treatments in indications of the invention (e.g. after a suicide attempt).

The pharmaceutical preparation according to the invention are usually containing the active agent in an amount of 0.1-95 weight%, preferably 1-50 weight%, subservient 5-30 weight%.

The pharmaceutical preparation is preferably useful for peroral, parenteral, subcutaneous or rectal administration.

The solid pharmaceutical preparations that can be administered perorally can be powders, capsules, tablets, film-coated tablets, microcapsules etc., and can contain carriers such as binders like gelatine, sorbitol, poly (vinyl pyrrolidone) etc.; fillers like lactose, glucose, starch, calcium phosphate etc.; tabletting aids like magnesium stearate, talc, poly (ethylene glycol), silicon dioxide etc.; wetting agents like sodium lauryl sulfate etc.

The liquid pharmaceutical preparations that can be administered perorally are solutions, suspensions or emulsions and can contain carriers such as suspending agents like gelatin, carboxy-methylcellulose etc.; emulsifiers like sorbitan monooleate etc.; solvents like water, oils, glycerol, propylene glycol, ethanol etc.; preservatives like p-hydroxybenzoic acid methyl- or -propyl ester etc.

The pharmaceutical preparations that can be administered parenterally are usually made from the sterile solution of the active agent.

The examples mentioned above and the other ways of dosing are known themselves by the state of the art, see e.g. the handbook of Remington's Pharmaceutical Sciences, 18th edition, Mack Publishing Co., Easton, USA (1990).

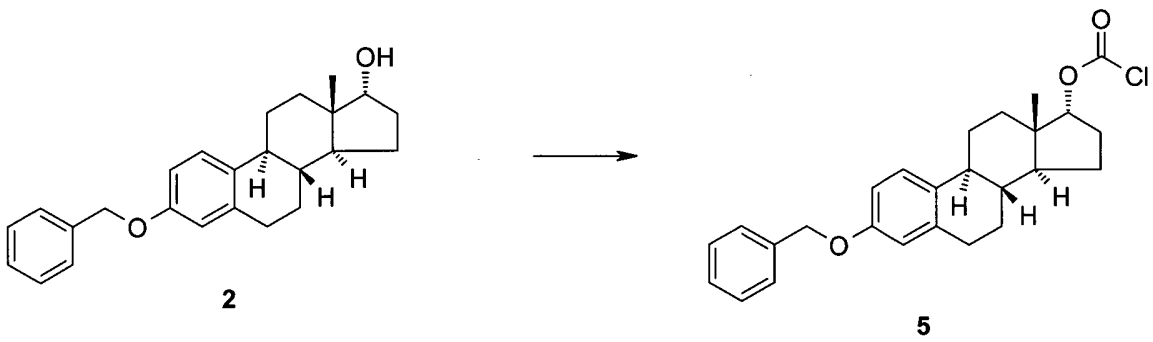
The pharmaceutical preparation mostly contains one unit the dose. The typical daily dose for an adult is of 0.2-300 µg/kg, preferably 0,5-100 µg/kg, most preferably 5-50 µg/kg. The daily dose can be administered in one or more portions. The concrete dose is depending on numerous factors and it is determined by the physician.

The pharmaceutical preparation is made by mixing the active agent with one or more carriers and by forming this mixture into the pharmaceutical preparation in a known manner. The applicable methods are known from the scientific literature such as the above mentioned handbook of Remington's Pharmaceutical Sciences.

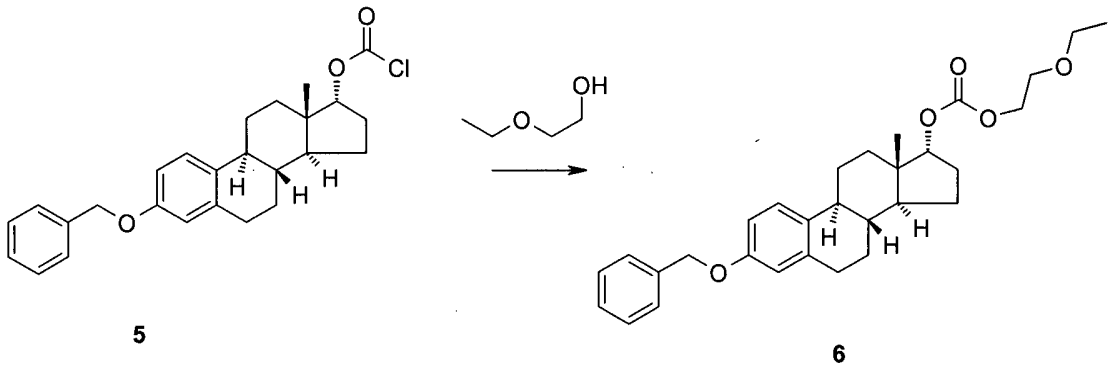
We are describing more details of our invention through the following examples without restricting it for the examples.

In case of contradiction between the chemical name and the structural formula described in the present description, the structural formula is to be taken into account.

