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**SOLICITUD DE REGISTRO DE:**

1 ☒ **Patente de Invención** ☐ **Patente Modelo de Utilidad.**  
☐ **Capítulo I** ☒ **Capítulo II**

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Cual \_\_\_\_\_  
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6 **Título (54) MÉTODO MEJORADO PARA PREPARAR EXTRACTOS DE GINKGO QUE TIENEN UN CONTENIDO BAJO DE 4"-O-METIL PIRIDOXINA Y/O BIFLAVONAS**

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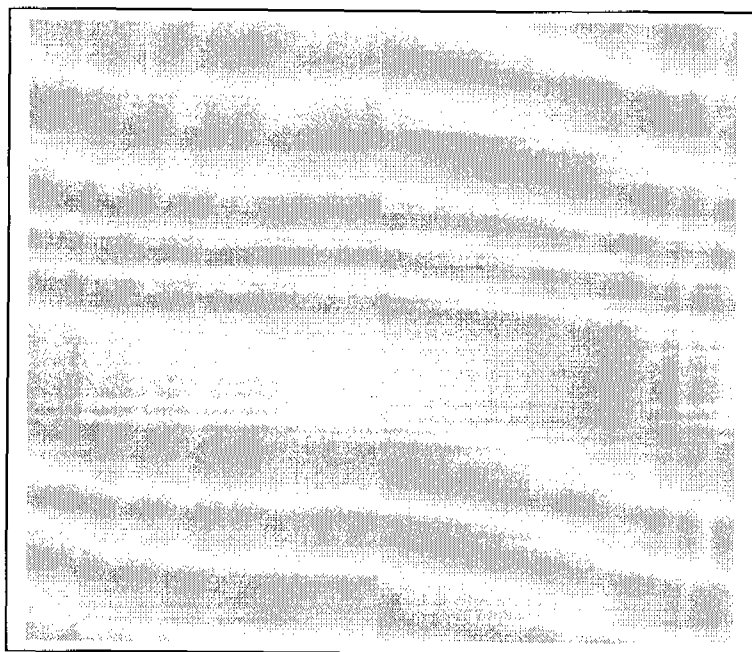
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- ☒ Comprobante de pago de la tasa de presentación de la solicitud
- ☐ Documento que acredita la existencia y representación legal cuando el solicitante sea persona jurídica
- ☐ Poderes si fuese el caso
- ☐ Documento de cesión del inventor al solicitante o a su causante
- ☐ Declaraciones
- ☒ Copia de la solicitud internacional (Resumen, descripción, reivindicaciones, dibujos)
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- ☐ Copia del examen preliminar internacional efectuado por la Autoridad Internacional de examen preliminar (IPER)
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- ☐ Arte final 12x12.
- ☐ De ser el caso, información sobre otras solicitudes de patente o títulos obtenidos en el extranjero por el mismo titular o su causante, relacionadas parcial o totalmente con la invención de esta solicitud.
- ☐ De ser el caso, modificaciones efectuadas a la solicitud internacional
- ☒ Otros: Informe de búsqueda
- ☒ Formulario modificación solicitante
- ☒ Request IB/304, Written Opinion

## 12 FIGURA CARACTERISTICA



13 Solicito la concesión de la patente,

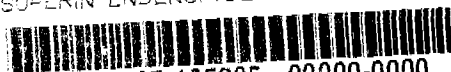
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2

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(54) Title: IMPROVED METHOD FOR PREPARING GINKGO EXTRACTS HAVING A LOW CONTENT OF 4'-O-METHYL PYRIDOXINE AND/OR BIFLAVONES

(57) Abstract: The present invention relates to an improved multi-step method for preparing an extract from ginkgo biloba having a reduced content of 4'-O-methyl pyridoxine and/or biflavones, wherein the depletion is carried out by filtration over an adsorber resin and/or an ion exchanger and the substances to be removed are retained on the resin. The invention further relates to an extract from ginkgo biloba having a reduced content of 4'-O-methyl pyridoxine and/or biflavones, which is obtainable by the method according to the present invention, as well as to its use.

WO 2006/117169 A1

**Improved method for preparing ginkgo extracts having a low  
content of 4'-O-methyl pyridoxine and/or biflavones**

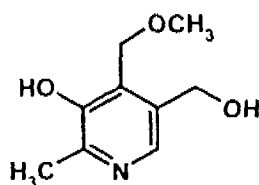
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The present invention relates to an improved multi-step method for preparing an extract from ginkgo biloba having a reduced content of 4'-O-methyl pyridoxine and/or biflavones, wherein the depletion is effected by filtration over an adsorber resin and/or an ion exchanger and the substances to be removed are retained on  
10 the resin. The invention further relates to an extract from ginkgo biloba having a reduced content of 4'-O-methyl pyridoxine and/or biflavones which is obtainable by the method according to the present invention, as well as to its use.

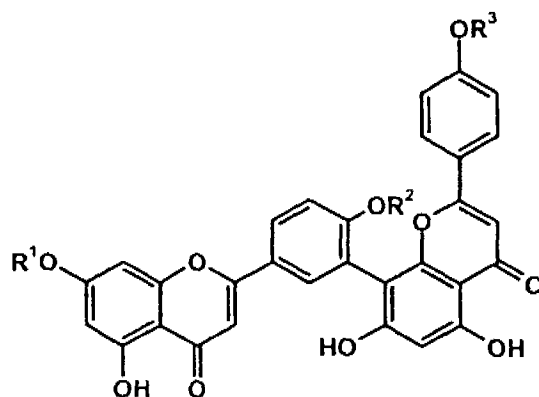
Since decades, extracts from the leaves of ginkgo biloba are used as a medica-  
15 ment. They are currently used for the treatment of different kinds of dementia and symptoms thereof as well as cerebral and peripheral blood circulation disorders. Ingredients, the efficacy is associated with, are terpene lactones (ginkgolides A, B, C and bilobalide) as well as glycosides of flavones (quercetin, kaempferol and isorhamnetin). The leaves of ginkgo biloba also contain components which do not  
20 contribute to the desired efficacy, but which may be responsible for risks and side effects. In addition to unpolar plant ingredients such as ginkgolic acids, those components are 4'-O-methyl pyridoxine and biflavones. In a ginkgo extract, which is efficacious and at the same time as safe as possible and as low in side effects as possible, these compounds should thus not be present to the largest possible  
25 extent.

4'-O-methyl pyridoxine may cause symptoms of poisoning such as convulsive sei-  
zures and unconsciousness. Thus, this compound is also referred to as ginkgo-  
toxin. The biflavones contained in ginkgo exhibit an immunotoxic potential and  
30 may elicit contact allergies. These biflavones contained in ginkgo are predomi-  
nantly the compounds amentoflavone, bilobetin, ginkgetin, isoginkgetin and  
sciadopitysin.

4'-O-Methyl pyridoxine:



5 Biflavones:



|                |                              |
|----------------|------------------------------|
| Amentoflavone: | $R^1 = R^2 = R^3 = H$        |
| Bilobetin:     | $R^1 = R^3 = H, R^2 = OCH_3$ |
| Ginkgetin:     | $R^3 = H, R^1 = R^2 = OCH_3$ |
| Isoginkgetin:  | $R^1 = H, R^2 = R^3 = OCH_3$ |
| Sciadopitysin: | $R^1 = R^2 = R^3 = OCH_3$    |

15 Methods for depleting 4'-O-methyl pyridoxine and simultaneously biflavones as well as the extracts obtained are already described in EP 1 037 646 B1. Therein, 4'-O-methyl pyridoxine is retained using an acidic cation exchanger and the biflavones are adsorbed on activated carbon. Preferably these depletion steps are carried out after step f) of the method according to EP 1 037 646 B1 (page 3), i.e., after "treating the solution with a lead compound or an insoluble polyamide". The method of EP 1 037 646 B1 is based on the method originally described in  
20 DE 39 40 091 C2.

In EP 1 037 646 B1 the depletion of the biflavones and 4'-O-methyl pyridoxine is carried out at a stage of the method (step f)), wherein relatively high contents of lead salts and ammonium salts are present such that an increased amount of ion exchanger has to be employed.

5

Furthermore, the depletions of biflavones and 4'-O-methyl pyridoxine are described and claimed in combination only. However, depending on the limits to be possibly established in the future and in view of the requirement of as low a modification of the extract composition as possible, it may be desirable to remove the  
10 biflavones or 4'-O-methyl pyridoxine only.

Furthermore, the contents of less than 50 ppm of 4'-O-methyl pyridoxine and less than 100 ppm of biflavones which are aspired in EP 1 037 646 B1, are still comparatively high. Since EP 1 037 646 B1 does not describe a comparative example,  
15 also the actual degree of depletion achieved in Examples 2 and 3 cannot be seen from this reference.

Thus, it is the object underlying the present invention to provide a method for preparing ginkgo extracts having a low content of 4'-O-methyl pyridoxine and/or a low  
20 content of bioflavones which does not have the disadvantages indicated above and, in particular,

- wherein well recyclable and/or very inexpensive adsorbents are employed,
- which leads to contents of 4'-O-methyl pyridoxine of less than 20 ppm, preferably less than 10 ppm and particularly preferred less than 2 ppm and/or  
25 contents of biflavones of less than 20 ppm, preferably less than 10 ppm and particularly preferred less than 5 ppm and
- which leads to extracts having preferred contents of flavonoides of 20 to 30% by weight (particularly preferred 22.0 to 27.0%), of terpene lactones of 4.5 to 8.5% by weight (particularly preferred 5.0 to 7.0%, preferably 2.8 to 3.4%  
30 ginkgolides A, B and C and 2.6 to 3.2% bilobalid) and of ginkgolic acids (5 ppm at the most), wherein said contents of flavonoids of 22.0 to 27.0%, of terpene lactones of 5.0 to 7.0% (2.8 to 3.4% thereof ginkgolides A, B and C

and 2.6 to 3.2% thereof bilobalid) and of ginkgolic acids (5 ppm at the most) fulfil the requirements of the German Pharmacopoeia 2003 ("Deutsches Arzneibuch"; DAB).

- 5 According to the DAB the flavonoids are determined in the form of quercetin, kaempferol and isorhamnetin after acidic hydrolysis and calculated as flavonoid glycosides.

This object could be solved by separating the biflavones using adsorber resins  
10 and/or separating 4'-O-methyl pyridoxine via acidic ion exchangers.

In order to produce the extracts according to the present invention, preferably the following method is carried out, wherein steps a) to i) as well as m) are essentially described already in DE 39 40 091 C2, the total disclosure content of which, in  
15 particular with respect to the details of the method, shall be incorporated into the present application by reference:

- a) extracting the leaves of ginkgo biloba (drug) at a temperature of 20-100°C, preferably 40-60°C, using water-containing acetone having a content of 20-90% by weight, a water-containing alkanol having 1 to 3 carbon atoms (methanol, ethanol,  
20 n-propanol and isopropanol) and a content of 20-90% by weight or anhydrous methanol, wherein the ratio drug : solvent amounts to 1:4 to 1:20, preferably 1:5 to 1:10,
- b) separating the organic solvent by evaporating the solvent to a content of 10% by weight at the most (preferably 5% by weight at the most), wherein water may  
25 be added during the final distillation steps,
- c) diluting the remaining aqueous solution with water to a solids content of 5 to 25% by weight (preferably 15 to 20% by weight), cooling the solution to a temperature below 25°C (preferably about 10 to 12°C) and allowing the remaining aqueous solution to stand until a precipitation is formed,
- 30 d) treating the remaining aqueous solution with ammonium sulfate (preferably to a content of up to 40% by weight, particularly preferred 25 to 35% by weight) and subsequently extracting the solution at least once using methyl ethyl ketone or a

mixture of methyl ethyl ketone and acetone (preferably in a ratio of 9:1 to 4:6, in particular 6:4),

e) concentrating the extract obtained (preferably to a solids content of 50 to 70% by weight) and diluting the extract with a mixture of ethanol and water to obtain a solution containing 50% by weight water and 50% by weight ethanol and having a solids content of 10% by weight,

f) treating the solution with a lead compound (preferably with lead acetate, lead hydroxide acetate, lead nitrate or an aqueous suspension of lead hydroxide) until a change in colour from brown to umber occurs, or an insoluble polyamide (preferably polyamide 6, polyamide 6,6 or crosslinked polyvinylpyrrolidone (polyvidone)),

g) extracting the filtered solution using an aliphatic or cycloaliphatic solvent having a boiling point of 60 to 100°C,

h) concentrating the remaining aqueous-alcoholic solution, subsequently treating the solution with ammonium sulfate and extracting the solution with methyl ethyl ketone and ethanol, (the solution is preferably concentrated under reduced pressure to an ethanol content of about 5% at the most, added with ammonium sulfate to a content of 20% by weight and extracted with the solvent in a ratio of 8:2 to 5:5, preferably 6:4),

i) concentrating the organic phase obtained to a solids content of 50 to 70% by weight,

k) dissolving the concentrate in an aqueous alkanol having 1 to 3 carbon atoms and optionally filtering the product,

l) filtration over an adsorber resin and/or an ion exchanger, wherein the substances to be removed are retained on the resin and

m) drying the concentrate (preferably at a maximum temperature of 60 to 80°C) under reduced pressure to obtain a dry extract having a water content of less than 5%.

It is the advantage of the method according to the present invention that the depletion step is carried out at the end of the method. Thus, a smaller amount of the

ion exchanger has to be employed and particularly low contents of 4'-O-methyl pyridoxine and/or biflavones are obtained.

Preferred adsorber resins in step l) are resins based on optionally substituted styrenes/divinylbenzenes such as Diaion HP-20, HP-21 or Sepabeads SP-207 and SP-850. Preferred ion exchangers are strongly acidic ion exchangers such as Merck I or Amberlite IR-120. The filtration optionally includes the usual rinsing using further solvent. The aqueous alkanol in step k) is aqueous methanol, ethanol, n-propanol or isopropanol having a content of 20-90% by weight, respectively, preferably aqueous ethanol, a concentration of 40 to 60% by weight being particularly preferred.

A subject of the present invention are extracts, in particular dry extracts, which are obtainable by the method according to the present invention and which exhibit the following contents:

- less than 20 ppm, preferably less than 10 ppm and in particular less than 2 ppm 4'-O-methyl pyridoxine and/or
- less than 20 ppm, preferably less than 10 ppm and in particular less than 5 ppm biflavones.

Furthermore, subject of the present invention are extracts, in particular dry extracts, which are obtainable by the method according to the present invention and which exhibit the following further contents:

- 20 to 30% by weight, preferably 22.0 to 27.0% by weight flavonoids,
- 4.5 to 8.5% by weight, preferably 5.0 to 7.0% by weight terpene lactones and
- less than 10 ppm, preferably not more than 5 ppm ginkgolic acids.

A further subject of the present invention are extracts, in particular dry extracts, which are obtainable by the method according to the present invention and which exhibit the following contents:

- 20 to 30% by weight, preferably 22.0 to 27.0% by weight flavonoids,
- 2.5 to 4.5% by weight ginkgolides A, B and C in total, preferably 2.8 to 3.4% by weight ginkgolides A, B and C in total,
- 5 • 2.0 to 4.0% by weight, preferably 2.6 to 3.2% by weight bilobalide,
- less than 10 ppm, preferably not more than 5 ppm ginkgolic acids and
- less than 20 ppm, preferably less than 10 ppm, particularly preferred less than 2 ppm of 4'-O-methyl pyridoxine and/or
- less than 20 ppm, preferably less than 10 ppm and particularly preferred less
- 10 than 5 ppm of biflavones.

According to the European Pharmacopoeia, dry extracts generally have a dry residue of at least 95% by weight.

