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- (71) **Applicants:** **DAEWOONG PHARMACEUTICAL CO., LTD.** [KR/KR]; 244, Galmachi-ro, Jungwon-gu, Seongnam-si, Gyeonggi-do 13211 (KR). **UNIVERSITY-INDUSTRY COOPERATION GROUP OF KYUNG HEE UNIVERSITY** [KR/KR]; Global Campus, 1732, Deogyong-daero, Giheung-gu, Yongin-si, Gyeonggi-do 17104 (KR).
- (72) **Inventors:** **CHOI, Jung-Hye**; #104-1902(Oksu-dong, Kukdong Green Apt.), 175, Dokseodang-ro, Seongdong-gu, Seoul 04732 (KR). **CHO, Yoon Jin**; #113-501(Beon-dong, Jugong Apt.), 23, Hancheon-ro, 105-gil, Gangbuk-gu, Seoul 01230 (KR). **RYU, Hyo Jeong**; #103-1502(Songdo-dong, Poongrim I-won Apt.), 6, Sinsong-ro 118beon-gil, Yeonsu-gu, Incheon 21994 (KR). **KANG, Hee Chol**; #106-401(Mok-dong, Mokyang Maeul Apt.), 37, Mokdong-ro, Jung-gu, Daejeon 34818 (KR). **KIM, Jin Pyo**; #205-603(Dunsan-dong, Gukhwa Apt.), 201, Dunsan-ro, Seo-gu, Daejeon 35245 (KR).

(74) **Agents:** **YOU, Chi-youl** et al.; Good Idea International Patent Office, 203-210, Bucheon Technopark Ssangyong 3rd, 397(Samjeong-dong), Seokcheon-ro, Bucheon-si, Gyeonggi-do 14449 (KR).

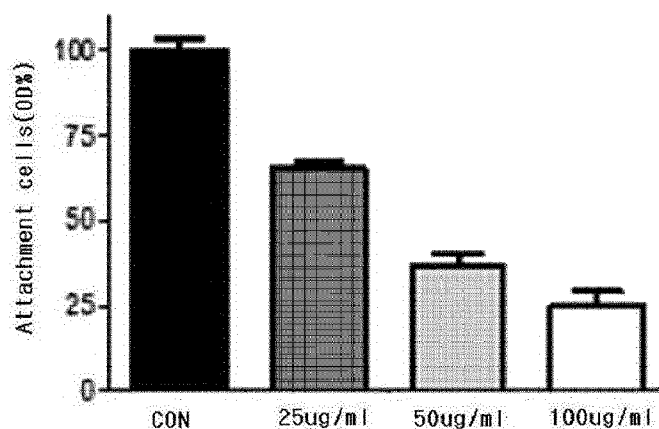
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(54) **Title:** NOVEL USE OF ALCOHOL EXTRACT OF TARAXACI HERBA



(57) **Abstract:** Disclosed is novel use of an alcohol extract of Taraxaci Herba, whereby endometriosis can be effectively treated, ameliorated, and/or prevented.

Description

Title of Invention: NOVEL USE OF ALCOHOL EXTRACT OF TARAXACI HERBA

Technical Field

- [1] The present invention relates to an alcohol extract of Taraxaci Herba and, more particularly, to a novel use of an alcohol extract of Taraxaci Herba.

Background Art

- [2] Endometriosis is a very common gynecological disease occurring in about 3 to 10% of fertile women and about 25 to 35% of infertile women, resulting from abnormally proliferating endometrial tissue in locations other than the uterus (ovaries, abdominal cavity, intestinal tract, bladder, etc.).
- [3] Endometriosis has a variety of pain symptoms, including dysmenorrhoea, dyspareunia and pain in various invasion portions, and is regarded as the cause of infertility in 30 to 50% of patients.
- [4] Endometriosis has been treated to date only via surgery for removing the endometrium, and other treatment methods have not yet been introduced. Such surgical therapy may limit future pregnancy options and suffers from a high recurrence rate.
- [5] Although not fatal, endometriosis incurs discomfort in daily life due to severe pain and results in infertility in severe cases, and has to be seriously recognized as affecting the quality of women's lives and causing pregnancy problems, and must be overcome. However, prevention and treatment methods thereof are still lacking.
- [6] There is thus an urgent need to develop a new concept of medicaments for the prevention and treatment of endometriosis through natural methods without any side effects in women, especially fertile women, in lieu of invasive surgical therapy.
- [7] Meanwhile, Taraxaci Herba is the whole plant of a dandelion, and a hydrothermal extract of Taraxaci Herba is reported to be effective in the treatment of endometriosis (The Journal of Oriental Gynecology, Vol. 19, No. 3: 069-082 (2006)).
- [8] [Citation List]
- [9] [Non-Patent Literature]
- [10] (Non-Patent Document 1) The Journal of Oriental Gynecology, Vol. 19, No. 3: 069-082 (2006)

Disclosure of Invention

Technical Problem

- [11] Accordingly, the present invention has been made keeping in mind the above problems encountered in the related art, and the present invention is intended to provide novel use of an extract, which is effective in the treatment, amelioration, or

prevention of endometriosis.