Example 1.:
Synthesis of 2-ethoxyetyl (1S,10R,11S,14R,15S)-5-hydroxy-15-methyltetracyclo[8.7.0.0^{2,7}.0^{11,15}] heptadeca-2,4,6-triene-14-yl carbonate

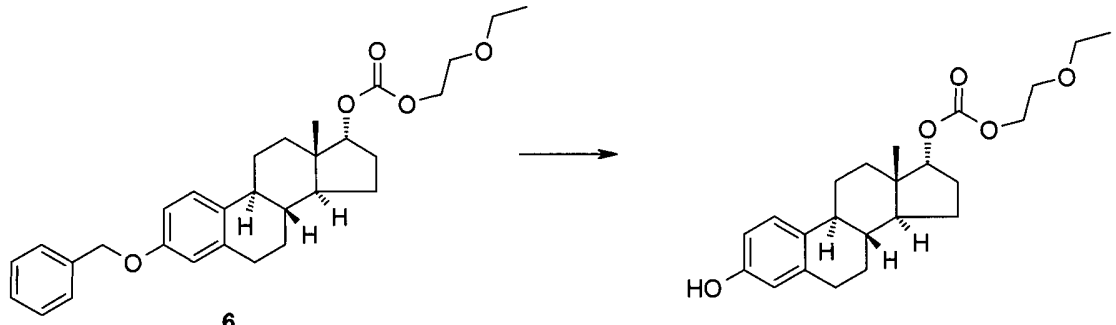


1.5 g (5.0 mmol) of triphosgene is dissolved in 15 ml of tetrahydrofuran and 10 mg of active carbon is added to the solution. 3 g (8.3 mmol)of compound 2 is dissolved in 20 ml of tetrahydrofuran at 0 °C and was added dropwise into the mixture and is stirred for 3 hours at room temperature. The reaction mixture is filtered and the filtrate is evaporated. Product: 4 g brown oil. We use it for the next step without purification.



3.2 g (7.5 mmol)of compound 5 is dissolved in 30 ml of dry dichloromethane and 0.72 ml (7.5 mmol) of cellosolve is added to it. The resulting solution is cooled down to 0 °C and 0.66 ml (8.2 mmol) of pyridine dissolved in 5 ml of dichloromethane was added dropwise into the reaction mixture. After dropping it in, it is stirred for 3 hours at room temperature. The reaction mixture is washed with 10 ml of 1 N hydrochloric acid and 10 ml of NaCl saturated saline,

and dried it over Na2SO4. After evaporation the resulting crude product is column chromatographed in hexane/ethylacetate = 3/1 eluent. Product: 1 g yellow oil.

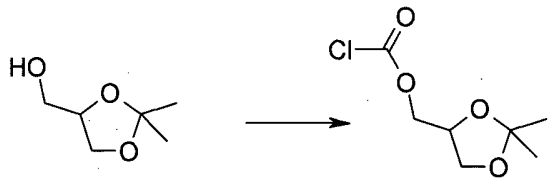


6 7

1 g (2.1 mmol) of compound 6 is hydrogenated in 12 ml of methanol using 0.1 g Pd/C catalyst under 4 bar pressure for 4 hours. We filter the catalyst off and evaporate the filtrate. Product: 810 mg yellow oil. ESI-MS: [M+H]+=389 NMR: NM03850

Example 2.:

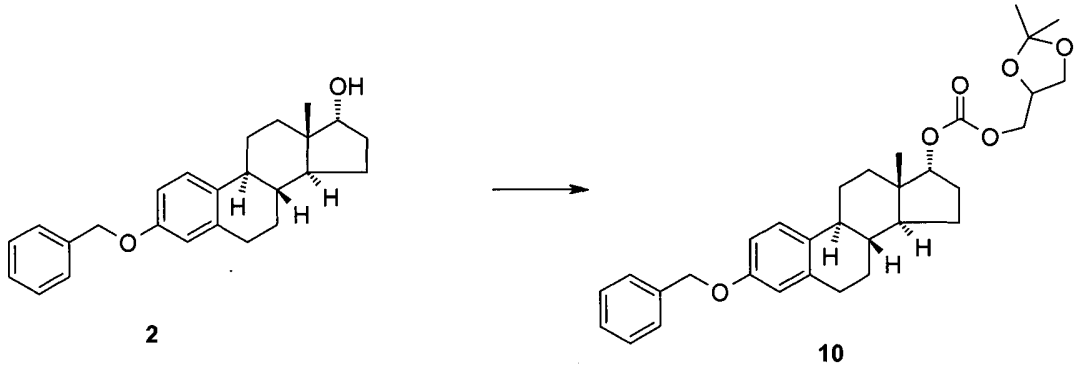
Synthesis of 3-[({[(1S,10R,11S,14R,15S)-5-hydroxy-15-methyltetracyclo[8.7.0.0^{2,7}.0^{11,15}]heptadeca-2,4,6-triene-14-yl]oxy}carbonyl)oxy]propane-1,2-diol