- 15 The extracts according to the present invention can be administered in the form of powders, granules, tablets, dragées (coated tablets) or capsules, preferably orally. In order to prepare tablets, the extract is mixed with suitable pharmaceutically acceptable adjuvants such as lactose, cellulose, silicon dioxide, croscarmellose and magnesium stearate, and pressed into tablets which are optionally provided with a
- 20 suitable coating, for example made of hydroxymethylcellulose, polyethyleneglycol, pigments (such as titanium dioxide, iron oxide) and talcum. The extract according to the present invention can also be filled into capsules, optionally under the addition of adjuvants such as stabilizers, fillers and the like. The dosage is such that 10 to 2000 mg, preferably 50 to 1000 mg and particularly preferred 100 to 500 mg
- 25 extract are administered per day.

A further subject of the present invention are medicaments, food products and other preparations containing these extracts, optionally in combination with other substances such as active ingredients and/or pharmaceutically acceptable adjuvants. The term "food product" as used herein particularly refers to dietetic food

30 products, dietary supplement products as well as medical food and dietary supplements.

## Examples

### Preparation of the extract solution used in Examples 1 and 2 as well as in 5 Comparative Examples 1 and 2

Dried leaves of ginkgo biloba (drug) were extracted at a temperature of about 50°C using seven times their weight (w/w) (i.e. drug-solvent ratio is 1:7) made up of acetone/water 60/40 (w/w) (step a)).

10

The organic solvent was largely separated from the extract (step b)) and the remaining concentrated aqueous solution was diluted with water to a solids content of about 10% by weight. The solution was cooled to a temperature of about 12°C under agitation and the resulting precipitate was removed (step c)).

15

About 30% by weight ammonium sulfate was added to the remaining aqueous solution and the solution formed was extracted using a mixture of methyl ethyl ketone and acetone in a ratio of 6:4 (step d)).

20

The extract obtained was largely concentrated and the concentrate thus obtained was diluted with water and ethanol such that a solution containing 50% by weight water and 50% by weight ethanol at a solids content of about 10% by weight was obtained (step e)). The solution was filtered and the filtrate was added with an aqueous solution of lead hydroxide acetate (step f)).

25

After a further filtration, the remaining aqueous alcoholic solution was extracted with heptane (step g)).

30

Subsequently, the remaining aqueous alcoholic solution was concentrated under reduced pressure to an ethanol content of about 5% and ammonium sulfate was added such that a content of about 20% by weight based on the water content was achieved.

The solution obtained was extracted using a mixture of methyl ethyl ketone and ethanol in a ratio of 6:4 (step h)).

- 5 The resulting organic phase was largely concentrated (solids content of about 55% by weight) (step i)). Then the product was diluted with ethanol and water such that a solution having an ethanol content of about 75% and a solids content of about 15% was obtained (step k)).

10 **Example 1: Ginkgo extract having a reduced content of 4'-O-methyl pyridoxine**

1249 g of the extract solution obtained according to the preparation method described above was adjusted with water to an ethanol content of about 40% (w/w) and applied to 50 ml (corresponding to 0.28 ml/g of the dry extract) Amberlite IR-120 (strongly acidic ion exchanger) (column: about 2 x 16.5 cm; flow: about 8 ml/min) (step l)), followed by rinsing with 200 ml of 50% ethanol (w/w). The resulting solution was concentrated on a rotary evaporator and dried in vacuum at 50°C (step m)): 179.4 g.

20

As can be seen from the following table, a content of 4'-O-methyl pyridoxine of 0.4 ppm was achieved without essentially modifying the contents of ingredients which are relevant with respect to the efficacy.

25 **Comparative Example 1: Ginkgo extract without depletion according to step l) of the present invention**

98.4 g of the extract solution employed in Example 1 were concentrated by evaporation and dried in vacuum at 50°C: 14.7 g.

30

**Example 2a): Ginkgo extract having a reduced content of biflavones**

500 g of the extract solution obtained according to the preparation method described above were adjusted with water to an ethanol content of about 40% (w/w) and applied to 400 ml (corresponding to 5.7 ml/g of the dry extract) Sepabeads SP-850 (adsorber resin) (column: about 4 x 32 cm; flow: about 8 ml/min) (step l)), followed by rinsing with about 2 l of 40% ethanol (w/w) in total. 10% of the resulting solution (259.3 g) were concentrated on a rotary evaporator and freeze-dried (step m)): 6.92 g. The remaining 90% of the solution were employed in Example 2b).

As can be seen from the following table, biflavone contents of less than 1 ppm were achieved, respectively, without essentially modifying the contents of ingredients which are relevant with respect to the efficacy.

**Example 2b): Ginkgo extract having a reduced content of 4'-O-methyl pyridoxine and biflavones**

90% of the solution obtained in Example 2a) (2334.1 g) were applied to 25 ml (corresponding to 0.40 ml/g of the dry extract) Amberlite IR-120 (strongly acidic ion exchanger) (column: about 1.5 x 11 cm; flow: about 10 ml/min) (step l)), followed by rinsing with 100 ml of 40% ethanol (w/w). The resulting solution was concentrated on a rotary evaporator and freeze-dried (step m)): 61.67 g.

As can be seen from the following table, a content of 4'-O-methyl pyridoxine of 0.4 ppm and contents of biflavones of less than 1 ppm were achieved, respectively, without essentially modifying the contents of ingredients that are relevant with respect to the efficacy.

**Comparative Example 2: Ginkgo extract without depletion according to step I) of the present invention**

5 50 g of the extract solution employed in Example 2a) were concentrated by evaporation and freeze-dried: 7.05 g.

**Comparative Example 3: Ginkgo extract with biflavone depletion and 4'-O-methyl pyridoxine depletion after treatment with a lead compound (step f) according to EP 1 037 646 B1**

10

Dried leaves of ginkgo biloba (drug) were extracted at a temperature of about 50°C using seven times their weight (w/w) (i.e. drug-solvent ratio is 1:7) made up of acetone/water 60/40 (w/w).

15 The organic solvent was largely separated from the extract and the remaining concentrated aqueous solution was diluted with water to a solids content of about 10% by weight, cooled to a temperature of about 12°C under agitation and the resulting precipitate was removed.

20 About 30% by weight ammonium sulfate was added to the remaining aqueous solution and the solution formed was extracted using a mixture of methyl ethyl ketone and acetone in a ratio of 6:4.

25 The extract obtained was largely concentrated and the concentrate thus obtained was diluted with water and ethanol such that a solution containing 50% by weight water and 50% by weight ethanol at a solids content of about 10% by weight was obtained. The solution was filtered, the filtrate was added with an aqueous solution of lead hydroxide acetate and filtered again (step f)).

30 150 ml of the solution thus obtained (portion of dry extract: 7.1%; ethanol content: about 50%) were stirred with 2.5 g (corresponding to 0.17 g/g of the final product) active carbon for 30 minutes at room temperature. The filtrate was applied to a

column with 4.2 ml (corresponding to 0.28 ml/g of the final product) strongly acidic cation exchanger (Merck I). The ion exchanger column was washed with 4 x 5 ml EtOH (50%).

- 5 The combined eluates were extracted by shaking using 3 x 70 ml n-hexane. The ethanol/water phase was concentrated on a rotary evaporator and diluted with water to 100 g. The extract solution thus obtained was extracted with 1 x 40 ml, 2 x 30 ml and 1 x 20 ml of a mixture of methyl ethyl ketone:ethanol in a ratio of 3:2, wherein 15 g ammonium sulfate was added to achieve phase separation.

10

The organic phase was stirred for 1 h at room temperature after adding 25 g ammonium sulfate. The undissolved ammonium sulfate as well as the aqueous phase formed were separated. The extract solution was concentrated on a rotary evaporator at 50°C and dried in vacuum at 60°C: 14.8 g.

15

As can be seen from the following table, a 4'-O-methyl pyridoxine content of 52 ppm and a biflavone content of 31.3 ppm and, thus, significantly higher contents and inferior depletion results are achieved compared to Example 2b) according to the present invention.

20

The determination of the contents of flavonoids, terpene lactones and ginkgolic acids listed in Table 1 was carried in accordance with the DAB.

25

The quantitative determination of 4'-O-methyl pyridoxine was performed by means of HPLC and fluorescence detection after enrichment via Sep Pak Plus C18 cartridges, followed by enrichment via Bond Elut LRC-SCX cartridges, wherein a Phenomenex Prodigy 5  $\mu$  ODS-3 HPLC column and a gradient of trifluoroacetic acid (pH 2) : methanol = 9:1 (v/v)  $\rightarrow$  methanol, were employed.

30

The determination of the contents of the biflavones listed in Table 1 was performed by means of HPLC and UV detection at 340 nm via a Waters Nova Pak

C18 column using a gradient of water: acetonitrile of 9:1 (v/v) + 0.3% by volume phosphoric acid (85%) → acetonitrile + 0.3% by volume phosphoric acid.

In each case, calibration was performed using reference substances having a  
5 high, defined purity grade.

Table 1: Composition of the extracts according to examples given above

| Extract according to                | Example 1 | Comparative<br>Example 1 | Example 2a) | Example 2b) | Comparative<br>Example 2 | Comparative<br>Example 3 |
|-------------------------------------|-----------|--------------------------|-------------|-------------|--------------------------|--------------------------|
| Flavonoids [%]                      | 24.86     | 24.51                    | 27.17       | 25.32       | 25.89                    | 22.78                    |
| Ginkgolides A, B, C [%]             | 3.28      | 2.94                     | 2.97        | 3.04        | 2.97                     | 3.40                     |
| Bilobalide [%]                      | 3.17      | 3.03                     | 3.18        | 3.14        | 3.07                     | 3.07                     |
| Terpene lactones [%]                | 6.45      | 5.97                     | 6.15        | 6.18        | 6.04                     | 6.47                     |
| Ginkgolic acids [ppm]               | < 5       | < 5                      | < 5         | < 5         | < 5                      | < 5                      |
| 4'-O-Methyl pyridoxine [ppm]        | 0.4       | 104.5                    | 164         | 0.8         | 89                       | 52                       |
| Amentoflavone [ppm]                 | 68.5      | 78.7                     | n. d. (< 1) | n. d. (< 1) | 81.6                     | 12.6                     |
| Bilobetin [ppm]                     | 123.3     | 120.4                    | n. d. (< 1) | n. d. (< 1) | 136.4                    | 10.4                     |
| Ginkgetin [ppm]                     | 86.3      | 82.1                     | n. d. (< 1) | n. d. (< 1) | 86.1                     | 2.7                      |
| Isoginkgetin [ppm]                  | 103.3     | 104.4                    | n. d. (< 1) | n. d. (< 1) | 111.5                    | 4.8                      |
| Sciadopitysin [ppm]                 | 44.0      | 43.0                     | n. d. (< 1) | n. d. (< 1) | 44.0                     | 0.8                      |
| Total amount of biflavones<br>[ppm] | 425.4     | 428.6                    | n. d. (< 5) | n. d. (< 5) | 459.6                    | 31.3                     |

n. d. = not detectable

**Claims**

1. Method for producing an extract from ginkgo biloba having a reduced content of 4'-O-methyl pyridoxine and/or biflavones, characterized by the following steps of the method:
- 5 a) extracting leaves of ginkgo biloba using water-containing acetone, a water-containing alkanol having 1 to 3 carbon atoms or anhydrous methanol,
- b) separating the organic solvent by concentrating the solvent to a content of 10% by weight at the most, wherein water may be added during the final distillation steps,
- 10 c) diluting the remaining aqueous solution with water to a solids content of 5 to 25% by weight, cooling to a temperature below 25°C and allowing to stand until a precipitate is formed,
- d) treating the remaining aqueous solution with ammonium sulfate and subsequently extracting at least once using methyl ethyl ketone or a mixture of methyl ethyl ketone and acetone,
- 15 e) concentrating the extract obtained and diluting with an ethanol-water mixture to obtain a solution containing 50% by weight water and 50% by weight ethanol at a solids content of 10% by weight,
- 20 f) treating the solution with a lead compound or an insoluble polyamide,
- g) extracting the filtered solution using an aliphatic or cycloaliphatic solvent having a boiling point of 60 to 100°C,
- h) concentrating the remaining aqueous-alcoholic solution, followed by treating with ammonium sulfate and extracting with methyl ethyl ketone and ethanol,
- 25 i) concentrating the organic phase obtained to a solids content of 50 to 70% by weight,
- k) dissolving the concentrate in an aqueous alkanol having 1 to 3 carbon atoms and optionally filtering,
- 30 l) filtration over an adsorber resin and/or an ion exchanger, wherein the substances to be removed are retained on the resin and

m) drying the concentrate at reduced pressure to obtain a dry extract having a water content of less than 5%.

2. Method according to claim 1, wherein the aqueous alkanol in step k) is aqueous ethanol.
3. Method according to claim 2, wherein the aqueous ethanol is ethanol having a content of 40 to 60% by weight.
4. Method according to any one of claims 1 to 3, wherein the adsorber resin in step l) is a resin based on optionally substituted styrenes/divinylbenzenes.
5. Method according to any one of claims 1 to 3, wherein the ion exchanger in step l) is a strongly acidic ion exchanger.
6. Extract from leaves of ginkgo biloba, characterized by having the following contents:
  - less than 20 ppm 4'-O-methyl pyridoxine and/or
  - less than 20 ppm biflavones.
7. Extract according to claim 6, characterized by having a content of less than 10 ppm 4'-O-methyl pyridoxine.
8. Extract according to claim 6, characterized by having a content of less than 2 ppm 4'-O-methyl pyridoxine.
9. Extract according to any one of claims 6 to 8, characterized by having a content of less than 10 ppm biflavones.
10. Extract according to any one of claims 6 to 8, characterized by having a content of less than 5 ppm biflavones.

11. Extract according to any of claims 6 to 10, characterized by having the following contents:
- 20 to 30% by weight flavonoids,
  - 4.5 to 8.5% by weight terpene lactones and
  - 5   • less than 10 ppm ginkgolic acids.
12. Extract according to any of claims 6 to 10, characterized by having the following contents:
- 22.0 to 27.0 % by weight flavonoids,
  - 10   • 5.0 to 7.0% by weight terpene lactones and
  - 5 ppm ginkgolic acids at the most.
13. Extract according to any of claims 6 to 10, characterized by having the following contents:
- 15   • 22.0 to 27.0 % by weight flavonoids,
  - 2.8 to 3.4% by weight ginkgolides A, B and C in total,
  - 2.6 to 3.2% by weight bilobalid and
  - 5 ppm ginkgolic acids at the most.
- 20   14. Use of an extract according to any one of claims 6 to 13 for the manufacture of a medicament, food product or other preparation for the treatment of dementia and symptoms thereof and/or cerebral or peripheral blood circulation disorders.
- 25   15. Medicament, food product or other preparation, characterized by a content of a ginkgo extract according to any one of claims 6 to 13.