- [12] The technical problem of the present invention is not limited to the foregoing, and additional technical problems, which are not mentioned in the foregoing, will be readily understood by those skilled in the art from the following description.

Solution to Problem

- [13] The present inventors have ascertained the surprisingly high effects of an alcohol extract of Taraxaci Herba on the treatment, amelioration and/or prevention of endometriosis, thus culminating in the present invention.
- [14] The present invention provides the use of an alcohol extract of Taraxaci Herba in a medicament and/or food for the treatment, amelioration and/or prevention of endometriosis.
- [15] In addition, the present invention provides a pharmaceutical composition for the treatment or prevention of endometriosis, containing the alcohol extract of Taraxaci Herba as an active ingredient.
- [16] The alcohol extract may result from extracting Taraxaci Herba with an alcohol or an aqueous alcohol solution.
- [17] The alcohol may be an alcohol having a purity of 99 vol% or more.
- [18] The aqueous alcohol solution may be an aqueous alcohol solution having an alcohol content of 30 to 99.98 vol%.
- [19] The aqueous alcohol solution may be an aqueous alcohol solution having an alcohol content of 30 to 70 vol%, or an aqueous alcohol solution having an alcohol content of 70 to 99.98 vol%. Given the above range, the resulting extract may be more effectively used for endometriosis.
- [20] The alcohol may be a lower alcohol, and is preferably at least one lower alcohol selected from among C1 to C5 alcohols. Given the above range, the resulting extract may be more effectively used for endometriosis.
- [21] The alcohol may be ethanol.
- [22] The treatment or prevention may be achieved by at least one selected from among inhibition of endometriotic cell migration, inhibition of endometriotic cell attachment or implantation, and inhibition of endometriotic tissue formation.
- [23] Also, the treatment or prevention may be achieved by inhibiting the expression of an enzyme associated with migration or attachment of endometriotic cells.
- [24] The enzyme associated with migration or attachment of endometriotic cells may be at least one selected from among MMP-2 and MMP-9.
- [25] Taraxaci Herba may be the whole plant of a dandelion, and may be those processed through drying or the like. The dandelion encompasses *Taraxacum platycarpum* H. Dahlstedt or other plants of the same genus (Compositae). For example, Taraxaci

Herba may be those disclosed in the Korean National Standard of Traditional Medicinal (Herbal and Botanical) Materials. In the Korean National Standard of Traditional Medicinal (Herbal and Botanical) Materials, Taraxaci Herba is described as the whole plant of *Taraxacum platycarpum* H. Dahlstedt, *Taraxacum officinale* Weber, *Taraxacum mongolicum* Handel-Mazzetti, or *Taraxacum coreanum* Nakai (Compositae). This Taraxaci Herba may be those that are commercially available or are directly collected.

- [26] In addition, the present invention provides a food composition for the amelioration or prevention of endometriosis, containing the alcohol extract of Taraxaci Herba as an active ingredient.
- [27] Unless otherwise mentioned, the matters described in connection with the pharmaceutical composition according to the present invention are applicable to food compositions so long as they are not incompatible with each other. Herein, "amelioration" is encompassed in the term "treatment", and means that conditions or symptoms are lessened.
- [28] The food composition may be variously contained in food including drinks, and may be provided in the form of a drink, gum, tea, functional health food and the like. The functional health food may be formulated in the dosage form of a tablet, capsule, etc. According to the Korean Health Functional Foods Act No. 12669, the term "functional health food" refers to food manufactured (including processing; hereinafter the same shall apply) using functional raw materials or ingredients beneficial to the human body, and the term "functionality" means controlling nutrients for the structure and functions of the human body or providing beneficial effects to health purposes, such as physiological effects. The food composition may include a typical food additive, and examples of the food additive may include chemical compounds, such as ketones, glycine, sodium citrate, nicotinic acid, and cinnamic acid, natural additives, such as persimmon color, licorice extract, crystalline cellulose, kaoliang colorant, and guar gum, and mixed formulations, such as a sodium L-glutamate formulation, an alkali agent for noodles, a preservative formulation, and a tar color formulation. The term "food composition" is to be understood as having a meaning that includes a food additive and the like.
- [29] In addition, the present invention provides a method of treating or preventing endometriosis, including administering the alcohol extract of Taraxaci Herba to mammals, including humans, in need of administration. In addition, the present invention provides the use of the alcohol extract of Taraxaci Herba in the manufacture of a medicament for the treatment or prevention of endometriosis. The administered alcohol extract of Taraxaci Herba may be an alcohol extract of Taraxaci Herba in an effective amount.