8 9

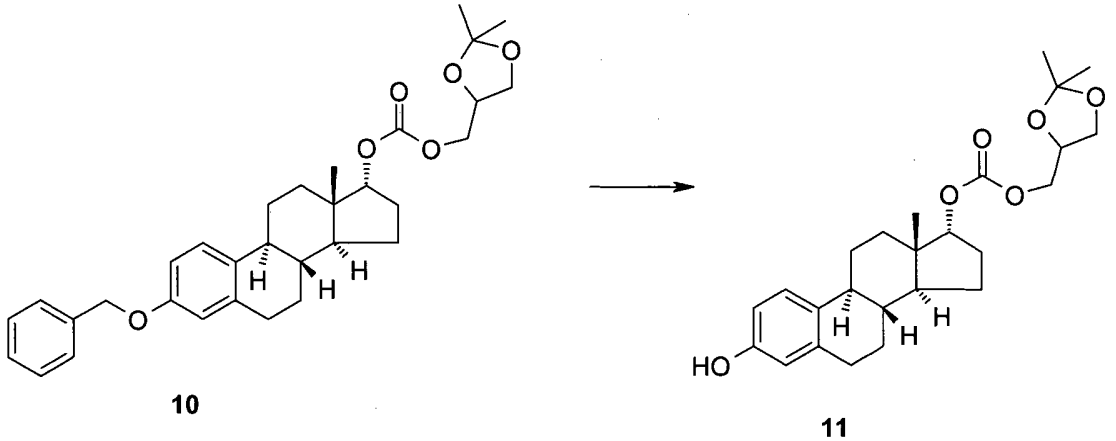
6.7 g (22.7 mmol) of triphosgene is dissolved in 60 ml of tetrahydrofuran and 60 mg active carbon is added to the solution. 5 g (37.8 mmol) of solketal (8) dissolved in 40 ml tetrahydrofuran is added dropwise to the mixture at 0°C and is mixed for 3 hours at room temperature. The reaction

mixture is filtered and the filtrate is evaporated. Product: 8.2 g transparent oil. We use it for the next step without purification.



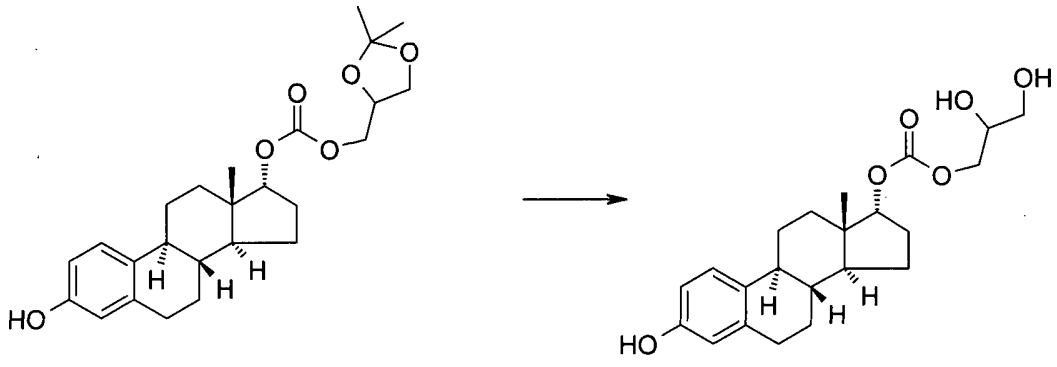
2 10

1.7 g (8.6 mmol) of compound 9 is dissolved in 30 ml dry dichloromethane and 3.1 g (8.6 mmol) of compound 2 is added to it. The resulting solution is cooled down to 0 °C and 0.72 ml (9.2 mmol) of pyridine dissolved in 5 ml of dichloromethane is added dropwise to the reaction mixture. After dropping it in, it is stirred for 3 hours at room temperature. The reaction mixture is washed with 10 ml of 1 N hydrochloric acid and 10 ml of NaCl saturated saline, dried over Na2SO4. After evaporation the resulting crude product was column chromatographed in hexane/ ethylacetate = 3/1 eluent. Product: 1.5 g. NMR: NM03803 ESI-MS:031119



10 11

1.5 g (2.9 mmol) of compound 10 is hydrogenated in 15 ml of methanol with 150 mg Pd/C catalyst under 4 bar pressure for 5 hours. The catalyst is filtered off and the filtrate is evaporated. Product: 1 g white powder.

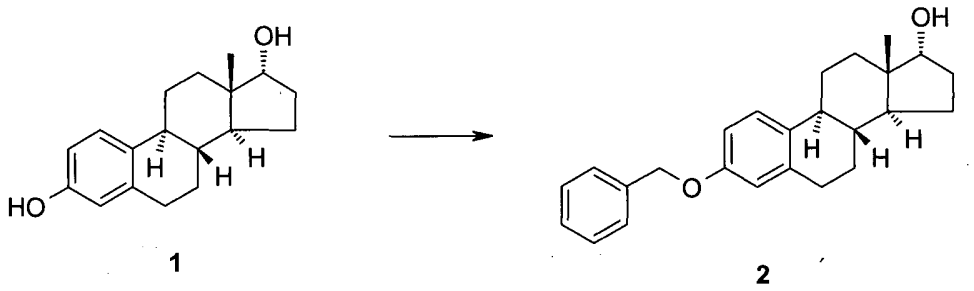


11 12

1 g (2.3 mmol) of compound 11 is dissolved in 10 ml of tetrahydrofuran and 1 ml of 2 N hydrochloric acid is added to it. The resulting reaction mixture is stirred for 5 hours at room temperature. After the reaction completed the reaction mixture is distilled to its half, 30 ml of dichloromethane is added to it and we wash it with 2×10 ml of water and 1×10 ml with NaCl saturated saline. The organic phase is dried over Na_2SO_4 and then evaporated. Product: 580 mg white powder. ESI-MS: [M+H]+=391