# INTERNATIONAL SEARCH REPORT

International application No  
PCT/EP2006/004029

## A. CLASSIFICATION OF SUBJECT MATTER

INV. A61K36/00

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 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 practical, search terms used)

EPO-Internal, WPI Data, BIOSIS, MEDLINE, EMBASE

## C. DOCUMENTS CONSIDERED TO BE RELEVANT

| Category* | Citation of document, with indication, where appropriate, of the relevant passages                                                                                                                                                                                  | Relevant to claim No. |
|-----------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-----------------------|
| X         | EP 1 037 646 A (DR. WILLMAR SCHWABE GMBH & CO) 27 September 2000 (2000-09-27)<br>claims 1,2,6,7                                                                                                                                                                     | 6-15                  |
| Y         | DE 39 40 091 A1 (DR. WILLMAR SCHWABE GMBH & CO, 7500 KARLSRUHE, DE)<br>6 June 1991 (1991-06-06)<br>the whole document                                                                                                                                               | 1-5                   |
| Y         | ARENZ ANSGAR ET AL: "Occurrence of neurotoxic 4'-O-methylpyridoxine in Ginkgo biloba leaves, Ginkgo medications and Japanese Ginkgo food"<br>PLANTA MEDICA,<br>vol. 62, no. 6, 1996, pages 548-551,<br>XP009070428<br>ISSN: 0032-0943<br>page 549, left-hand column | 1-5                   |

☒ 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 document 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.
- \*Z\* document member of the same patent family

Date of the actual completion of the international search

11 September 2006

Date of mailing of the international search report

25/09/2006

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## INTERNATIONAL SEARCH REPORT

International application No

PCT/EP2006/004029

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

| Category* | Citation of document, with indication, where appropriate, of the relevant passages                                                                                                                                                                | Relevant to claim No. |
|-----------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-----------------------|
| Y         | DATABASE WPI<br>Section Ch, Week 200525<br>Derwent Publications Ltd., London, GB;<br>Class B04, AN 2005-234134<br>XP002398187<br>& CN 1 557 455 A (ZHANG Z)<br>29 December 2004 (2004-12-29)<br>abstract                                          | 1-5                   |
| Y         | EP 0 360 556 A (INDENA S.P.A)<br>28 March 1990 (1990-03-28)<br>claims 1,15; example 3                                                                                                                                                             | 1-5                   |
| A         | KRESSMANN S ET AL: "Pharmaceutical quality<br>of different Ginkgo biloba brands"<br>May 2002 (2002-05), JOURNAL OF PHARMACY<br>AND PHARMACOLOGY, VOL. 54, NR. 5, PAGE(S)<br>661-669, XP009070389<br>ISSN: 0022-3573<br>page 662, left-hand column | 12,13                 |
| A         | BEEK VAN T A: "GINKGO BILOBA L"<br>FITOTERAPIA, IDB HOLDING, MILAN, IT,<br>vol. I.XIX, no. 3, 1998, pages 195-244,<br>XP000700334<br>ISSN: 0367-326X<br>page 198; table 1                                                                         | 1-15                  |

# INTERNATIONAL SEARCH REPORT

International application No.  
PCT/EP2006/004029

## Box 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:  
see FURTHER INFORMATION sheet PCT/ISA/210
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 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:

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 fee, this Authority did not invite payment of any additional fee.
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.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.  
☐ No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Continuation of Box II.2

Claims Nos.: -

It is pointed out to the Applicant concerning claims 6-15, that the term "flavonoids" used in claims 11-13 normally encompasses also "biflavones", see claim 6. According to pages 2 and 4 of the description, Applicant means under "biflavones" amentoflavone, bilobetin, ginkgetin, isoginkgetin and sciadopitysin and under "flavonoids" quercetin, kaempferol and isorhamnetin calculated as flavonoid glycosides.

Applicant is asked to remove said unclarity / ambiguity.

Moreover, having regard to claim 1, it is indicated that it is not clear whether in step 1) the filtration over an adsorber resin or over an ion exchanger leads to the reduced content of 4'-O-methyl pyridoxine or to the reduced content of biflavones, respectively. This should be clarified.

The applicant's attention is drawn to the fact that claims relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure. If the application proceeds into the regional phase before the EPO, the applicant is reminded that a search may be carried out during examination before the EPO (see EPO Guideline C-VI, 8.5), should the problems which led to the Article 17(2) declaration be overcome.

# INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No

PCT/EP2006/004029

| Patent document<br>cited in search report |    | Publication<br>date | Patent family<br>member(s) | Publication<br>date |
|-------------------------------------------|----|---------------------|----------------------------|---------------------|
| EP 1037646                                | A  | 27-09-2000          | CN 1282252 A               | 31-01-2001          |
|                                           |    |                     | WO 9932129 A1              | 01-07-1999          |
|                                           |    |                     | DE 19756848 A1             | 01-07-1999          |
|                                           |    |                     | JP 2001526235 T            | 18-12-2001          |
|                                           |    |                     | US 6328999 B1              | 11-12-2001          |
| DE 3940091                                | A1 | 06-06-1991          | CA 2031385 A1              | 05-06-1991          |
|                                           |    |                     | DK 431535 T3               | 28-03-1994          |
|                                           |    |                     | EP 0431535 A1              | 12-06-1991          |
|                                           |    |                     | ES 2024400 T3              | 16-04-1994          |
|                                           |    |                     | GR 91300122 T1             | 20-03-1992          |
|                                           |    |                     | JP 2045722 C               | 25-04-1996          |
|                                           |    |                     | JP 3279331 A               | 10-12-1991          |
|                                           |    |                     | JP 7076176 B               | 16-08-1995          |
|                                           |    |                     | KR 154977 B1               | 16-11-1998          |
| CN 1557455                                | A  | 29-12-2004          | NONE                       |                     |
| EP 0360556                                | A  | 28-03-1990          | CA 1337973 C               | 23-01-1996          |
|                                           |    |                     | DE 68906108 D1             | 27-05-1993          |
|                                           |    |                     | DE 68906108 T2             | 29-07-1993          |
|                                           |    |                     | ES 2054018 T3              | 01-08-1994          |
|                                           |    |                     | GR 3007688 T3              | 31-08-1993          |
|                                           |    |                     | HK 43694 A                 | 13-05-1994          |
|                                           |    |                     | JP 2121998 A               | 09-05-1990          |
|                                           |    |                     | JP 2800040 B2              | 21-09-1998          |
|                                           |    |                     | KR 169957 B1               | 01-02-1999          |

15

**MÉTODO MEJORADO PARA PREPARAR EXTRACTOS DE GINKGO QUE TIENEN UN CONTENIDO BAJO DE 4'-O-METIL PIRIDOXINA Y/O BIFLAVONAS**

La presente invención se relaciona con un método mejorado de pasos múltiples para preparar un extracto de ginkgo biloba que tiene un contenido reducido de 4'-O-metil piridoxina y/o biflavonas, donde la reducción del contenido se efectúa por filtración a través de una resina de adsorción y/o un intercambiador de iones, y las sustancias a eliminar se retienen en la resina. Además, la invención se relaciona con un extracto de ginkgo biloba que tiene un contenido reducido de 4'-O-metil piridoxina y/o biflavonas que puede obtenerse con el método de acuerdo con la presente invención, así como con su uso.

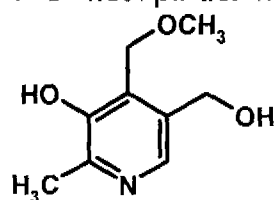
Los extractos de hojas de ginkgo biloba se usan como medicamentos desde hace décadas. En la actualidad se los usa para el tratamiento de distintos tipos de demencia, y de los síntomas relacionados con esta enfermedad, así como trastornos de la circulación sanguínea cerebral y periférica. Los ingredientes con los cuales se asocia la eficacia son las lactonas de terpenos (ginkgólidos A, B, C y bilobalida), así como los glicoósidos de flavonas (quercetina, kaempferol e isoramnetina). Las hojas de ginkgo biloba también contienen componentes que no contribuyen a la eficacia deseada, sino que pueden ser responsables de riesgos y efectos colaterales. Además de los ingredientes vegetales no polares, tales como los ácidos ginkgólicos, aquellos componentes son la 4'-O-metil piridoxina y las biflavonas. Por lo tanto, en un extracto de ginkgo, que al mismo tiempo es eficaz, es tan seguro como

15

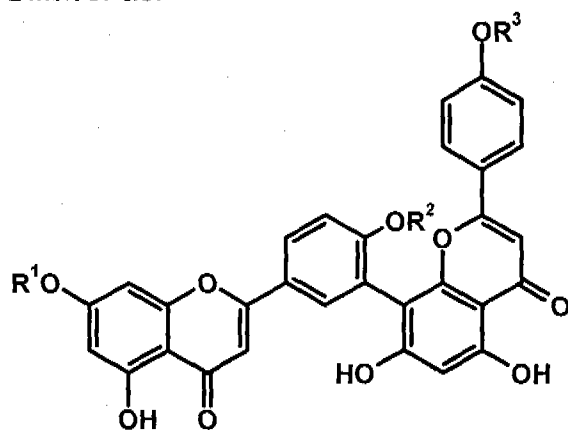
resulta posible y presenta la menor cantidad de efectos colaterales posibles, estos compuestos no deberían estar presentes en la mayor medida posible.

La 4'-O-metil piridoxina puede provocar síntomas de envenenamiento, tales como ataques convulsivos e inconsciencia. Por consiguiente, este compuesto también se conoce como ginkgotoxina. Las biflavonas contenidas en el ginkgo presentan potencial inmunotóxico y pueden provocar alergias de contacto. Estas biflavonas contenidas en ginkgo son predominantemente los compuestos amentoflavona, bilobetina, ginkgetina, isoginkgetina y sciadopitisina.

4'-O-Metil piridoxina:



Biflavonas:



Amentoflavona:  $R^1 = R^2 = R^3 = H$

Bilobetina:  $R^1 = R^3 = H, R^2 = OCH_3$

Ginkgetina:  $R^3 = H, R^1 = R^2 = OCH_3$

Isoginkgetina:  $R^1 = H, R^2 = R^3 = OCH_3$

Sciadopitisina:  $R^1 = R^2 = R^3 = OCH_3$

Se han descripto métodos para reducir simultáneamente el contenido de 4'-O-metil piridoxina y biflavonas, así como los extractos obtenidos, en EP 1 037 646 B1. En dicha publicación, se retiene la 4'-O-metil piridoxina usando un intercambiador de cationes ácidos, y las biflavonas se adsorben en carbón activado. Preferiblemente, estos pasos de reducción de contenido se realizan después del paso f) del método de acuerdo con EP 1 037 646 B1 (página 3), es decir, después de "tratar la solución con un compuesto de plomo o una poliamida insoluble". El método de EP 1 037 646 B1 se basa en el método descripto originalmente en DE 39 40 091 C2.

En EP 1 037 646 B1, la reducción del contenido de biflavonas y 4'-O-metil piridoxina se realiza en una etapa del método (paso f)) en la que hay contenidos relativamente elevados de sales de plomo y sales de amonio presentes, de modo que es necesario emplear una mayor cantidad de intercambiador de iones.

Además, se describen y reivindican las reducciones en el contenido de biflavonas y 4'-O-metil piridoxina solamente en combinación. Sin embargo, dependiendo de los límites que posiblemente se establezcan en el futuro, y considerando el requerimiento de realizar una modificación tan pequeña de la composición del extracto como sea posible, puede ser deseable eliminar solamente las biflavonas o la 4'-O-metil piridoxina.

Además, los contenidos de menos de 50 ppm de 4'-O-metil piridoxina y menos de 100 ppm de biflavonas indicados en EP 1 037 646 B1 aún son comparativamente elevados. Como en EP 1 037 646 B1 no se describe un ejemplo comparativo, tampoco resulta posible observar el grado de reducción real del contenido obtenido en los Ejemplos 2 y 3 a partir de esta referencia.

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Por consiguiente, el objeto subyacente de la presente invención es proporcionar un método para preparar extractos de ginkgo que tienen un contenido bajo de 4'-O-metil piridoxina y/o un contenido bajo de bioflavonas, que no presente las desventajas indicadas previamente y, en particular,

- en el que se empleen adsorbentes reciclables y/o de muy bajo costo,
- que resulte en contenidos de 4'-O-metil piridoxina menores que 20 ppm, preferiblemente menores que 10 ppm, y en particular preferiblemente menores que 2 ppm, y/o contenidos de biflavonas menores que 20 ppm, preferiblemente menores que 10 ppm y en particular preferiblemente menores que 5 ppm, y
- que resulte en extractos que tengan contenidos preferidos de flavonoides de entre 20 y 30% en peso (en particular preferiblemente, entre 22,0 y 27,0%), de lactonas de terpenos de entre 4,5 y 8,5% en peso (en particular preferiblemente, entre 5,0 y 7,0%, preferiblemente entre 2,8 y 3,4% de los ginkgólidos A, B y C, y entre 2,6 y 3,2% de bilobalida), y de ácidos ginkgólicos (a lo sumo de 5 ppm), donde dichos contenidos de flavonoides de entre 22,0 y 27,0%, de lactonas de terpenos de entre 5,0 y 7,0% (entre 2,8 y 3,4% de ginkgólidos A, B y C, y entre 2,6 y 3,2% de bilobalida), y de ácidos ginkgólicos (a lo sumo de 5 ppm) satisfacen los requerimientos de la Farmacopea Alemana 2003 ("Deutsches Arzneibuch"; DAB).

De acuerdo con la DAB, los flavonoides se determinan bajo la forma de quercetina, kaempferol e isoramnetina después de una hidrólisis ácida, y se calculan como glicósidos flavonoides.

Este objeto podría alcanzarse separando las biflavonas con resinas de adsorción, y/o separando la 4'-O-metil piridoxina con intercambiadores de iones ácidos.

Con el fin de producir los extractos de acuerdo con la presente invención, preferiblemente se pone en práctica el siguiente método, donde los pasos a) a i), así como m), son como se ha descrito esencialmente en DE 39 40 091 C2, cuya descripción total, en particular respecto de los detalles del método, se incorporará en la presente solicitud a modo de referencia:

- a) extraer las hojas de ginkgo biloba (droga) a una temperatura de 20-100°C, preferiblemente 40-60°C, usando acetona con agua, con un contenido de 20-90% en peso, un alcohol que contiene agua con entre 1 y 3 átomos de carbono (metanol, etanol, n-propanol e isopropanol), y un contenido de 20-90% en peso de metanol anhidro, donde la relación droga: solvente es de entre 1:4 y 1:20, preferiblemente entre 1:5 y 1:10,
- b) separar el solvente orgánico evaporando el solvente hasta un contenido de a lo sumo 10% en peso (preferiblemente a lo sumo 5% en peso), donde puede agregarse agua durante los pasos de destilación finales,
- c) diluir la solución acuosa remanente con agua hasta obtener un contenido de sólidos de entre 5 y 25% en peso (preferiblemente entre 15 y 20% en peso), enfriar la solución a una temperatura inferior a 25°C (preferiblemente de entre aproximadamente 10 y 12°C), y permitir que la solución acuosa remanente repose hasta que se forme un precipitado,
- d) tratar la solución acuosa remanente con sulfato de amonio (preferiblemente hasta un contenido de hasta 40% en peso, en particular preferiblemente entre 25 y 35% en peso), y luego extraer la solución al menos una vez usando metil

etil cetona o una mezcla de metil etil cetona y acetona (preferiblemente en una relación de entre 9:1 y 4:6, en particular 6:4),

e) concentrar el extracto obtenido (preferiblemente hasta obtener un contenido de sólidos de entre 50 y 70% en peso) y diluir el extracto con una mezcla de etanol y agua para obtener una solución que contiene 50% en peso de agua y 50% en peso de etanol, y que tiene un contenido de sólidos de 10% en peso,

f) tratar la solución con un compuesto de plomo (preferiblemente con acetato de plomo, acetato de hidróxido de plomo, nitrato de plomo o una suspensión acuosa de hidróxido de plomo) hasta que se obtiene un cambio de color, de marrón a pardo, o una poliamida insoluble (preferiblemente poliamida 6, poliamida 6,6 o polivinilpirrolidona reticulada (polividona)),

g) extraer la solución filtrada usando un solvente alifático o cicloalifático con un punto de fusión de entre 60 y 100°C,

h) concentrar la solución acuosa alcohólica remanente, luego tratar la solución con sulfato de amonio y extraer la solución con metil etil cetona y etanol, (preferiblemente se concentra la solución a presión reducida, hasta obtener un contenido de etanol de hasta aproximadamente 5%, agregado con sulfato de amonio hasta un contenido de 20% en peso y extraído con el solvente en una relación de entre 8:2 y 5:5, preferiblemente 6:4),

i) concentrar la fase orgánica obtenida hasta obtener un contenido de sólidos de entre 50 y 70% en peso,

k) disolver el concentrado en un alcohol acuoso que tiene entre 1 y 3 átomos de carbono, y opcionalmente filtrar el producto,

l) filtrar sobre una resina de adsorción y/o un intercambiador de iones, donde las sustancias a eliminar se retienen en la resina, y

m) secar el concentrado (preferiblemente a una temperatura máxima de entre 60 y 80°C) a presión reducida para obtener un extracto seco que tiene un contenido de agua menor que 5%.