- [30] Unless otherwise mentioned, the matters described in connection with the pharmaceutical composition, the food composition, the method, and the use according to the present invention are applicable to each other so long as they are not incompatible with each other.
- [31] The alcohol extract of Taraxaci Herba or the composition thereof may be orally or parenterally administered to mammals including humans, and the active ingredient and a pharmaceutically acceptable vehicle may be mixed, formulated and administered. When formulations are produced, a filler, an extender, a binder, a humectant, a disintegrant, a surfactant, a diluent, or an excipient, which is typically useful, may be used. A solid formulation for oral administration may be provided in the form of a tablet, a pill, a powder, a granule, or a capsule, and such a solid formulation is produced by adding the composition of the present invention with at least one excipient, for example, starch, calcium carbonate, sucrose, lactose, and/or gelatin. Furthermore, a lubricant such as magnesium or talc may be used. A liquid formulation for oral administration may be provided in the form of a suspension, an oral solution, an emulsion or a syrup, and may include not only a simple diluent, such as water or liquid paraffin, but also various excipients, for example, a humectant, a sweetener, a fragrance and/or a preservative. A formulation for parenteral administration includes an injectable solution, a suspension, an emulsion, a lyophilisate, an intranasal cleaner and a suppository. The injectable solution, suspension, or emulsion may be prepared by mixing water, a non-aqueous solvent or a suspending solvent with the active ingredient, and the non-aqueous solvent and the suspending solvent may include propylene glycol, polyethylene glycol, a vegetable oil such as olive oil, and an injectable ester such as ethyl oleate. The substrate for a suppository may include Witepsol, Macrogol, Tween 61, cacao oil, laurin oil, glycerol and/or gelatin. Upon parenteral administration, subcutaneous injection, intravenous injection or intramuscular injection is possible.
- [32] The alcohol extract of Taraxaci Herba, which is contained in the composition of the present invention or is applied to the method or use of the present invention, may be used daily in an amount of 0.0001 to 1000 mg/kg, preferably 0.0001 to 100 mg/kg, and more preferably 0.001 to 10 mg/kg for a female adult. It may be administered once a day, or may be divided in multiple applications to be administered several times per day. However, the scope of the present invention is not limited to the administration amount and frequency set forth above.
- [33] The composition of the present invention may contain the active ingredient in an amount of 0.1 to 99.9 wt% based on the total weight of the composition.
- [34] The alcohol extract of Taraxaci Herba may be formulated together with an additive, such as a pharmaceutically or sitologically acceptable vehicle, excipient or diluent.

For the description of formulations, reference may be made to Remington's Pharmaceutical Science (latest edition), Mack Publishing Company, Easton PA.

- [35] Accordingly, the composition of the present invention further includes a pharmaceutically or sitologically acceptable additive, and may thus be composed of the active ingredient and the pharmaceutically or sitologically acceptable additive.

Advantageous Effects of Invention

- [36] According to the present invention, endometriosis can be effectively treated, ameliorated and/or prevented.

Brief Description of Drawings

- [37] FIG. 1 is of graphs showing the results of testing of the effects of the alcohol extract of Taraxaci Herba on inhibiting the migration or attachment of endometriotic cells;
- [38] FIG. 2 is of graphs showing the results of testing of the effects of the alcohol extract of Taraxaci Herba on inhibiting the migration or attachment of endometriotic cells;
- [39] FIG. 3 is of graphs showing the results of testing of the effects of the alcohol extract of Taraxaci Herba on inhibiting the migration or attachment of endometriotic cells;
- [40] FIG. 4 is a graph showing the results of testing of the effects of the alcohol extract of Taraxaci Herba on inhibiting the migration of endometriotic cells;
- [41] FIG. 5 is a graph showing the results of testing of the effects of the hydrothermal extract of Taraxaci Herba on inhibiting the migration of endometriotic cells;
- [42] FIG. 6 is a graph showing the results of testing of the effects of the alcohol extract of Taraxaci Herba on inhibiting the attachment and implantation of endometriotic cells;
- [43] FIG. 7 is a graph showing the results of testing of the effects of the hydrothermal extract of Taraxaci Herba on inhibiting the attachment and implantation of endometriotic cells; and
- [44] FIG. 8 is a graph showing the results of testing of the effects of the alcohol extract of Taraxaci Herba in an endometriosis animal model.