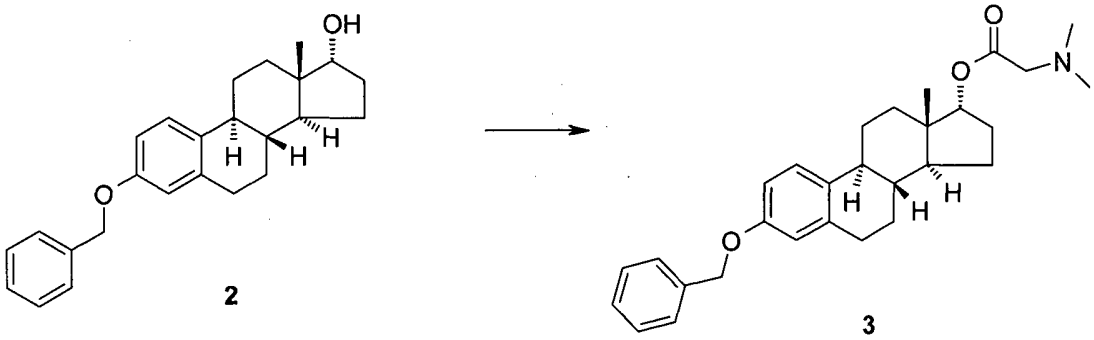
Example 3.:

Synthesis of (1S,10R,11S,14R,15S)-5-hydroxy-15-methyltetracyclo[8.7.0.0^{2,7}.0^{11,15}]heptadeca-2,4,6-triene-14-yl 2-(dimethylamino)acetate



1 2

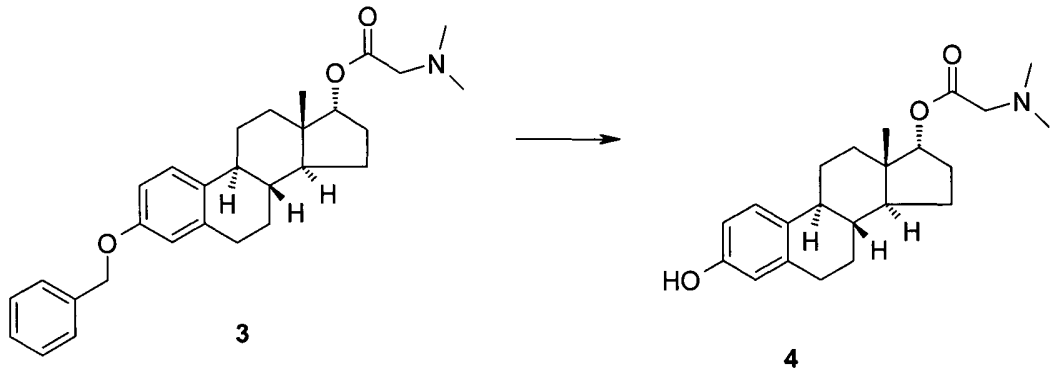
10 g (39 mmol) of 17-alpha-estradiol (1) is dissolved in 200 ml of dry acetone. 20 g (78 mmol) of potassium carbonate is added to the resulting solution and then carefully drop 11.4 g (66 mmol) of benzyl bromide at 0-10 °C to it. The resulting suspension is stirred for 2 hours at reflux temperature. The suspension is filtered, washed with acetone and the filtrate is evaporated. The resulting crude product is crystallized from hexane. Product: 11.9 g off-white, solid substance.



2 3

2 g (5.6 mmol) of benzyl protected 17-alpha-estradiol (**2**) is dissolved in 30 ml of ethyl acetate and 1.2 g (8.4 mmol) N,N-dimethylglycine hydrochloride, 0.8 ml (8.4 mmol) triethylamine, 1.6 g (8.4 mmol) ethyldicyclocarbodiimide and 300 mg dimethyl-aminopyridine are added. The reaction mixture is stirred overnight at room temperature. The reaction mixture is washed with 3×40 ml of water and 1×40 ml with NaCl saturated saline, the organic phase is dried over Na_2SO_4 and then evaporated. The crude product is purified by

column chromatography using hexane/ethyl acetate = 2/1 eluent. Product: 560 mg, white powder.



3 4

560 mg (1.25 mmol) compound **3** is dissolved in 16 ml dichloromethane/isopropanol mixture and hydrogenated using 56 mg Pd/C catalyst under 4 bar pressure for 4 hours. The catalyst is filtered off and the filtrate is evaporateed. Product: 455 mg white powder. ESI-MS: $[M+H]^+$=358 NMR: NM03905

Example 4.:
The fast antidepressant effect of the compound of example 3.