El método de acuerdo con la presente invención tiene la ventaja de que el paso de reducción de contenido se realiza al final del método. Por consiguiente, es necesario emplear una menor cantidad de intercambiador de iones, y se obtienen contenidos particularmente bajos de 4'-O-metil piridoxina y/o biflavonas.

Las resinas de adsorción preferidas en el paso l) son resinas basadas en estirenos/divinilbencenos opcionalmente sustituidos, tales como Diaion HP-20, HP-21 o Sepabeads SP-207 y SP-850. Los intercambiadores de iones preferidos son intercambiadores de iones fuertemente ácidos, tales como Merck I o Amberlite IR-120. La filtración incluye opcionalmente el lavado usual usando solvente adicional. El alcohol acuoso en el paso k) es metanol, etanol, n-propanol o isopropanol acuoso, preferiblemente con un contenido de 20-90% en peso, respectivamente, de etanol acuoso, donde se prefiere en particular una concentración de entre 40 y 60% en peso.

Un sujeto de la presente invención lo constituyen los extractos, en particular los extractos secos, que pueden obtenerse con el método de acuerdo con la presente invención, y que presentan los siguientes contenidos:

- menos de 20 ppm, preferiblemente menos de 10 ppm y en particular menos de 2 ppm de 4'-O-metil piridoxina, y/o
- menos de 20 ppm, preferiblemente menos de 10 ppm y en particular menos de 5 ppm de biflavonas.

Además, un sujeto de la presente invención lo constituyen los extractos, en particular los extractos secos, que pueden obtenerse con el método de acuerdo con la presente invención, y que presentan los siguientes contenidos adicionales:

- entre 20 y 30% en peso, preferiblemente entre 22,0 y 27,0% en peso de flavonoides,
- entre 4,5 y 8,5% en peso, preferiblemente entre 5,0 y 7,0% en peso de lactonas de terpenos, y
- menos de 10 ppm, preferiblemente no más de 5 ppm de ácidos ginkgólicos.

Otro sujeto de la presente invención lo constituyen los extractos, en particular los extractos secos, que pueden obtenerse con el método de acuerdo con la presente invención, y que presentan los siguientes contenidos:

- entre 20 y 30% en peso, preferiblemente entre 22,0 y 27,0% en peso flavonoides,
- entre 2,5 y 4,5% en peso de ginkgólidos A, B y C en total, preferiblemente entre 2,8 y 3,4% en peso de ginkgólidos A, B y C en total,
- entre 2,0 y 4,0% en peso, preferiblemente entre 2,6 y 3,2% en peso de bilobalida,
- menos de 10 ppm, preferiblemente no más de 5 ppm de ácidos ginkgólicos, y
- menos de 20 ppm, preferiblemente menos de 10 ppm, en particular preferiblemente menos de 2 ppm de 4'-O-metil piridoxina, y/o
- menos de 20 ppm, preferiblemente menos de 10 ppm y en particular preferiblemente menos de 5 ppm de biflavonas,

De acuerdo con la Farmacopea Europea, los extractos secos generalmente tienen un residuo seco de al menos 95% en peso.

Los extractos de acuerdo con la presente invención pueden administrarse bajo la forma de polvos, gránulos, tabletas, grageas (tabletas recubiertas) o cápsulas, preferiblemente por vía oral. Con el fin de preparar tabletas, el extracto se mezcla con coadyuvantes apropiados aceptables para el uso farmacéutico, tales como lactosa, celulosa, dióxido de silicio, croscarmellose y estearato de magnesio, y se comprime en tabletas, sobre que las opcionalmente se aplica un recubrimiento apropiado, por ejemplo, hecho de hidroximetilcelulosa, polietilenglicol, pigmentos (tales como dióxido de titanio, óxido de hierro) y talco. El extracto de acuerdo con la presente invención también puede colocarse en cápsulas, opcionalmente con la adición de coadyuvantes, tales como estabilizadores, rellenos y semejantes. La dosificación es tal que se administran entre 10 y 2000 mg, preferiblemente entre 50 y 1000 mg, y en particular preferiblemente entre 100 y 500 mg de extracto por día.

Otro sujeto de la presente invención lo constituyen los medicamentos, los productos alimenticios y otras preparaciones que contienen estos extractos, opcionalmente en combinación con otras sustancias, tales como ingredientes activos y/o coadyuvantes aceptables para el uso farmacéutico. El término "producto alimenticio", tal como se lo usa en la presente documentación, particularmente hace referencia a productos alimenticios dietéticos, suplementos para productos dietéticos, y también suplementos para alimentos médicos y dietéticos.

## EJEMPLOS

### **Preparación de la solución de extracto usada en los ejemplos 1 y 2, y también en los ejemplos comparativos 1 y 2**

Se extrajeron hojas secas de ginkgo biloba (droga) a una temperatura de aproximadamente 50°C usando siete veces su peso (p/p) (es decir, la relación de droga-solvente es de 1:7) de acetona/agua 60/40 (p/p) (paso a)).

El solvente orgánico se separó en gran medida del extracto (paso b)), y la solución acuosa concentrada remanente se diluyó con agua hasta obtener un contenido de sólidos de aproximadamente 10% en peso. La solución se enfrió hasta una temperatura de aproximadamente 12°C con agitación, y se eliminó el precipitado resultante (paso c)).

Se agregó aproximadamente 30% en peso de sulfato de amonio a la solución acuosa remanente, y se extrajo la solución formada usando una mezcla de metil etil cetona y acetona en una relación de 6:4 (paso d)).

El extracto obtenido se concentró en gran medida, y el concentrado obtenido de este modo se diluyó con agua y etanol, de modo que se obtuvo una solución que contenía 50% en peso de agua y 50% en peso de etanol, con un contenido de sólidos de aproximadamente 10% en peso (paso e)). Se filtró la solución y se agregó el filtrado con una solución acuosa de acetato de hidróxido de plomo (paso f)).

Después de una filtración adicional, se extrajo la solución acuosa alcohólica remanente con heptano (paso g)).

Luego se concentró la solución acuosa remanente a presión reducida hasta obtener un contenido de etanol de aproximadamente 5%, y se agregó

sulfato de amonio hasta obtener un contenido de aproximadamente 20% en peso, sobre la base del contenido de agua.

La solución obtenida se extrajo usando una mezcla de metil etil cetona y etanol en una relación de 6:4 (paso h)).

La fase orgánica resultante se extrajo en gran medida (con un contenido de sólidos de aproximadamente 55% en peso) (paso i)). Luego se diluyó el producto con etanol y agua, de modo que se obtuvo una solución con un contenido de etanol de aproximadamente 75% y un contenido de sólidos de aproximadamente 15% (paso k)).

**Ejemplo 1: Extracto de ginkgo que tiene un contenido reducido de 4'-O-metil piridoxina**

Se ajustaron con agua 1249 g de la solución de extracto, obtenida de acuerdo con el método de preparación descrito previamente, hasta alcanzar un contenido de etanol de aproximadamente 40% (p/p), y se la aplicó a 50 ml (que corresponden a 0,28 ml/g del extracto seco) de Amberlite IR-120 (intercambiador de iones fuertemente ácido) (columna: de aproximadamente 2 x 16,5 cm; flujo: aproximadamente 8 ml/minuto) (paso l)), a lo que siguió un lavado con 200 ml de etanol al 50% (p/p). La solución resultante se concentró en un evaporador rotativo y se secó al vacío a 50°C (paso m)): 179,4 g.

Como puede apreciarse en la siguiente tabla, se obtuvo un contenido de 4'-O-metil piridoxina de 0,4 ppm, sin modificar esencialmente los contenidos de los ingredientes con relevancia en la eficacia.

**Ejemplo comparativo 1: Extracto de ginkgo sin reducción de contenido de acuerdo con el paso l) de la presente invención**

Se concentraron por evaporación 98,4 g de la solución de extracto empleada en el ejemplo 1, se secó al vacío a 50°C: 14,7 g.

**Ejemplo 2a): Extracto de ginkgo que tiene un contenido reducido de biflavonas**

Se ajustaron con agua 500 g de la solución de extracto obtenida de acuerdo con el método de preparación descrito previamente, hasta alcanzar un contenido de etanol de aproximadamente 40% (p/p), y se los aplicó a 400 ml (que corresponden a 5,7 ml/g del extracto seco) de Sepabeads SP-850 (resina de adsorción) (columna: de aproximadamente 4 x 32 cm; flujo: aproximadamente 8 ml/minuto) (paso l)), a lo que siguió un lavado con aproximadamente 2 l de 40% de etanol (p/p) en total. Se concentró 10% de la solución resultante (259,3 g) en un evaporador rotativo y se secó por congelamiento (paso m)): 6,92 g. El 90% restante de la solución se empleó en el ejemplo 2b).

Como puede apreciarse en la siguiente tabla, se obtuvieron contenidos de biflavona menores que 1 ppm, respectivamente, sin modificar esencialmente los contenidos de los ingredientes con relevancia en la eficacia.

**Ejemplo 2b): Extracto de ginkgo que tiene un contenido reducido de 4'-O-metil piridoxina y biflavonas**

Se aplicó 90% de la solución obtenida en el ejemplo 2a) (2334,1 g) a 25 ml (que corresponde a 0,40 ml/g del extracto seco) de Amberlite IR-120 (intercambiador de iones fuertemente ácido) (columna: de aproximadamente 1,5 x 11 cm; flujo: aproximadamente 10 ml/minuto) (paso l)), a lo que siguió un lavado con 100 ml de etanol al 40% (p/p). La solución resultante se concentró en un evaporador rotativo y se secó por congelamiento (paso m)): 61,67 g.

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Como puede apreciarse en la siguiente tabla, se obtuvo un contenido de 4'-O-metil piridoxina de 0,4 ppm y contenidos de biflavonas menores que 1 ppm, respectivamente, sin modificar esencialmente los contenidos de los ingredientes con relevancia en la eficacia.

**Ejemplo comparativo 2: Extracto de ginkgo sin reducción de contenido de acuerdo con el paso I) de la presente invención**

Se concentraron por evaporación 50 g de la solución de extracto empleada en el ejemplo 2a), y se secó por congelamiento: 7,05 g.

**Ejemplo comparativo 3: Extracto de ginkgo con reducción del contenido de biflavona y reducción del contenido de 4'-O-metil piridoxina después de un tratamiento con un compuesto de plomo (paso f)) de acuerdo con EP 1 037 646 B1**

Se extrajeron hojas secas de ginkgo biloba (drug) a una temperatura de aproximadamente 50°C usando siete veces su peso (p/p) (es decir, la relación de droga-solvente es de 1:7) de acetona/agua 60/40 (p/p) (paso a)).

El solvente orgánico se separó en gran medida del extracto y la solución acuosa concentrada remanente se diluyó con agua hasta obtener un contenido de sólidos de aproximadamente 10% en peso, se enfrió hasta una temperatura de aproximadamente 12°C con agitación, y se eliminó el precipitado resultante.

Se agregó aproximadamente 30% en peso de sulfato de amonio a la solución acuosa remanente, y se extrajo la solución formada usando una mezcla de metil etil cetona y acetona en una relación de 6:4.

El extracto obtenido se concentró en gran medida, y el concentrado obtenido de este modo se diluyó con agua y etanol, de modo que se obtuvo una solución que contenía 50% en peso de agua y 50% en peso de etanol, con

un contenido de sólidos de aproximadamente 10% en peso. Se filtró la solución y se agregó el filtrado con una solución acuosa de acetato de hidróxido de plomo (paso f)).

Se agitaron 150 ml de la solución obtenida de este modo (porción de extracto seco: 7,1%; contenido de etanol: aproximadamente 50%) con 2,5 g (que corresponden a 0,17 g/g de producto final) de carbón activado por 30 minutos a temperatura ambiente. El filtrado se aplicó a una columna con 4,2 ml (que corresponden a 0,28 ml/g de producto final) de un intercambiador de cationes fuertemente ácido (Merck I). La columna intercambiadora de iones se lavó 4 veces con 5 ml de EtOH (al 50%).

Las porciones eluidas combinadas se extrajeron con agitación usando 3 x 70 ml de n-hexano. La fase de etanol/agua se concentró en un evaporador rotativo y se diluyó con agua hasta 100 g. La solución de extracto obtenida de este modo se extrajo una vez con 40 ml, 2 veces con 30 ml y una vez con 20 ml de una mezcla de metil etil cetona:etanol en una relación de 3:2, donde se agregaron 15 g de sulfato de amonio para efectuar la separación de fases.

La fase orgánica se agitó por 1 hora a temperatura ambiente después de agregar 25 g de sulfato de amonio. Se separó el sulfato de amonio no disuelto, así como la fase acuosa formada. La solución de extracto se concentró en un evaporador rotativo a 50°C y se secó al vacío a 60°C: 14,8 g.

Como puede observarse en la siguiente tabla, se obtuvo un contenido de 4'-O-metil piridoxina de 52 ppm y un contenido de biflavona de 31,3 ppm, lo que representa contenidos significativamente mayores y resultados de reducción de contenido inferiores respecto del ejemplo 2b) de acuerdo con la presente invención.

La determinación de los contenidos de flavonoides, lactonas de terpenos y ácidos ginkgólicos indicados en la Tabla 1 se realizó de acuerdo con la DAB.

La determinación cuantitativa de la 4'-O-metil piridoxina se realizó por medio de una detección con HPLC y fluorescencia, después de efectuar un enriquecimiento con cartuchos Sep Pak Plus C18, a lo que siguió un enriquecimiento con cartuchos Bond Elut LRC-SCX, donde se empleó una columna Fenomenex Prodigy ODS-3 de HPLC de 5  $\mu$  y un gradiente de ácido trifluoroacético (pH 2): metanol = 9:1 (v/v)  $\rightarrow$  metanol.

Se realizó la determinación de los contenidos de las biflavonas indicadas en la Tabla 1 por medio de una detección con HPLC y UV a 340 nm, empleando una columna Waters Nova Pak C18 con un gradiente de agua: acetonitrilo de 9:1 (v/v) + 0,3% en volumen de ácido fosfórico (85%)  $\rightarrow$  acetonitrilo + 0,3% en volumen de ácido fosfórico.

En cada caso, se realizó la calibración usando sustancias de referencia con un grado elevado de pureza definida.

**Tabla 1: Composición de los extractos de acuerdo con los ejemplos proporcionados previamente**

| Extracto de acuerdo con            | Ejemplo 1 | Ejemplo comparativo 1 | Ejemplo 2a) | Ejemplo 2b) | Ejemplo comparativo 2 | Ejemplo comparativo 3 |
|------------------------------------|-----------|-----------------------|-------------|-------------|-----------------------|-----------------------|
| Flavonoides [%]                    | 24,86     | 24,51                 | 27,17       | 25,32       | 25,89                 | 22,78                 |
| Ginkgólidos A, B, C [%]            | 3,28      | 2,94                  | 2,97        | 3,04        | 2,97                  | 3,40                  |
| Bilobalida [%]                     | 3,17      | 3,03                  | 3,18        | 3,14        | 3,07                  | 3,07                  |
| Lactonas de terpenos [%]           | 6,45      | 5,97                  | 6,15        | 6,18        | 6,04                  | 6,47                  |
| Ácidos ginkgólicos [ppm]           | < 5       | < 5                   | < 5         | < 5         | < 5                   | < 5                   |
| 4'-O-metil piridoxina [ppm]        | 0,4       | 104,5                 | 164         | 0,8         | 89                    | 52                    |
| Amentoflavona [ppm]                | 68,5      | 78,7                  | n. d. (< 1) | n. d. (< 1) | 81,6                  | 12,6                  |
| Bilobetina [ppm]                   | 123,3     | 120,4                 | n. d. (< 1) | n. d. (< 1) | 136,4                 | 10,4                  |
| Ginkgetina [ppm]                   | 86,3      | 82,1                  | n. d. (< 1) | n. d. (< 1) | 86,1                  | 2,7                   |
| Isoginkgetina [ppm]                | 103,3     | 104,4                 | n. d. (< 1) | n. d. (< 1) | 111,5                 | 4,8                   |
| Sciadopitisina [ppm]               | 44,0      | 43,0                  | n. d. (< 1) | n. d. (< 1) | 44,0                  | 0,8                   |
| Cantidad total de biflavonas [ppm] | 425,4     | 428,6                 | n. d. (< 5) | n. d. (< 5) | 459,6                 | 31,3                  |

n. d. = no detectable

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**REIVINDICACIONES:**

1. Un método para producir un extracto de ginkgo biloba que tiene un contenido reducido de 4'-O-metil piridoxina y/o biflavonas, CARACTERIZADO PORQUE comprende los siguientes pasos:
  - a) extraer hojas de ginkgo biloba usando acetona con agua, un alcohol que contiene agua con entre 1 y 3 átomos de carbono, o metanol anhidro,
  - b) separar el solvente orgánico concentrando el solvente hasta obtener un contenido de a lo sumo 10% en peso, donde puede agregarse agua durante los pasos de destilación finales,
  - c) diluir la solución acuosa remanente con agua hasta obtener un contenido de sólidos de entre 5 y 25% en peso, enfriar a una temperatura inferior a 25°C, y permitir el reposo hasta la formación de un precipitado,
  - d) tratar la solución acuosa remanente con sulfato de amonio y luego realizar al menos una extracción usando metil etil cetona o una mezcla de metil etil cetona y acetona,
  - e) concentrar el extracto obtenido y diluir con una mezcla de etanol y agua para obtener una solución que contiene 50% en peso de agua y 50% en peso de etanol, con un contenido de sólidos de 10% en peso,
  - f) tratar la solución con un compuesto de plomo o una poliamida insoluble,
  - g) extraer la solución filtrada usando un solvente alifático o cicloalifático con un punto de fusión de entre 60 y 100°C,
  - h) concentrar la solución acuosa alcohólica remanente, luego realizar un tratamiento con sulfato de amonio y una extracción con metil etil cetona y etanol,

- i) concentrar la fase orgánica obtenida hasta obtener un contenido de sólidos de entre 50 y 70% en peso,
- k) disolver el concentrado en un alcohol acuoso que tiene entre 1 y 3 átomos de carbono, y opcionalmente filtrar,
- l) filtrar sobre una resina de adsorción y/o un intercambiador de iones, donde las sustancias a eliminar se retienen en la resina, y
- m) secar el concentrado a presión reducida para obtener un extracto seco que tiene un contenido de agua menor que 5%.
2. Un método de acuerdo con la reivindicación 1, CARACTERIZADO PORQUE el alcohol acuoso en el paso k) es etanol acuoso.
  3. Un método de acuerdo con la reivindicación 2, CARACTERIZADO PORQUE el etanol acuoso es etanol con un contenido de entre 40 y 60% en peso.
  4. Un método de acuerdo con cualquiera de las reivindicaciones 1 a 3, CARACTERIZADO PORQUE la resina de adsorción del paso l) es una resina basada en estirenos/divinilbencenos opcionalmente sustituidos.
  5. Un método de acuerdo con cualquiera de las reivindicaciones 1 a 3, CARACTERIZADO PORQUE el intercambiador de iones del paso l) es un intercambiador de iones fuertemente ácido.
  6. Un extracto de hojas de ginkgo biloba CARACTERIZADO PORQUE tiene los siguientes contenidos:
    - menos de 20 ppm de 4'-O-metil piridoxina y/o
    - menos de 20 ppm de biflavonas.

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7. Un extracto de acuerdo con la reivindicación 6, CARACTERIZADO PORQUE tiene un contenido de 4'-O-metil piridoxina de menos de 10 ppm.
  8. Un extracto de acuerdo con la reivindicación 6, CARACTERIZADO PORQUE tiene un contenido de 4'-O-metil piridoxina de menos de 2 ppm.
  9. Un extracto de acuerdo con cualquiera de las reivindicaciones 6 a 8, CARACTERIZADO PORQUE tiene un contenido de biflavonas de menos de 10 ppm.
  10. Un extracto de acuerdo con cualquiera de las reivindicaciones 6 a 8, CARACTERIZADO PORQUE tiene un contenido de biflavonas de menos de 5 ppm.
  11. Un extracto de acuerdo con cualquiera de las reivindicaciones 6 a 10, CARACTERIZADO PORQUE tiene los siguientes contenidos:
    - entre 20 y 30% en peso de flavonoides,
    - entre 4,5 y 8,5% en peso de lactonas de terpenos, y
    - menos de 10 ppm de ácidos ginkgólicos.
  12. Un extracto de acuerdo con cualquiera de las reivindicaciones 6 a 10, CARACTERIZADO PORQUE tiene los siguientes contenidos:
    - entre 22,0 y 27,0 % en peso de flavonoides,
    - entre 5,0 y 7,0% en peso de lactonas de terpenos, y
    - a lo sumo 5 ppm de ácidos ginkgólicos.
  13. Un extracto de acuerdo con cualquiera de las reivindicaciones 6 a 10, CARACTERIZADO PORQUE tiene los siguientes contenidos:
    - entre 22,0 y 27,0 % en peso de flavonoides,
    - entre 2,8 y 3,4% en peso de ginkgólidos A, B y C en total,
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- entre 2,6 y 3,2% en peso de bilobalida, y
  - a lo sumo 5 ppm de ácidos ginkgólicos.
14. El uso de un extracto de acuerdo con cualquiera de las reivindicaciones 6 a 13, CARACTERIZADO PORQUE es en la fabricación de un medicamento, un producto alimenticio u otra preparación para el tratamiento de la demencia y sus síntomas, y/o de trastornos de la circulación sanguínea cerebral o periférica.
15. Un medicamento, un producto alimenticio u otra preparación, CARACTERIZADA PORQUE tiene un contenido de un extracto de ginkgo de acuerdo con cualquiera de las reivindicaciones 6 a 13.

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**MÉTODO MEJORADO PARA PREPARAR EXTRACTOS DE GINKGO QUE  
TIENEN UN CONTENIDO BAJO DE 4'-O-METIL PIRIDOXINA Y/O  
BIFLAVONAS**

**RESUMEN:**

Método mejorado de pasos múltiples para preparar un extracto de ginkgo biloba que tiene un contenido reducido de 4'-O-metil piridoxina y/o biflavonas, donde la reducción del contenido se efectúa por filtración a través de una resina de adsorción y/o un intercambiador de iones, y las sustancias a eliminar se retienen en la resina. Extracto de ginkgo biloba que tiene un contenido reducido de 4'-O-metil piridoxina y/o biflavonas que puede obtenerse con el método de acuerdo con la invención y su uso.

# PATENT COOPERATION TREATY

## PCT

### INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

(Chapter II of the Patent Cooperation Treaty)

(PCT Article 36 and Rule 70)

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|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|----------------------------------------------------------|-----------------------------------------------------------------------------|----------------------|
| Applicant's or agent's file reference<br>15406WO/eI                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                | <b>FOR FURTHER ACTION</b>                                |                                                                             | See Form PCT/PEA/416 |
| International application No.<br>PCT/EP2006/004029                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                 | International filing date (day/month/year)<br>28.04.2006 | Priority date (day/month/year)<br>03.05.2005                                |                      |
| International Patent Classification (IPC) or national classification and IPC<br>INV. A61K36/00                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                     |                                                          |                                                                             |                      |
| Applicant<br>Dr. Willmar Schwabe GmbH & Co. KG                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                     |                                                          |                                                                             |                      |
| <p>1. This report is the international preliminary examination report, established by this International Preliminary Examining Authority under Article 35 and transmitted to the applicant according to Article 36.</p> <p>2. This REPORT consists of a total of <u>5</u> sheets, including this cover sheet.</p> <p>3. This report is also accompanied by ANNEXES, comprising:</p> <p>a. <input checked="" type="checkbox"/> sent to the applicant and to the International Bureau) a total of <u>3</u> sheets, as follows:</p> <p><input checked="" type="checkbox"/> sheets of the description, claims and/or drawings which have been amended and are the basis of this report and/or sheets containing rectifications authorized by this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions).</p> <p><input type="checkbox"/> sheets which supersede earlier sheets, but which this Authority considers contain an amendment that goes beyond the disclosure in the international application as filed, as indicated in item 4 of Box No. I and the Supplemental Box.</p> <p>b. <input type="checkbox"/> (sent to the International Bureau only) a total of (indicate type and number of electronic carrier(s)) , containing a sequence listing and/or tables related thereto, in electronic form only, as indicated in the Supplemental Box Relating to Sequence Listing (see Section 802 of the Administrative Instructions).</p> |                                                          |                                                                             |                      |
| <p>4. This report contains indications relating to the following items:</p> <p><input checked="" type="checkbox"/> Box No. I Basis of the report</p> <p><input type="checkbox"/> Box No. II Priority</p> <p><input type="checkbox"/> Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability</p> <p><input type="checkbox"/> Box No. IV Lack of unity of invention</p> <p><input checked="" type="checkbox"/> Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement</p> <p><input type="checkbox"/> Box No. VI Certain documents cited</p> <p><input type="checkbox"/> Box No. VII Certain defects in the international application</p> <p><input type="checkbox"/> Box No. VIII Certain observations on the international application</p>                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                |                                                          |                                                                             |                      |
| Date of submission of the demand<br><br>2007-03-02                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                 |                                                          | Date of completion of this report<br><br>13.04.2007                         |                      |
| Name and mailing address of the international preliminary examining authority:<br> European Patent Office<br>D-80298 Munich<br>Tel. +49 89 2399 - 0 Tx: 523656 epmu d<br>Fax: +49 89 2399 - 4465                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                |                                                          | Authorized officer<br><br>THALMAIR, M<br><br>Telephone No. +49 89 2399-2177 |                      |

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**INTERNATIONAL PRELIMINARY REPORT  
ON PATENTABILITY**

International application No.  
PCT/EP2006/004029

**Box No. I Basis of the report**

1. With regard to the **language**, this report is based on

- ☒ the international application in the language in which it was filed
- ☐ a translation of the international application into , which is the language of a translation furnished for the purposes of:
  - ☐ international search (under Rules 12.3(a) and 23.1(b))
  - ☐ publication of the international application (under Rule 12.4(a))
  - ☐ international preliminary examination (under Rules 55.2(a) and/or 55.3(a))

2. With regard to the **elements\*** of the international application, this report is based on *(replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report):*

**Description, Pages**

1-14 as originally filed

**Claims, Numbers**

1-14 received on 02.03.2007 with letter of 02.03.2007

- ☐ a sequence listing and/or any related table(s) - see Supplemental Box Relating to Sequence Listing

3. ☐ The amendments have resulted in the cancellation of:

- ☐ the description, pages
- ☐ the claims, Nos.
- ☐ the drawings, sheets/figs
- ☐ the sequence listing (*specify*):
- ☐ any table(s) related to sequence listing (*specify*):

4. ☐ This report has been established as if (some of) the amendments annexed to this report and listed below had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).

- ☐ the description, pages
- ☐ the claims, Nos.
- ☐ the drawings, sheets/figs
- ☐ the sequence listing (*specify*):
- ☐ any table(s) related to sequence listing (*specify*):

\* If item 4 applies, some or all of these sheets may be marked "superseded."

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**INTERNATIONAL PRELIMINARY REPORT  
ON PATENTABILITY**

International application No.  
PCT/EP2006/004029

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**Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement**

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1. Statement

|                               |             |             |
|-------------------------------|-------------|-------------|
| Novelty (N)                   | Yes: Claims | <u>1-14</u> |
|                               | No: Claims  |             |
| Inventive step (IS)           | Yes: Claims | <u>1-14</u> |
|                               | No: Claims  |             |
| Industrial applicability (IA) | Yes: Claims | <u>1-14</u> |
|                               | No: Claims  |             |

2. Citations and explanations (Rule 70.7):

see separate sheet

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Section V

Responsive to the Written Opinion of the International Searching Authority dated September 25, 2006, Applicant has submitted an amended set of claims 1 to 14 which forms the basis of this International Preliminary Report on Patentability.

In summary, documents D 1 to D 5 do not directly and unambiguously disclose or suggest to disclose an extract from leaves of Ginkgo biloba containing less than 10 ppm 4'-O-methyl pyridoxine and/or less than 20 ppm biflavones.

Reference to a content of less than 100 ppm biflavones and less than 50 ppm 4'-O-Methylpyridoxine (see page 2 lines 46-47 of D 1) comprises the range down to the limit of detection of the respective component at the priority date of **D 1**. It has to be interpreted in the light of the disclosure content of document **D 1** as a whole and has to be understood in the light of the specific Examples 2 and 3 of D 1 such that contents of less than 20 ppm biflavones and less than 10 ppm 4'-O-methyl pyridoxine are not comprised by the disclosure content of D 1. Document **D 2** is not closer to the present application than document D 1. D 2 does not relate to the preparation of an extract from Ginkgo biloba having a reduced content of 4'-O-methyl pyridoxine and/or biflavones.

**D 3** is an article by Ansgar Arenz et al. disclosing that 4'-O-methyl pyridoxine (ginkgotoxin) is a neurotoxic antivitamin B6 which occurs in Ginkgo biloba L. seeds. D 3 describes the preparative isolation of 4'-O-methyl pyridoxine from leaves. However, it neither teaches nor suggests as to how to reduce the content of 4'-O-methyl pyridoxine in an extract from Ginkgo biloba containing all the other desired active ingredients relevant for the activity of the extract from Ginkgo biloba.

According to **D 4** total Ginkgetin (biflavones) is extracted through alcohol extraction and water precipitation process and purified with macroporous adsorbing resin. Thus, rather than disclosing a method for preparing an extract from Ginkgo biloba containing all the desired active ingredients, however, having a reduced content of biflavones, D 4 discloses a method for the isolation of total ginkgetin and teaches the purification of total ginkgetin with macroporous adsorbing resin. It does not disclose or suggest to disclose the adsorption of biflavones on an adsorber resin in order to reduce the content thereof in an extract from Ginkgo biloba. Finally, document **D 5** teaches a method for preparing an extract from Ginkgo biloba being constituted by a dimeric flavonoids fraction i.e. biflavones fraction. Thus, rather than suggesting a method for preparing an extract from Ginkgo biloba having a reduced content of biflavones, D 5 suggests to enrich the biflavones content in the extracts in order to

**INTERNATIONAL PRELIMINARY  
REPORT ON PATENTABILITY  
(SEPARATE SHEET)**

International application No.

PCT/EP2006/004029

provide new active products useful in therapy of disturbances of the cerebral and peripheral circulation, etc..

## Claims

1. Method for producing an extract from ginkgo biloba having a reduced content of 4'-O-methyl pyridoxine and/or biflavones, characterized by the following steps of the method:

a) extracting leaves of ginkgo biloba using water-containing acetone, a water-containing alkanol having 1 to 3 carbon atoms or anhydrous methanol,

b) separating the organic solvent by concentrating the solvent to a content of 10% by weight at the most, wherein water may be added during the final distillation steps,

c) diluting the remaining aqueous solution with water to a solids content of 5 to 25% by weight, cooling to a temperature below 25°C and allowing to stand until a precipitate is formed,

d) treating the remaining aqueous solution with ammonium sulfate and subsequently extracting at least once using methyl ethyl ketone or a mixture of methyl ethyl ketone and acetone,

e) concentrating the extract obtained and diluting with an ethanol-water mixture to obtain a solution containing 50% by weight water and 50% by weight ethanol at a solids content of 10% by weight,

f) treating the solution with a lead compound or an insoluble polyamide,

g) extracting the filtered solution using an aliphatic or cycloaliphatic solvent having a boiling point of 60 to 100°C,

h) concentrating the remaining aqueous-alcoholic solution, followed by treating with ammonium sulfate and extracting with methyl ethyl ketone and ethanol,

i) concentrating the organic phase obtained to a solids content of 50 to 70% by weight,

k) dissolving the concentrate in an aqueous alkanol having 1 to 3 carbon atoms and optionally filtering,

l) filtration over an adsorber resin to reduce the content of biflavones and/or an ion exchanger to reduce the content of 4'-O-methyl pyridoxine, wherein the substances to be removed are retained on the resin and

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m) drying the concentrate at reduced pressure to obtain a dry extract having a water content of less than 5%.