During our experiment we tested the compound of example 3 in the so called „learned helplessness" animal model. The „learned helplessness" is said to be the most reliable model of depression and antidepressant response (Willner P: Animal models of depression: an overview. Pharmacol Ther 45:425-455, 1990; Cryan JF, Markou A, Lucki I: Assessing antidepressant activity in rodents: recent developments and future needs. Trends Pharmacol Sci 23:238-245, 2002; O'Neil MF, Moore NA: Animal models of depression: are there any? Hum Psychopharmacol 18:239-254, 2003). The „learned helplessness" test performs outstandingly regarding all the three criteria of the disease models. It is excellent at

„face validity", that is, the model reproduces the symptoms of depression trustworthily. It is excellent at „predictive validity", that is, the model identifies agents with antidepressant effect precisely. It is also good at „construct validity", that is, the model is suitable for studying the pathomechanism of depression.(Willner P: Animal models of depression: an overview. Pharmacol Ther 45:425-455, 1990; Cryan JF, Markou A, Lucki I: Assessing antidepressant activity in rodents: recent developments and future needs. Trends Pharmacol Sci 23:238-245, 2002). Regarding our experiment, it is of vital importance, that the „learned helplessness" is one of the few animal models, which are able to reproduce the delayed effect of the monoaminergic antidepressants (Hajszan T, Dow A, Warner-Schmidt JL, Szigeti-Buck K, Sallam NL, Parducz A, Leranth C, Duman RS: Remodeling of hippocampal spine synapses in the rat learned helplessness model of depression. Biol Psychiatry 65:392-400, 2009), making it possible to filter for antidepressants with fast effect.

We used the „learned helplessness" protocol during our experiments, which is routinely applied by our laboratory, as described it in our previous studies (Hajszan T, Dow A, Warner-Schmidt JL, Szigeti-Buck K, Sallam NL, Parducz A, Leranth C, Duman RS: Remodeling of hippocampal spine synapses in the rat learned helplessness model of depression. Biol Psychiatry 65:392-400, 2009; Hajszan T, Szigeti-Buck K, Sallam NL, Bober J, Parducz A, Maclusky NJ, Leranth C, Duman RS: Effects of estradiol on learned helplessness and associated remodeling of hippocampal spine synapses in female rats. Biol Psychiatry 67:168-174, 2010). The essence of the „learned helplessness" animal model is that the animals are put under very intense stress, from which it is impossible to escape (inescapable stress). The

inescapable stress greatly diminishes the animals' ability to acquire escape reactions in similar situations in the future, even though the stress is already escapable (escapable stress). This escape/ learning deficit evolves in an acute way within 24 hours as a result of the inescapable stress and without treatment it can last for weeks (Hajszan T, Dow A, Warner-Schmidt JL, Szigeti-Buck K, Sallam NL, Parducz A, Leranth C, Duman RS: Remodeling of hippocampal spine synapses in the rat learned helplessness model of depression. Biol Psychiatry 65:392-400, 2009).

The „learned helplesness" experiments were performed in the so called shuttle box conditioning system.(Med Associates, St. Albans, VT, USA). The shuttle boxes of our laboratory are specifically equipped cages sized for rats, made of transparent plexiglass, which are divided into two equal parts with a central traverse. There is a computer-controlled, built-in guillotine door in the traverse, which if open, the rat can freely move between the two sides of the cage. The stress effect is caused by an electric shock given to the animals' leg (footshock), that is why the flooring of the shuttle box cages are made of metal grid. The metal grid is made of 5 mm diameter non-corrosive steel rods, which are placed 1.7 cm apart from each other. A shock-generator provides the electric shocks through the steel rods. We always make the experiments in a dark or dim room between 10:00- 16:00 h.

We used male 200-250 gram SD rats (n = 14) for our experiment, which were coming from the first generation of the SD rat colony maintained by the Biological Research Centre of the Hungarian Academy of Sciences in Szeged. We kept the rats in 12/12 h illumination cycle (light phase from 07:00 to 19:00 h) and we provided constantly available

water and rodent food for them. We divided the animals into two groups: (1) „control" group (n = 7)and (2) group treated with the compound of example 3 (n = 7).

On the first day of the experiment the animals got the inescapable stress conditioning, which was carried out in one side of the shuttle box cages. The inescapable stress conditioning consisted of 60 footshocks, at 0.85 mA amperage, as well as of a footshock period randomized by a computer and an inter-shock interval (on the average 15 s footshock period and 45 s inter-shock interval). The guillotine door dividing the two sides was closed during the whole length of the conditioning. During our laboratory's previous experiments these parameters were proved to be adequate for effectively provoking the pathological helpless behavioral form (Hajszan T, Dow A, Warner-Schmidt JL, Szigeti-Buck K, Sallam NL, Parducz A, Leranth C, Duman RS: Remodeling of hippocampal spine synapses in the rat learned helplessness model of depression. Biol Psychiatry 65:392-400, 2009; Hajszan T, Szigeti-Buck K, Sallam NL, Bober J, Parducz A, Maclusky NJ, Leranth C, Duman RS: Effects of estradiol on learned helplessness and associated remodeling of hippocampal spine synapses in female rats. Biol Psychiatry 67:168-174, 2010).