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2. Method according to claim 1, wherein the aqueous alkanol in step k) is aqueous ethanol.
3. Method according to claim 2, wherein the aqueous ethanol is ethanol having a content of 40 to 60% by weight.
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4. Method according to any one of claims 1 to 3, wherein the adsorber resin in step l) is a resin based on optionally substituted styrenes/divinylbenzenes.
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5. Method according to any one of claims 1 to 3, wherein the ion exchanger in step l) is a strongly acidic ion exchanger.
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6. Extract from leaves of ginkgo biloba, characterized by having the following contents:
- less than 10 ppm 4'-O-methyl pyridoxine and/or
  - less than 20 ppm biflavones selected from the group comprising amentoflavone, bilobetin, ginkgetin, isoginkgetin and sciadopitysin.
- 25
7. Extract according to claim 6, characterized by having a content of less than 2 ppm 4'-O-methyl pyridoxine.
- 30
8. Extract according to claim 6 or 7, characterized by having a content of less than 10 ppm biflavones.
9. Extract according to claim 6 or 7, characterized by having a content of less than 5 ppm biflavones.

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10. Extract according to any of claims 6 to 9, characterized by having the following contents:
- 20 to 30% by weight flavonoids, selected from the group comprising quercetin, kaempferol and isorhamnetin glycosides,
  - 4.5 to 8.5% by weight terpene lactones and
  - less than 10 ppm ginkgolic acids.
11. Extract according to any of claims 6 to 9, characterized by having the following contents:
- 22.0 to 27.0 % by weight flavonoids, selected from the group comprising quercetin, kaempferol and isorhamnetin glycosides,
  - 5.0 to 7.0% by weight terpene lactones and
  - 5 ppm ginkgolic acids at the most.
12. Extract according to any of claims 6 to 9, characterized by having the following contents:
- 22.0 to 27.0 % by weight flavonoids, selected from the group comprising quercetin, kaempferol and isorhamnetin glycosides,
  - 2.8 to 3.4% by weight ginkgolides A, B and C in total,
  - 2.6 to 3.2% by weight bilobalid and
  - 5 ppm ginkgolic acids at the most.
13. Use of an extract according to any one of claims 6 to 12 for the manufacture of a medicament, food product or other preparation for the treatment of dementia and symptoms thereof and/or cerebral or peripheral blood circulation disorders.
14. Medicament, food product or other preparation, characterized by a content of a ginkgo extract according to any one of claims 6 to 12.

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## REIVINDICACIONES:

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1. Un método para producir un extracto de ginkgo biloba que tiene un contenido reducido de 4'-O-metil piridoxina y/o biflavonas, CARACTERIZADO PORQUE comprende los siguientes pasos:
  - a) extraer hojas de ginkgo biloba usando acetona con agua, un alcohol que contiene agua con entre 1 y 3 átomos de carbono, o metanol anhidro,
  - b) separar el solvente orgánico concentrando el solvente hasta obtener un contenido de a lo sumo 10% en peso, donde puede agregarse agua durante los pasos de destilación finales,
  - c) diluir la solución acuosa remanente con agua hasta obtener un contenido de sólidos de entre 5 y 25% en peso, enfriar a una temperatura inferior a 25°C, y permitir el reposo hasta la formación de un precipitado,
  - d) tratar la solución acuosa remanente con sulfato de amonio y luego realizar al menos una extracción usando metil etil cetona o una mezcla de metil etil cetona y acetona,
  - e) concentrar el extracto obtenido y diluir con una mezcla de etanol y agua para obtener una solución que contiene 50% en peso de agua y 50% en peso de etanol, con un contenido de sólidos de 10% en peso,
  - f) tratar la solución con un compuesto de plomo o una poliamida insoluble,
  - g) extraer la solución filtrada usando un solvente alifático o cicloalifático con un punto de fusión de entre 60 y 100°C,
  - h) concentrar la solución acuosa alcohólica remanente, luego realizar un tratamiento con sulfato de amonio y una extracción con metil etil cetona y etanol,
  - i) concentrar la fase orgánica obtenida hasta obtener un contenido de sólidos de entre 50 y 70% en peso,
  - k) disolver el concentrado en un alcohol acuoso que tiene entre 1 y 3 átomos de carbono, y opcionalmente filtrar,

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- l) filtrar sobre una resina de adsorción para reducir el contenido de biflavones y/o un intercambiador de iones para reducir el contenido de 4'-O-metil piridoxina, donde las sustancias a eliminar se retienen en la resina, y
- m) secar el concentrado a presión reducida para obtener un extracto seco que tiene un contenido de agua menor que 5%.
2. Un método de acuerdo con la reivindicación 1, CARACTERIZADO PORQUE el alcohol acuoso en el paso k) es etanol acuoso.
  3. Un método de acuerdo con la reivindicación 2, CARACTERIZADO PORQUE el etanol acuoso es etanol con un contenido de entre 40 y 60% en peso.
  4. Un método de acuerdo con cualquiera de las reivindicaciones 1 a 3, CARACTERIZADO PORQUE la resina de adsorción del paso l) es una resina basada en estirenos/divinilbencenos opcionalmente sustituidos.
  5. Un método de acuerdo con cualquiera de las reivindicaciones 1 a 3, CARACTERIZADO PORQUE el intercambiador de iones del paso l) es un intercambiador de iones fuertemente ácido.
  6. Un extracto de hojas de ginkgo biloba CARACTERIZADO PORQUE tiene los siguientes contenidos:
    - menos de 10 ppm de 4'-O-metil piridoxina y/o
    - menos de 20 ppm de biflavonas seleccionadas del grupo que comprende amentoflavona, bilobetina, ginkgetin, isoginkgetina y siadopitisina.
  7. Un extracto de acuerdo con la reivindicación 6, CARACTERIZADO PORQUE tiene un contenido de 4'-O-metil piridoxina de menos de 2 ppm.
  8. Un extracto de acuerdo con las reivindicaciones 6 o 7, CARACTERIZADO PORQUE tiene un contenido de biflavones de menos de 10 ppm.
  9. Un extracto de acuerdo con las reivindicaciones 6 o 7, CARACTERIZADO PORQUE tiene un contenido de biflavonas de menos de 5 ppm.

10. Un extracto de acuerdo con cualquiera de las reivindicaciones 6 a 9, CARACTERIZADO PORQUE tiene los siguientes contenidos:
- entre 20 y 30% en peso de flavonoides, seleccionados del grupo que comprende cuercetina, kaempferol y glicosidas isorhamnetin,
  - entre 4,5 y 8,5% en peso de lactonas de terpenos, y
  - menos de 10 ppm de ácidos ginkgólicos.
11. Un extracto de acuerdo con cualquiera de las reivindicaciones 6 a 9, CARACTERIZADO PORQUE tiene los siguientes contenidos:
- entre 22,0 y 27,0 % en peso de flavonoides, seleccionados del grupo que comprende cuercetina, kaempferol y glicosidas isorhamnetin,
  - entre 5,0 y 7,0% en peso de lactonas de terpenos, y
  - a lo sumo 5 ppm de ácidos ginkgólicos.
12. Un extracto de acuerdo con cualquiera de las reivindicaciones 6 a 9, CARACTERIZADO PORQUE tiene los siguientes contenidos:
- entre 22,0 y 27,0 % en peso de flavonoides, seleccionados del grupo que comprende cuercetina, kaempferol y glicosidas isorhamnetin,
  - entre 2,8 y 3,4% en peso de ginkgólidos A, B y C en total,
  - entre 2,6 y 3,2% en peso de bilobalida, y
  - a lo sumo 5 ppm de ácidos ginkgólicos.
13. El uso de un extracto de acuerdo con cualquiera de las reivindicaciones 6 a 12, CARACTERIZADO PORQUE es en la fabricación de un medicamento, un producto alimenticio u otra preparación para el tratamiento de la demencia y sus síntomas, y/o de trastornos de la circulación sanguínea cerebral o periférica.
14. Un medicamento, un producto alimenticio u otra preparación, CARACTERIZADA PORQUE tiene un contenido de un extracto de ginkgo de acuerdo con cualquiera de las reivindicaciones 6 a 12.

# PATENT COOPERATION TREATY

From the  
INTERNATIONAL SEARCHING AUTHORITY

## PCT

To:

see form PCT/ISA/220

### WRITTEN OPINION OF THE INTERNATIONAL SEARCHING AUTHORITY (PCT Rule 43bis.1)

Date of mailing  
(day/month/year) see form PCT/ISA/210 (second sheet)

Applicant's or agent's file reference  
see form PCT/ISA/220

**FOR FURTHER ACTION**  
See paragraph 2 below

International application No.  
PCT/EP2006/004029

International filing date (day/month/year)  
28.04.2006

Priority date (day/month/year)  
03.05.2005

International Patent Classification (IPC) or both national classification and IPC  
INV. A61K36/00

Applicant  
BIOPLANTA ARZNEIMITTEL GMBH

**1. This opinion contains indications relating to the following items:**

- ☒ Box No. I Basis of the opinion
- ☐ Box No. II Priority
- ☐ Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
- ☐ Box No. IV Lack of unity of invention
- ☒ Box No. V Reasoned statement under Rule 43bis.1(a)(i) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
- ☐ Box No. VI Certain documents cited
- ☐ Box No. VII Certain defects in the international application
- ☒ Box No. VIII Certain observations on the international application

**2. FURTHER ACTION**

If a demand for international preliminary examination is made, this opinion will usually be considered to be a written opinion of the International Preliminary Examining Authority ("IPEA") except that this does not apply where the applicant chooses an Authority other than this one to be the IPEA and the chosen IPEA has notified the International Bureau under Rule 66.1bis(b) that written opinions of this International Searching Authority will not be so considered.

If this opinion is, as provided above, considered to be a written opinion of the IPEA, the applicant is invited to submit to the IPEA a written reply together, where appropriate, with amendments, before the expiration of 3 months from the date of mailing of Form PCT/ISA/220 or before the expiration of 22 months from the priority date, whichever expires later.

For further options, see Form PCT/ISA/220.

**3. For further details, see notes to Form PCT/ISA/220.**

Name and mailing address of the ISA:



European Patent Office  
D-80298 Munich  
Tel. +49 89 2399 - 0 Tx: 523656 epmu d  
Fax: +49 89 2399 - 4465

Date of completion of  
this opinion

see form  
PCT/ISA/210

Authorized Officer

THALMAIR, M

Telephone No. +49 89 2399-2177



**WRITTEN OPINION OF THE  
INTERNATIONAL SEARCHING AUTHORITY**

International application No.  
PCT/EP2006/004029

**Box No. I Basis of the opinion**

1. With regard to the **language**, this opinion has been established on the basis of:
  - ☒ the international application in the language in which it was filed
  - ☐ a translation of the international application into , which is the language of a translation furnished for the purposes of international search (Rules 12.3(a) and 23.1 (b)).
2. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application and necessary to the claimed invention, this opinion has been established on the basis of:
  - a. type of material:
    - ☐ a sequence listing
    - ☐ table(s) related to the sequence listing
  - b. format of material:
    - ☐ on paper
    - ☐ in electronic form
  - c. time of filing/furnishing:
    - ☐ contained in the international application as filed.
    - ☐ filed together with the international application in electronic form.
    - ☐ furnished subsequently to this Authority for the purposes of search.
3. ☐ In addition, in the case that more than one version or copy of a sequence listing and/or table relating thereto has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.
4. Additional comments:

**WRITTEN OPINION OF THE  
INTERNATIONAL SEARCHING AUTHORITY**

International application No.  
PCT/EP2006/004029

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**Box No. V Reasoned statement under Rule 43bis.1(a)(i) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement**

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1. Statement

|                               |             |      |
|-------------------------------|-------------|------|
| Novelty (N)                   | Yes: Claims | 1-5  |
|                               | No: Claims  | 6-15 |
| Inventive step (IS)           | Yes: Claims |      |
|                               | No: Claims  | 1-15 |
| Industrial applicability (IA) | Yes: Claims | 1-15 |
|                               | No: Claims  |      |

2. Citations and explanations

**see separate sheet**

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**Box No. VIII Certain observations on the international application**

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The following observations on the clarity of the claims, description, and drawings or on the question whether the claims are fully supported by the description, are made:

**see separate sheet**

## Section VIII

Having regard to claim 1, it is pointed out to the Applicant that it is not clear whether in step I) the filtration over an adsorber resin or over an ion exchanger leads to the reduced content of 4'-O-methyl pyridoxine or to the reduced content of biflavones, respectively. This should be clarified.

Moreover, concerning claims 6-15, it is indicated that the term "flavonoids" used in claims 11-13 normally encompasses also "biflavones", see claim 6.

According to pages 2 and 4 of the description, Applicant seems to mean under "biflavones" amentoflavone, bilobetin, ginkgetin, isoginkgetin and sciadopitysin and under "flavonoids" quercetin, kaempferol and isorhamnetin calculated as flavonoid glycosides.

Applicant is asked to remove said unclarity / ambiguity in the said claims.

## Section V

The subject-matter of claims 6-15 is not novel, since **D 1** (EP 1037 646) already reports on extracts from leaves of *Ginkgo biloba* comprising (in wt.%) flavonol glycoside (20-30); ginkgolide A, B, C and J (2.5-4.5); bilobalid (2.0-4.0); alkyl phenols such as ginkgolic acids (at less than 10 ppm); proanthocyanidins (less than 10); 4'-O-methylpyridoxine (less than 50 ppm) and biflavone (less than 10 ppm), and their inclusion into medicaments for the treatment of disorders of the brain, arterial peripheral blood circulation disorders etc.

The subject-matter of claims 1-5 is not considered as involving inventive merits.

**D 2** (DE 394091), the total disclosure of which, in particular with respect to the details of the method, is incorporated into the present application, describes already steps a) to i) as well as m) of the claimed method.

**D 3** (Arenz Ansgar et al.) discloses that 4'-O-methylpyridoxine is a neurotoxic antivitamin B 6 which occurs in *Ginkgo biloba* seeds and is also present in *Ginkgo* medications. Preparative isolation of the toxin is possible by passing the acidified extract over a cation exchange column.

**D 4** (WPI abstract of CN1557 455) reports that total ginkgetin (biflavone) is extracted through alcohol extraction and water precipitation process and purified with macroporous adsorbing resin. Through mixing total ginkgetin and total bilobalide the ginkgo-dipyridamole powder for injection is prepared and has higher pharmacological effect.

**D 5** (EP 0360 556) shows a process for extracting flavanoids from an aqueous extract of *Ginkgo biloba* which comprises contacting the extract with an absorption resin and eluting absorbed flavanoids with an organic solvent.

Hence, the object as defined on page 3, line 18 ff of the present application was solved by the skilled person in an obvious manner by combining the teachings of **D 2** with **D 3** or with **D 4** and/or **D 5**.

**D 6** (Kressmann et al.) mentions that most manufacturers of *Ginkgo* preparations refer to the standards published by the German Commission E. Accordingly, the extract should contain 22-27% flavone glycosides, 5-7% terpene lactones (2.8-3.4% ginkgolides A, B and C and 2.6-3.2% bilobalide thereof) and not more than 5 ppm ginkgolic acids.

Pharmacological and clinical studies reported in the review **D 7** (van Beek) confirm the therapeutic value and the good tolerability of the standardized extract of *G. biloba* leaves in the treatment of disorders of cerebral function and peripheral vascular insufficiency. Terpene trilactones (ginkgolides and bilobalide) and flavonoids are likely responsible for its biological efficacy.

# PATENT COOPERATION TREATY

# PCT

## INTERNATIONAL SEARCH REPORT

(PCT Article 18 and Rules 43 and 44)

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|                                                         |                                                                                                      |                                                             |
|---------------------------------------------------------|------------------------------------------------------------------------------------------------------|-------------------------------------------------------------|
| Applicant's or agent's file reference<br><br>15406WO/e1 | <b>FOR FURTHER ACTION</b><br><br>see Form PCT/ISA/220<br>as well as, where applicable, item 5 below. |                                                             |
| International application No.<br><br>PCT/EP2006/004029  | International filing date (day/month/year)<br><br>28/04/2006                                         | (Earliest) Priority Date (day/month/year)<br><br>03/05/2005 |
| Applicant<br><br>BIOPLANTA ARZNEIMITTEL GMBH            |                                                                                                      |                                                             |

This international search report has been prepared by this International Searching Authority and is transmitted to the applicant according to Article 18. A copy is being transmitted to the International Bureau.

This international search report consists of a total of 6 sheets.

☒ It is also accompanied by a copy of each prior art document cited in this report.

**1. Basis of the report**

a. With regard to the **language**, the international search was carried out on the basis of:

- ☒ the international application in the language in which it was filed  
☐ a translation of the international application into \_\_\_\_\_, which is the language of a translation furnished for the purposes of international search (Rules 12.3(a) and 23.1(b))

b. ☐ With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, see Box No. I.

2. ☒ **Certain claims were found unsearchable** (See Box No. II)

3. ☐ **Unity of invention is lacking** (see Box No. III)

4. With regard to the **title**,

- ☒ the text is approved as submitted by the applicant  
☐ the text has been established by this Authority to read as follows:

5. With regard to the **abstract**,

- ☒ the text is approved as submitted by the applicant  
☐ the text has been established, according to Rule 38.2(b), by this Authority as it appears in Box No. IV. The applicant may, within one month from the date of mailing of this international search report, submit comments to this Authority

6. With regard to the **drawings**,

a. the figure of the **drawings** to be published with the abstract is Figure No. \_\_\_\_\_

- ☐ as suggested by the applicant  
☐ as selected by this Authority, because the applicant failed to suggest a figure  
☐ as selected by this Authority, because this figure better characterizes the invention

b. ☐ none of the figures is to be published with the abstract

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## INTERNATIONAL SEARCH REPORT

International application No

PCT/EP2006/004029

## A. CLASSIFICATION OF SUBJECT MATTER

INV. A61K36/00

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 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 practical, search terms used)

EPO-Internal, WPI Data, BIOSIS, MEDLINE, EMBASE

## C. DOCUMENTS CONSIDERED TO BE RELEVANT

| Category* | Citation of document, with indication, where appropriate, of the relevant passages                                                                                                                                                                                  | Relevant to claim No. |
|-----------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-----------------------|
| X         | EP 1 037 646 A (DR. WILLMAR SCHWABE GMBH & CO) 27 September 2000 (2000-09-27)<br>claims 1,2,6,7                                                                                                                                                                     | 6-15                  |
| Y         | DE 39 40 091 A1 (DR. WILLMAR SCHWABE GMBH & CO, 7500 KARLSRUHE, DE)<br>6 June 1991 (1991-06-06)<br>the whole document                                                                                                                                               | 1-5                   |
| Y         | ARENZ ANSGAR ET AL: "Occurrence of neurotoxic 4'-O-methylpyridoxine in Ginkgo biloba leaves, Ginkgo medications and Japanese Ginkgo food"<br>PLANTA MEDICA,<br>vol. 62, no. 6, 1996, pages 548-551,<br>XP009070428<br>ISSN: 0032-0943<br>page 549, left-hand column | 1-5                   |

☒ 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 document 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

11 September 2006

Date of mailing of the international search report

25/09/2006

Name and mailing address of the ISA/

European Patent Office, P.B. 5618 Patentlaan 2  
NL - 2280 HV Rijswijk  
Tel. (+31-70) 340-2040, Tx. 31 651 epo nl,  
Fax: (+31-70) 340-3016

Authorized officer

THALMAIR, M

## INTERNATIONAL SEARCH REPORT

International application No  
PCT/EP2006/004029

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| C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT |                                                                                                                                                                                                                                                            |                       |
|------------------------------------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-----------------------|
| Category*                                            | Citation of document, with indication, where appropriate, of the relevant passages                                                                                                                                                                         | Relevant to claim No. |
| Y                                                    | DATABASE WPI<br>Section Ch, Week 200525<br>Derwent Publications Ltd., London, GB;<br>Class B04, AN 2005-234134<br>XP002398187<br>& CN 1 557 455 A (ZHANG Z)<br>29 December 2004 (2004-12-29)<br>abstract<br>-----                                          | 1-5                   |
| Y                                                    | EP 0 360 556 A (INDENA S.P.A)<br>28 March 1990 (1990-03-28)<br>claims 1,15; example 3<br>-----                                                                                                                                                             | 1-5                   |
| A                                                    | KRESSMANN S ET AL: "Pharmaceutical quality<br>of different Ginkgo biloba brands"<br>May 2002 (2002-05), JOURNAL OF PHARMACY<br>AND PHARMACOLOGY, VOL. 54, NR. 5, PAGE(S)<br>661-669, XP009070389<br>ISSN: 0022-3573<br>page 662, left-hand column<br>----- | 12,13                 |
| A                                                    | BEEK VAN T A: "GINKGO BILOBA L"<br>FITOTERAPIA, IDB HOLDING, MILAN, IT,<br>vol. I.XIX, no. 3, 1998, pages 195-244,<br>XP000700334<br>ISSN: 0367-326X<br>page 198; table 1<br>-----                                                                         | 1-15                  |

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Continuation of Box II.2

Claims Nos.: -

It is pointed out to the Applicant concerning claims 6-15, that the term "flavonoids" used in claims 11-13 normally encompasses also "biflavones", see claim 6. According to pages 2 and 4 of the description, Applicant means under "biflavones" amentoflavone, bilobetin, ginkgetin, isoginkgetin and sciadopitysin and under "flavonoids" quercetin, kaempferol and isorhamnetin calculated as flavonoid glycosides.

Applicant is asked to remove said unclarity / ambiguity.

Moreover, having regard to claim 1, it is indicated that it is not clear whether in step 1) the filtration over an adsorber resin or over an ion exchanger leads to the reduced content of 4'-O-methyl pyridoxine or to the reduced content of biflavones, respectively. This should be clarified.

The applicant's attention is drawn to the fact that claims relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure. If the application proceeds into the regional phase before the EPO, the applicant is reminded that a search may be carried out during examination before the EPO (see EPO Guideline C-VI, 8.5), should the problems which led to the Article 17(2) declaration be overcome.

# INTERNATIONAL SEARCH REPORT

International application No.  
PCT/EP2006/004029

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## Box 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:  
see FURTHER INFORMATION sheet PCT/ISA/210
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 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:

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 fee, this Authority did not invite payment of any additional fee.
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.:

### Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☐ No protest accompanied the payment of additional search fees.

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# INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No

PCT/EP2006/004029

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| Patent document<br>cited in search report |    | Publication<br>date | Patent family<br>member(s) | Publication<br>date |
|-------------------------------------------|----|---------------------|----------------------------|---------------------|
| EP 1037646                                | A  | 27-09-2000          | CN 1282252 A               | 31-01-2001          |
|                                           |    |                     | WO 9932129 A1              | 01-07-1999          |
|                                           |    |                     | DE 19756848 A1             | 01-07-1999          |
|                                           |    |                     | JP 2001526235 T            | 18-12-2001          |
|                                           |    |                     | US 6328999 B1              | 11-12-2001          |
| DE 3940091                                | A1 | 06-06-1991          | CA 2031385 A1              | 05-06-1991          |
|                                           |    |                     | DK 431535 T3               | 28-03-1994          |
|                                           |    |                     | EP 0431535 A1              | 12-06-1991          |
|                                           |    |                     | ES 2024400 T3              | 16-04-1994          |
|                                           |    |                     | GR 91300122 T1             | 20-03-1992          |
|                                           |    |                     | JP 2045722 C               | 25-04-1996          |
|                                           |    |                     | JP 3279331 A               | 10-12-1991          |
|                                           |    |                     | JP 7076176 B               | 16-08-1995          |
|                                           |    |                     | KR 154977 B1               | 16-11-1998          |
| CN 1557455                                | A  | 29-12-2004          | NONE                       |                     |
| EP 0360556                                | A  | 28-03-1990          | CA 1337973 C               | 23-01-1996          |
|                                           |    |                     | DE 68906108 D1             | 27-05-1993          |
|                                           |    |                     | DE 68906108 T2             | 29-07-1993          |
|                                           |    |                     | ES 2054018 T3              | 01-08-1994          |
|                                           |    |                     | GR 3007688 T3              | 31-08-1993          |
|                                           |    |                     | HK 43694 A                 | 13-05-1994          |
|                                           |    |                     | JP 2121998 A               | 09-05-1990          |
|                                           |    |                     | JP 2800040 B2              | 21-09-1998          |
|                                           |    |                     | KR 169957 B1               | 01-02-1999          |

From the INTERNATIONAL BUREAU

**PCT**NOTIFICATION OF THE RECORDING  
OF A CHANGE(PCT Rule 92bis.1 and  
Administrative Instructions, Section 422)

To:

ADAM, Holger  
Kraus & Weisert  
Thomas-Wimmer-Ring 15  
80539 München  
ALLEMAGNE**EINGEGANGEN**

29. Dez. 2006

Patentanwälte  
**KRAUS & WEISERT**

|                                                                   |                                                                          |
|-------------------------------------------------------------------|--------------------------------------------------------------------------|
| Date of mailing (day/month/year)<br>21 December 2006 (21.12.2006) | <b>IMPORTANT NOTIFICATION</b>                                            |
| Applicant's or agent's file reference<br>15406WO/el               |                                                                          |
| International application No.<br>PCT/EP2006/004029                | International filing date (day/month/year)<br>28 April 2006 (28.04.2006) |

## 1. The following indications appeared on record concerning:

☒ the applicant
                     ☐ the inventor
                     ☐ the agent
                     ☐ the common representative

Name and Address

 BIOPLANTA ARZNEIMITTEL GMBH  
 Bahnhofstr. 5  
 76275 Ettlingen  
 Germany

State of Nationality

DE

State of Residence

DE

Telephone No.

Facsimile No.

Teleprinter No.

## 2. The International Bureau hereby notifies the applicant that the following change has been recorded concerning:

☒ the person
                     ☐ the name
                     ☐ the address
                     ☐ the nationality
                     ☐ the residence

Name and Address

 DR. WILLMAR SCHWABE GMBH & CO. KG  
 Willmar-Schwabe-Strasse 4  
 76227 Karlsruhe  
 Germany

State of Nationality

DE

State of Residence

DE

Telephone No.

Facsimile No.

Teleprinter No.

## 3. Further observations, if necessary:

## 4. A copy of this notification has been sent to:

☒ the receiving Office
                     ☒ the designated Offices concerned  
☐ the International Searching Authority
                     ☐ the elected Offices concerned  
☐ the International Preliminary Examining Authority
                     ☐ other:

 The International Bureau of WIPO  
 34, chemin des Colombettes  
 1211 Geneva 20, Switzerland

Authorized officer

**Suaton Sophie**

Facsimile No. +41 22 338 82 70

Facsimile No. +41 22 338 89 75

Telephone No. +41 22 338 70 89

mi

From the INTERNATIONAL BUREAU

**PCT**NOTIFICATION CONCERNING  
SUBMISSION OR TRANSMITTAL  
OF PRIORITY DOCUMENT

(PCT Administrative Instructions, Section 411)

To:

ADAM, Holger  
Kraus & Weisert  
Thomas-Wimmer-Ring 15  
80539 München  
ALLEMAGNE

EINGEGANGEN

31. Juli 2006

Patentanwälte  
KRAUS & WEISERT

|                                                                      |                                                                          |
|----------------------------------------------------------------------|--------------------------------------------------------------------------|
| Date of mailing (day/month/year)<br>20 July 2006 (20.07.2006)        | IMPORTANT NOTIFICATION                                                   |
| Applicant's or agent's file reference<br>15406WO/el                  |                                                                          |
| International application No.<br>PCT/EP2006/004029                   | International filing date (day/month/year)<br>28 April 2006 (28.04.2006) |
| International publication date (day/month/year)<br>Not yet published | Priority date (day/month/year)<br>03 May 2005 (03.05.2005)               |
| Applicant<br>BIOPLANTA ARZNEIMITTEL GMBH et al                       |                                                                          |

- By means of this Form, which replaces any previously issued notification concerning submission or transmittal of priority documents, the applicant is hereby notified of the date of receipt by the International Bureau of the priority document(s) relating to all earlier application(s) whose priority is claimed. Unless otherwise indicated by the letters "NR", in the right-hand column or by an asterisk appearing next to a date of receipt, the priority document concerned was submitted or transmitted to the International Bureau in compliance with Rule 17.1(a) or (b).
- (If applicable)* The letters "NR" appearing in the right-hand column denote a priority document which, on the date of mailing of this Form, had not yet been received by the International Bureau under Rule 17.1(a) or (b). Where, under Rule 17.1(a), the priority document must be submitted by the applicant to the receiving Office or the International Bureau, but the applicant fails to submit the priority document within the applicable time limit under that Rule, the attention of the applicant is directed to Rule 17.1(c) which provides that no designated Office may disregard the priority claim concerned before giving the applicant an opportunity, upon entry into the national phase, to furnish the priority document within a time limit which is reasonable under the circumstances.
- (If applicable)* An asterisk (\*) appearing next to a date of receipt, in the right-hand column, denotes a priority document submitted or transmitted to the International Bureau but not in compliance with Rule 17.1(a) or (b) (the priority document was received after the time limit prescribed in Rule 17.1(a) or the request to prepare and transmit the priority document was submitted to the receiving Office after the applicable time limit under Rule 17.1(b)). Even though the priority document was not furnished in compliance with Rule 17.1(a) or (b), the International Bureau will nevertheless transmit a copy of the document to the designated Offices, for their consideration. In case such a copy is not accepted by the designated Office as the priority document, Rule 17.1(c) provides that no designated Office may disregard the priority claim concerned before giving the applicant an opportunity, upon entry into the national phase, to furnish the priority document within a time limit which is reasonable under the circumstances.

| Priority date                   | Priority application No. | Country or regional Office<br>or PCT receiving Office | Date of receipt<br>of priority document |
|---------------------------------|--------------------------|-------------------------------------------------------|-----------------------------------------|
| 03 May 2005 (03.05.2005) ✓      | 10 2005 020 685.9 ✓      | DE ✓                                                  | 12 June 2006 (12.06.2006)               |
| 23 December 2005 (23.12.2005) ✓ | 10 2005 061 948.7 ✓      | DE ✓                                                  | 12 June 2006 (12.06.2006)               |

The International Bureau of WIPO  
34, chemin des Colombettes  
1211 Geneva 20, Switzerland