After one week, on the 8th day of the experiment the rats received treatments according to the two kinds of groups.
(1) The animals of the „control" group received 1 ml/kg vehiculum [10% DMSO (Sigma-Aldrich, St. Louis, MO, USA)and 10% Solutol (BASF) dissolved in distilled water] injected intraperitoneally .
(2) The members of the group treated with the compound of example 3 received 500 µg/kg of the compound of example 3 injected intraperitioneally in a 1 ml/kg volume vehiculum.

One hour after the injection we examined the severity of the rats' depressive behavior with the active escape test. The active escape test consisted of 30 footshocks at 0.65 mA amperage with a footshock-length of maximum 35 s and a 60 s average interval between the trials. Simultaneously, with the beginning of the footshock the guillotine door was opened and the animal could escape from the stress effect by running to the other side of the shuttle box cage (shuttle crossing). During the first five trials the rat had to accomplish one shuttle crossing for the escape, while in the rest (6-30 trials) the requirement was two shuttle crossings. During the trials the time needed for a successful escape was measured and recorded by the control computer (escape latency). In case the animal did not provide a successful escape reaction within 35 s, the footshock was terminated, the computer recorded the escape latency at 35 s and entered the trial as an escape error. (Hajszan T, Dow A, Warner-Schmidt JL, Szigeti-Buck K, Sallam NL, Parducz A, Leranth C, Duman RS: Remodeling of hippocampal spine synapses in the rat learned helplessness model of depression. Biol Psychiatry 65:392-400, 2009; Hajszan T, Szigeti-Buck K, Sallam NL, Bober J, Parducz A, Maclusky NJ, Leranth C, Duman RS: Effects of estradiol on learned helplessness and associated remodeling of hippocampal spine synapses in female rats. Biol Psychiatry 67:168-174, 2010).

We averaged the escape latency rates coming from different animals by fives (trials 1-5, 6-10, 11-15, ... 26-30), then we calculated group averages from them. We illustrated the rates obtained in form of average ± standard error (SEM) on an escape latency diagram (Figure 1). We also calculated group averages from the escape latency averages of the total active escape experiments (trials 1-30) and the number of

escape errors, which were illustrated on diagrams in form of average ± SEM (Figure 1, MLa and MH graphs). First, we examined the escape latency rates with the two-way repeated measures ANOVA (group × trial number) test, then we compared the data of the two groups with the Tukey post-hoc test. We compared the escape error results of the two groups by means of a t-test. To determine the significance we took the $p < 0.05$ value as a basis.

The escape latency graph according to Figure 1 shows, that the rats treated with the compound of example 3 every time accomplished their escape task within a shorter time than the animals belonging to the „control" group. The two-way repeated measures ANOVA test found a significant change in case of both the „ group" factor ($F_{1,12} = 14.378$, $p = 0.003$) and the „trial number" factor ($F_{5,60} = 5.529$, $p < 0.001$), however, the group x trial number interaction was not significant ($F_{5,60} = 0.824$, $p = 0.537$). This analysis proves, that both the treatment with the compound of example 3 and the progress of time significantly changed the escape latency of the animals, however, the progress of time did not influence the effect of the treatment. Our measurement results show, that the average escape latency of the rats treated with the compound of example 3 was 8.354 seconds lower compared to the "control" group (Figure 1, MLa graph), which is a 26.6% improvement on the average ("control" group: 31.455 ± 0.800 s escape latency; group treated with the compound of example 3: 23.101 ± 2.050 s escape latency). According to the conservative Tukey post-hoc test, the occurred decline in the average escape latency of the animals treated with the compound of example 3 was significant ($p = 0.003$). The number of escape errors made by rats treated with the compound of example 3 were on the average 10.714 lower compared to the "control" group (Figure

1., MH graph), which is a 4.44% improvement on the average ("control" group: 24.143 ± 1.534 escape error; group treated with the compound of example 3: 13.429 ± 2.590 escape error). According to the t-test, the decline in the number of escape errors occurred as a result of the treatment with the compound of example 3 was significant ($p = 0.004$).

According to the analysis of our results we can draw the conclusions, that the treatment with the compound of example 3 in the animal model of learned helplessness of depression significantly reduces the escape latency and the number of escape errors of rats, that is, the compound of example 3 has an antidepressant effect. During the previous experiments of our laboratory, in case of the use of the tricyclic antidepressant desipramine a six day treatment was necessary to achieve antidepressant effect in the rat learned helplessness model. (Hajszan T, Dow A, Warner-Schmidt JL, Szigeti-Buck K, Sallam NL, Parducz A, Leranth C, Duman RS: Remodeling of hippocampal spine synapses in the rat learned helplessness model of depression. Biol Psychiatry 65:392-400, 2009). Contrarily, in our present experiment the treatment with the compound of example 3 achieved antidepressant effect in one hour, that is, the compound of example 3 is a fast-acting antidepressant.