Facsimile No. +41 22 338 82 70

Authorized officer

Ellen Moyse

Facsimile No. +41 22 338 89 75

Telephone No. +41 22 338 91 54

## PCT REQUEST

Original (for SUBMISSION)

|         |                                                                                                                                         |                                                                                                                |
|---------|-----------------------------------------------------------------------------------------------------------------------------------------|----------------------------------------------------------------------------------------------------------------|
| 0       | For receiving Office use only                                                                                                           |                                                                                                                |
| 0-1     | International Application No.                                                                                                           | PCT/EP 2006 / 0 0 4 0 2 9                                                                                      |
| 0-2     | International Filing Date                                                                                                               | 28 APR 2006 ( 28. 04. 06 ) ✓                                                                                   |
| 0-3     | Name of receiving Office and "PCT International Application"                                                                            | RO/EP                                                                                                          |
| 0-4     | Form PCT/RO/101 PCT Request                                                                                                             |                                                                                                                |
| 0-4-1   | Prepared Using                                                                                                                          | PCT-SAFE [EASY model]<br>Version 3.51.005.180 MT/FOP<br>20060401/0.20.4rc.2.7                                  |
| 0-5     | Petition<br>The undersigned requests that the present international application be processed according to the Patent Cooperation Treaty |                                                                                                                |
| 0-6     | Receiving Office (specified by the applicant)                                                                                           | European Patent Office (EPO) (RO/EP)                                                                           |
| 0-7     | Applicant's or agent's file reference                                                                                                   | 15406WO/el                                                                                                     |
| I       | Title of Invention                                                                                                                      | IMPROVED METHOD FOR PREPARING GINKGO EXTRACTS HAVING A LOW CONTENT OF 4'-O-METHYL PYRIDOXINE AND/OR BIFLAVONES |
| II      | Applicant                                                                                                                               |                                                                                                                |
| II-1    | This person is                                                                                                                          | applicant only                                                                                                 |
| II-2    | Applicant for                                                                                                                           | all designated States except US                                                                                |
| II-4    | Name                                                                                                                                    | BIOPLANTA ARZNEIMITTEL GMBH                                                                                    |
| II-5    | Address                                                                                                                                 | Bahnhofstr. 5<br>76275 Ettlingen<br>Germany                                                                    |
| II-6    | State of nationality                                                                                                                    | DE                                                                                                             |
| II-7    | State of residence                                                                                                                      | DE                                                                                                             |
| III-1   | Applicant and/or inventor                                                                                                               |                                                                                                                |
| III-1-1 | This person is                                                                                                                          | applicant and inventor                                                                                         |
| III-1-2 | Applicant for                                                                                                                           | US only                                                                                                        |
| III-1-4 | Name (LAST, First)                                                                                                                      | ERDELMEIER, Clemens                                                                                            |
| III-1-5 | Address                                                                                                                                 | Glogauer Str. 32<br>76139 Karlsruhe<br>Germany                                                                 |
| III-1-6 | State of nationality                                                                                                                    | DE                                                                                                             |
| III-1-7 | State of residence                                                                                                                      | DE                                                                                                             |

## PCT REQUEST

Original (for SUBMISSION)

|         |                                                                                                                                                            |                                                                      |
|---------|------------------------------------------------------------------------------------------------------------------------------------------------------------|----------------------------------------------------------------------|
| III-2   | <b>Applicant and/or inventor</b>                                                                                                                           |                                                                      |
| III-2-1 | This person is                                                                                                                                             | applicant and inventor                                               |
| III-2-2 | Applicant for                                                                                                                                              | US only                                                              |
| III-2-4 | Name (LAST, First)                                                                                                                                         | HAUER, Hermann                                                       |
| III-2-5 | Address                                                                                                                                                    | Trollingerstr. 7<br>76228 Karlsruhe<br>Germany                       |
| III-2-6 | State of nationality                                                                                                                                       | DE                                                                   |
| III-2-7 | State of residence                                                                                                                                         | DE                                                                   |
| III-3   | <b>Applicant and/or inventor</b>                                                                                                                           |                                                                      |
| III-3-1 | This person is                                                                                                                                             | applicant and inventor                                               |
| III-3-2 | Applicant for                                                                                                                                              | US only                                                              |
| III-3-4 | Name (LAST, First)                                                                                                                                         | KOCH, Egon                                                           |
| III-3-5 | Address                                                                                                                                                    | Am Gießbach 11a<br>76229 Karlsruhe<br>Germany                        |
| III-3-6 | State of nationality                                                                                                                                       | DE                                                                   |
| III-3-7 | State of residence                                                                                                                                         | DE                                                                   |
| III-4   | <b>Applicant and/or inventor</b>                                                                                                                           |                                                                      |
| III-4-1 | This person is                                                                                                                                             | applicant and inventor                                               |
| III-4-2 | Applicant for                                                                                                                                              | US only                                                              |
| III-4-4 | Name (LAST, First)                                                                                                                                         | LANG, Friedrich                                                      |
| III-4-5 | Address                                                                                                                                                    | Berwartsteinstr. 18<br>76767 Hagenbach<br>Germany                    |
| III-4-6 | State of nationality                                                                                                                                       | DE                                                                   |
| III-4-7 | State of residence                                                                                                                                         | DE                                                                   |
| IV-1    | <b>Agent or common representative; or<br/>address for correspondence</b>                                                                                   |                                                                      |
|         | The person identified below is hereby/<br>has been appointed to act on behalf of<br>the applicant(s) before the competent<br>International Authorities as: | agent                                                                |
| IV-1-1  | Name (LAST, First)                                                                                                                                         | ADAM, Holger                                                         |
| IV-1-2  | Address                                                                                                                                                    | KRAUS & WEISERT<br>Thomas-Wimmer-Ring 15<br>80539 München<br>Germany |
| IV-1-3  | Telephone No.                                                                                                                                              | ++49-89-29060-0                                                      |
| IV-1-4  | Facsimile No.                                                                                                                                              | ++49-89-29060-111                                                    |
| IV-1-5  | e-mail                                                                                                                                                     | office@kraus-weisert.de                                              |
| IV-2    | <b>Additional agent(s)</b>                                                                                                                                 | additional agent(s) with same address as                             |

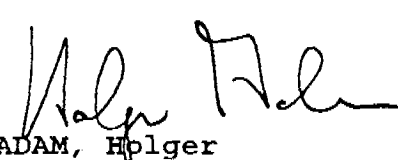
## PCT REQUEST

Original (for SUBMISSION)

|        |                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                      |                                                                                                                                    |
|--------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------------------------------------------|
| IV-2-1 | Name(s)                                                                                                                                                                                                                                                                                                                                                                                                                                                                                              | first named agent<br>ALBRECHT, Thomas; BANZER, Hans-Jörg;<br>BECKMANN, Claus; HIEBL, Inge;<br>BERTSCH, Florian; NIELSEN, Ferdinand |
| V      | DESIGNATIONS                                                                                                                                                                                                                                                                                                                                                                                                                                                                                         |                                                                                                                                    |
| V-1    | The filing of this request constitutes under Rule 4.9(a), the designation of all Contracting States bound by the PCT on the international filing date, for the grant of every kind of protection available and, where applicable, for the grant of both regional and national patents.                                                                                                                                                                                                               |                                                                                                                                    |
| V-2    | Item V-2 may only be used to exclude (irrevocably) the designations concerned if, at the time of filing, the international application contains a priority claim to an earlier national application filed in the particular State concerned, in order to avoid the ceasing of the effect, under the national law, of this earlier national application. As to the consequences of such national law provisions in these States, see the PCT Applicant's Guide, Volume I/A, in the relevant Annex B1. | DE                                                                                                                                 |
| VI-1   | Priority claim of earlier national application                                                                                                                                                                                                                                                                                                                                                                                                                                                       |                                                                                                                                    |
| VI-1-1 | Filing date                                                                                                                                                                                                                                                                                                                                                                                                                                                                                          | 03 May 2005 (03.05.2005)                                                                                                           |
| VI-1-2 | Number                                                                                                                                                                                                                                                                                                                                                                                                                                                                                               | 102005020685.9                                                                                                                     |
| VI-1-3 | Country                                                                                                                                                                                                                                                                                                                                                                                                                                                                                              | DE                                                                                                                                 |
| VI-2   | Priority claim of earlier national application                                                                                                                                                                                                                                                                                                                                                                                                                                                       |                                                                                                                                    |
| VI-2-1 | Filing date                                                                                                                                                                                                                                                                                                                                                                                                                                                                                          | 23 December 2005 (23.12.2005)                                                                                                      |
| VI-2-2 | Number                                                                                                                                                                                                                                                                                                                                                                                                                                                                                               | 102005061948.7                                                                                                                     |
| VI-2-3 | Country                                                                                                                                                                                                                                                                                                                                                                                                                                                                                              | DE                                                                                                                                 |
| VII-1  | International Searching Authority Chosen                                                                                                                                                                                                                                                                                                                                                                                                                                                             | European Patent Office (EPO) (ISA/EP)                                                                                              |

## PCT REQUEST

Original (for SUBMISSION )

|        |                                                                                                                                      |                                                                                      |                             |
|--------|--------------------------------------------------------------------------------------------------------------------------------------|--------------------------------------------------------------------------------------|-----------------------------|
| VIII   | <b>Declarations</b>                                                                                                                  | Number of declarations                                                               |                             |
| VIII-1 | Declaration as to the identity of the inventor                                                                                       | -                                                                                    |                             |
| VIII-2 | Declaration as to the applicant's entitlement, as at the international filing date, to apply for and be granted a patent             | -                                                                                    |                             |
| VIII-3 | Declaration as to the applicant's entitlement, as at the international filing date, to claim the priority of the earlier application | -                                                                                    |                             |
| VIII-4 | Declaration of inventorship (only for the purposes of the designation of the United States of America)                               | -                                                                                    |                             |
| VIII-5 | Declaration as to non-prejudicial disclosures or exceptions to lack of novelty                                                       | -                                                                                    |                             |
| IX     | <b>Check list</b>                                                                                                                    | number of sheets                                                                     | electronic file(s) attached |
| IX-1   | Request (including declaration sheets)                                                                                               | 5                                                                                    | ✓                           |
| IX-2   | Description                                                                                                                          | 14                                                                                   | -                           |
| IX-3   | Claims                                                                                                                               | 3                                                                                    | -                           |
| IX-4   | Abstract                                                                                                                             | 1                                                                                    | ✓                           |
| IX-5   | Drawings                                                                                                                             | 0                                                                                    | -                           |
| IX-7   | TOTAL                                                                                                                                | 23                                                                                   |                             |
|        | <b>Accompanying Items</b>                                                                                                            | paper document(s) attached                                                           | electronic file(s) attached |
| IX-8   | Fee calculation sheet                                                                                                                | ✓                                                                                    | -                           |
| IX-13  | Priority document(s)                                                                                                                 | Item(s) VI-1 VI-2                                                                    | -                           |
| IX-17  | PCT-SAFE physical media                                                                                                              | -                                                                                    | ✓                           |
| IX-19  | Figure of the drawings which should accompany the abstract                                                                           |                                                                                      |                             |
| IX-20  | Language of filing of the international application                                                                                  | English                                                                              |                             |
| X-1    | Signature of applicant, agent or common representative                                                                               |  |                             |
| X-1-1  | Name (LAST, First)                                                                                                                   | ADAM, Hölger                                                                         |                             |
| X-1-2  | Name of signatory                                                                                                                    |                                                                                      |                             |
| X-1-3  | Capacity                                                                                                                             |                                                                                      |                             |

## PCT REQUEST

Original (for SUBMISSION )

## FOR RECEIVING OFFICE USE ONLY

|        |                                                                                                                                         |                               |
|--------|-----------------------------------------------------------------------------------------------------------------------------------------|-------------------------------|
| 10-1   | Date of actual receipt of the purported international application                                                                       | ( 2 8. 04. 06 ) 28 APR 2006 ✓ |
| 10-2   | Drawings:                                                                                                                               |                               |
| 10-2-1 | Received                                                                                                                                |                               |
| 10-2-2 | Not received                                                                                                                            |                               |
| 10-3   | Corrected date of actual receipt due to later but timely received papers or drawings completing the purported international application |                               |
| 10-4   | Date of timely receipt of the required corrections under PCT Article 11(2)                                                              |                               |
| 10-5   | International Searching Authority                                                                                                       | ISA/EP                        |
| 10-6   | Transmittal of search copy delayed until search fee is paid                                                                             |                               |

## FOR INTERNATIONAL BUREAU USE ONLY

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|------|----------------------------------------------------------------|--|
| 11-1 | Date of receipt of the record copy by the International Bureau |  |
|------|----------------------------------------------------------------|--|



No. 07-105205- -00000-0000

Fecha: 2007-10-08 15:58:47 Dep. 2020 NUECREACIONI  
Fra. 11 PATENTEIYII Eve: 379 FASENACIONALI  
Act. 411 PRESENTACION Folios: 63



**Industria y Comercio**  
SUPERINTENDENCIA

## REQUISITOS MINIMOS PARA ADMISION A TRAMITE PCT

PATENTE DE INVENCION

☒

MODELO DE UTILIDAD ☐

- ◆ Indicación de que se solicita la concesión de una patente ☒
- ◆ Datos de identificación del solicitante o de la persona que presenta la solicitud ☒
- ◆ Descripción de la invención ☒
- ◆ Dibujos de ser estos pertinentes ☐
- ◆ Comprobante de pago de las tasas establecidas ☒

COMPLETA ☒

INCOMPLETA ☐

DISEÑO INDUSTRIAL ☐

- ◆ Indicación de que se solicita el registro de Diseño Industrial ☐
- ◆ Datos de identificación del solicitante o de la persona que presenta la solicitud ☐
- ◆ Representación gráfica y fotográfica del Diseño Industrial o muestra del material que incorpora el diseño ☐
- ◆ Comprobante de pago de las tasas establecidas ☐

COMPLETA ☐

INCOMPLETA ☐

ESQUEMA DE TRAZADO ☐

- ◆ Indicación de que se solicita el registro de un Esquema de Trazado ☐
- ◆ Datos de identificación del solicitante o de la persona que presenta la solicitud ☐
- ◆ Representación gráfica del esquema trazado ☐
- ◆ Comprobante de pago de las tasas establecidas ☐

COMPLETA ☐

INCOMPLETA ☐

Firma Responsable

Fecha

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**REPUBLICA DE COLOMBIA**  
**SUPERINTENDENCIA DE INDUSTRIA Y COMERCIO**

Bogotá,  
2020

Doctor(a)  
RODRIGUEZ D'ALEMAN DILIA MARIA

|            |                            |                   |
|------------|----------------------------|-------------------|
| REFERENCIA | Radicación No.             | 7 105205          |
|            | Solicitud internacional    | PCT/EP2006/004029 |
|            | Fecha inicio fase nacional | 03/12/2007        |
|            | Trámite                    | 11                |
|            | Evento                     | 379               |
|            | Actuación                  | 430               |
|            | Oficio No.                 | 15860             |

Como resultado del examen de forma, realizado con fundamento en el artículo 38 de la Decisión 486 de la Comisión de la Comunidad Andina, artículo 27 del Tratado de Cooperación en Materia de Patentes (PCT), regla 51 bis del Reglamento y Circular Única de la Superintendencia de Industria y Comercio, se le comunica que:

1. La solicitud no contiene la copia del documento en que consta la cesión del derecho a la patente del inventor al solicitante o a su causante. (Art. 26-k) Decisión 486.

2. La solicitud no contiene el poder debidamente otorgado, con el lleno de los requisitos exigidos por los artículos 63 y subsiguientes del Código de Procedimiento Civil. Adicionalmente, deberá presentar el documento que acredita la existencia y representación legal de la sociedad solicitante, cuando ésta fuere persona jurídica.

En cumplimiento de lo dispuesto por el artículo 39 de la Decisión 486, se le requiere para que dentro del término de dos meses siguientes a la fecha de notificación del presente oficio, complete los requisitos anteriormente enunciados. En caso de no dar cumplimiento al presente requerimiento, la solicitud se considerará abandonada y perderá su prelación.

Notifíquese el presente oficio conforme a lo dispuesto en el numeral 5.2, literal e), del capítulo quinto del título primero de la Circular Unica No.10 de 2001.

ALIX CARMENZA CESPEDES DE VERGEL  
Jefe de la división de Nuevas Creaciones

El anterior requerimiento fue notificado por fijación en lista de fecha \_\_\_\_\_.  
adpall