The meanings of the abbreviations of Figure 1 are the following:

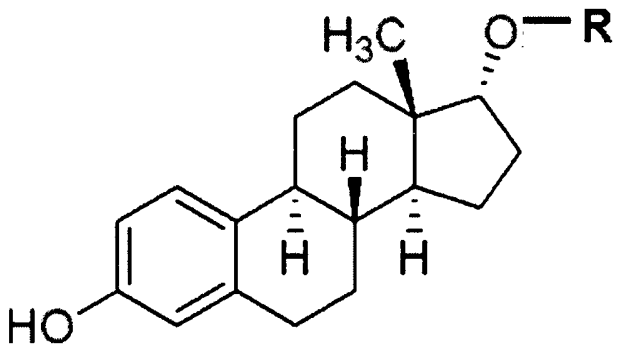
- meaning of the asterix: $p < 0.01$;
- vehicle = control, vehiculum (vehicle without active agent) group;
- compound no 3 according to the invention = animals treated with 500 µg/kg solution of compound of example 3;
- meaning of MLa: escape latency;

- meaning of MH: escape error.

The graphs of Figure 1 show, that the treatment with the compound of example 3 in the animal model of learned helplessness significantly reduces the escape latency and the number of escape errors of rats, that is, the compound of example 3 has an antidepressant effect.

Claims

1. Process for the preparation of pharmaceutical products, characterized in that one or more derivatives of the 17-alpha-estradiol of formula (I) produced in a known manner



(I),

where

R means

- ethyl, higher alkyl, cycloalkyl, aryl and heterocycle as well as hydroxyl, alkyl groups containing amino and carboxy groups; or
- CO-O-R', where R' means alkyl, cycloalkyl, aryl and heterocycle; or
- CO-(R")-O-Q, where R" means alkyl, Q means alkyl, cycloalkyl, aryl and heterocycle.

are mixed with inert pharmaceutical carriers and/or excipients and form them into pharmaceutical preparations that are capable of treating and/or preventing psychiatric diseases.

2. Process according to claim 1, characterized in that pharmaceutical preparations with antidepressant effect are prepared.

3. Process according to claim 2, characterized in that fast-acting pharmaceutical preparations with antidepressant effect are prepared.

4. Process according to claim 2 and 3, characterized in that pharmaceutical preparations that are capable of treating post-partum depression and/or peri-menopausal depression are produced.

5. Process according to claim 1, characterized in that pharmaceutical preparations that are capable of treating anxiety, schizophrenia, memory disturbance associated with depression, bipolar depression, Alzheimer-disease and post-traumatic stress disease are produced.

6. Use of one or more derivatives of the 17-alpha-estradiol of formula (I) for the treatment and/or prevention of psychiatric diseases.

7. Use according to claim 6 for the treatment of depression.

8. Use according to claim 7 as a fast-acting antidepressant.

9. Use according to claim 7 and 8 for the treatment of post-partum depression or peri-menopausal depression.

10. Use according to claim 6 for the treatment of anxiety, schizophrenia, memory disturbance associated with depression, bipolar depression, Alzheimer-disease or post-traumatic stress disease.

11. Pharmaceutical preparation capable of the treatment and/or prevention of psychiatric diseases, which contains one or more derivatives of 17-alpha-estradiol of formula (I)

as the active agent and inert solid or liquid pharmaceutically acceptable carriers and/or excipients.

12. Pharmaceutical preparation according to claim 11 with antidepressant effect.

13. Pharmaceutical preparation according to claim 12 with fast-acting antidepressant effect.

14. Pharmaceutical preparation according to claims 12 and 13 with post-partum antidepressant or peri-menopausal antidepressant effect.

15. Pharmaceutical preparation according to claim 11 that is capable of the treatment and/or prevention of anxiety, schizophrenia, memory disturbance associated with depression, bipolar depression, Alzheimer-disease or post-traumatic stress disease.

16. Process for the treatment and/or prevention of psychiatric diseases, especially depression, characterized in that a pharmaceutically suitable amount of pharmaceutical preparation containing one or more derivatives of 17-alpha-estradiol of formula (I) is administered for the patients in need of treatment.

17. Process according to claim 16, characterized in that the derivative of 17-alpha-estradiol of formula (I) is used in an amount of 0.2-300 µg/kg, preferably 0.5-100 µg/kg, more preferably 5-50 µg/kg.

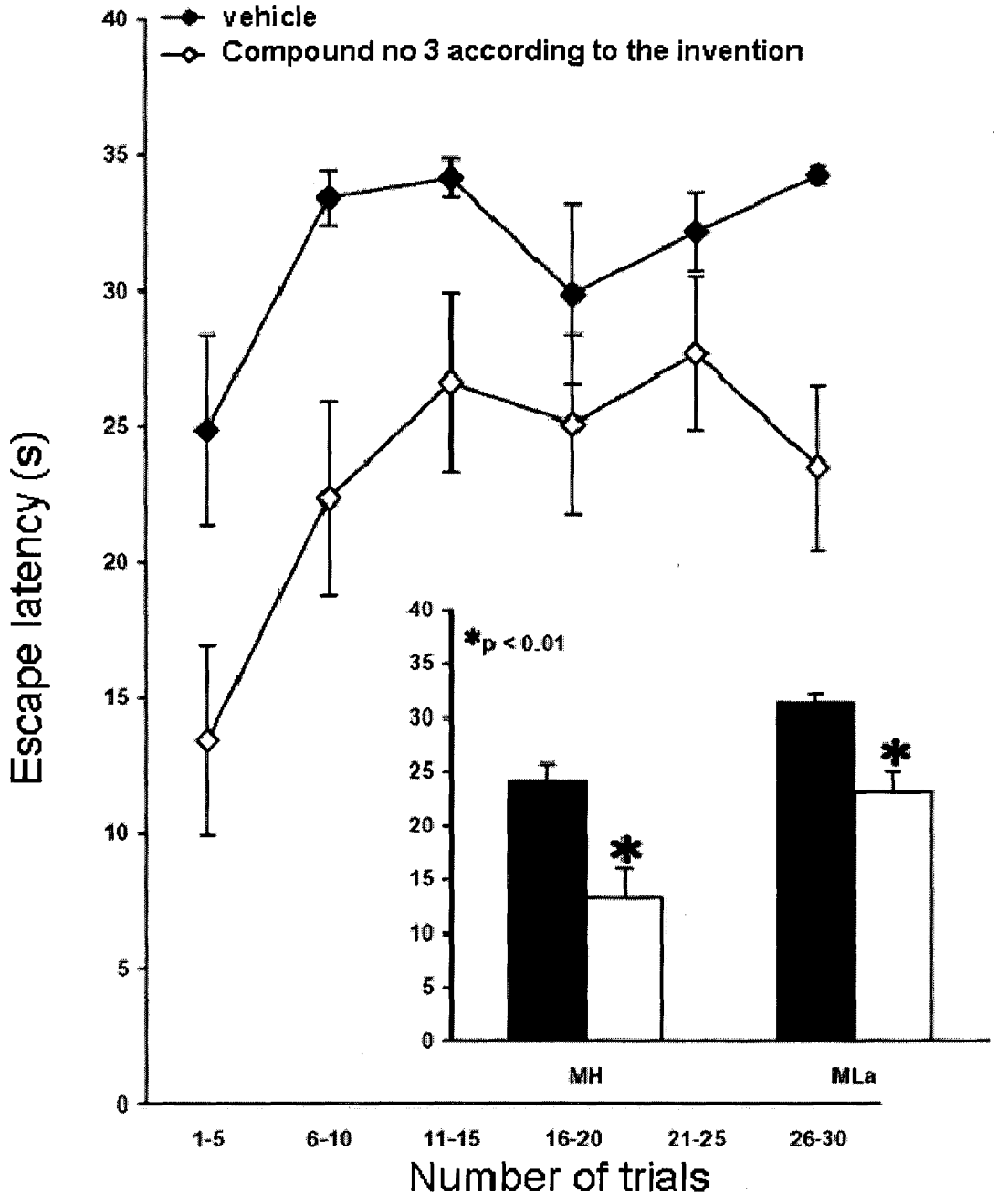

vehicle
Compound no 3 according to the invention
Escape latency (s)
Number of trials
1-5
6-10
11-15
16-20
21-25
26-30
*p < 0.01
MH
MLa

Figure 1.

INTERNATIONAL SEARCH REPORT

International application No
PCT/HU2012/000141

A. CLASSIFICATION OF SUBJECT MATTER
INV. A61K31/565 A61P25/24
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)
A61K

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, WPI Data, BIOSIS, EMBASE

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 02/00619 A2 (UNIV FLORIDA [US]) 3 January 2002 (2002-01-03) figure 1 page 6, line 11 - line 12 page 7, line 21 page 9, line 25 - line 30 -----	1,5,6, 10,11, 15-17
A	WO 03/074058 A1 (CHIESI FARMA SPA [IT]; DAVIES REBECCA JAINE [IT]; GANDERTON DAVID [IT]) 12 September 2003 (2003-09-12) claims 1-3 -----	1-17

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

☒ See patent family annex.

* Special categories of cited documents :

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

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

"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	Date of mailing of the international search report
12 March 2013	21/03/2013

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

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INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/HU2012/000141

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 0200619 A2	03-01-2002	AU 7725801 A	08-01-2002
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		US 2002035100 A1	21-03-2002
		WO 0200619 A2	03-01-2002
WO 03074058 A1	12-09-2003	AT 381339 T	15-01-2008
		AU 2003215588 A1	16-09-2003
		BR 0303347 A	13-07-2004
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		EP 1340503 A1	03-09-2003
		EP 1480651 A1	01-12-2004
		ES 2297183 T3	01-05-2008
		NO 20034873 A	23-12-2003
		US 2005209207 A1	22-09-2005
		WO 03074058 A1	12-09-2003