Mode for the Invention

- [45] A better understanding of the present invention may be obtained through the following examples and preparation examples, which are set forth to illustrate, but are not to be construed as limiting the present invention.
- [46] By evaluating the influences of the alcohol extract of Taraxaci Herba on the features of endometriosis, including changes in expression of an enzyme associated with migration or attachment of endometriotic cells, endometriotic cell migration, and endometriotic cell attachment and implantation, the effects of the alcohol extract of Taraxaci Herba on the treatment, amelioration or prevention of endometriosis were confirmed as follows.
- [47] In the following Examples and Comparative Examples, DMEM/F12 medium was

purchased from Life Technologies (Grand Island, New York, U.S.A.), and cell tracker CMFDA was purchased from Invitrogen (Paisley, United Kingdom). Unless otherwise mentioned, reagents were purchased from Sigma. Also, dry Taraxaci Herba, as disclosed in the Korean National Standard of Traditional Medicinal (Herbal and Botanical) Materials, was purchased from Kyung Dong Market (Seoul, Korea).

[48] The extracts used in the following Examples and Comparative Examples were prepared as follows.

[49] <Preparation of hydrothermal extract of Taraxaci Herba>

[50] Taraxaci Herba, dried in the shade, was milled to a particle size of about 400 μm . 200 g of the Taraxaci Herba thus prepared was added with 1 L of tertiary distilled water and refluxed at 90°C for 3 hr, thus obtaining a filtrate, after which extraction was repeated once more in the same manner. The extracted filtrate was concentrated under reduced pressure at 60°C, thus yielding 46 g of a hydrothermal extract of Taraxaci Herba in powder form.

[51]

[52] <Preparation of alcohol extract of Taraxaci Herba>

[53] Taraxaci Herba, dried in the shade, was milled to a particle size of about 400 μm . 200 g of the Taraxaci Herba thus prepared was added with 2 L of a 30% (v/v) ethanol aqueous solution and immersed at room temperature for 24 hr, thus obtaining a filtrate, after which extraction was repeated once more in the same manner. The extracted filtrate was concentrated under reduced pressure at 60°C, thus yielding 38 g of a 30% ethanol extract of Taraxaci Herba in powder form.

[54] Also, Taraxaci Herba, dried in the shade, was milled to a particle size of about 400 μm . 600 g of the Taraxaci Herba thus prepared was added with 6 L of a 70% (v/v) ethanol aqueous solution, and immersed at room temperature for 24 hr, thus obtaining a filtrate, after which extraction was repeated once more in the same manner. The extracted filtrate was concentrated under reduced pressure at 60°C, thus yielding 84 g of a 70% ethanol extract of Taraxaci Herba in powder form.

[55] Also, Taraxaci Herba, dried in the shade, was milled to a particle size of about 400 μm . 200 g of the Taraxaci Herba thus prepared was added with 2 L of a 99.98% ethanol aqueous solution and immersed at room temperature for 24 hr, thus obtaining a filtrate, after which extraction was repeated once more in the same manner. The extracted filtrate was concentrated under reduced pressure at 60°C, thus yielding 8 g of a 99.98% ethanol extract of Taraxaci Herba in soft extract form.

[56]

[57] <Example 1> Evaluation of effects of alcohol extract of Taraxaci Herba on inhibiting the migration or attachment of endometriotic cells

[58] In order to evaluate whether the alcohol extract of Taraxaci Herba was able to inhibit

mRNA expression of MMP-2 (matrix metalloproteinase-2) and MMP-9 (matrix metalloproteinase-9), real-time PCR was performed. This is because MMP-2 and MMP-9 are known to be proteases that play an important role in the migration, attachment and invasion of endometrial cells.

- [59] Specifically, endometrial cells, namely 12Z cells {epithelial endometrial cells; Johann-Wolfgang-Goethe-Universität (Germany), Dr. Starzinski-Powitz} were treated for 24 hr with the 30% ethanol extract of Taraxaci Herba, the 70% ethanol extract of Taraxaci Herba, and the 99.98% ethanol extract of Taraxaci Herba at respective concentrations of 50 $\mu\text{g/mL}$, 100 $\mu\text{g/mL}$, and 200 $\mu\text{g/mL}$, after which total RNA was extracted from the 12Z cells using TRIzol (Invitrogen Canada, Burlington, ON, Canada) by a method known in the art, and reverse transcription polymerase chain reaction (RT-PCR) was carried out to synthesize first-strand DNA. SYBR Green real-time PCR was conducted using the first-strand DNA as a template by means of a Thermal Cycler Dice Real-Time PCR System (Takara, Japan) in a manner such that reactions were cycled 45 times under conditions of denaturation at 95°C for 5 sec, annealing at 57°C for 10 sec, and extension at 72°C for 20 sec. Used for the SYBR Green real-time PCR, MMP-2 primers, MMP-9 primers, and as a control group, GAPDH primers are shown in Table 1 below. The average Ct values of MMP-2 and MMP-9 were determined through triplicate measurement, and corrected by the Ct value of the GAPDH control group.

- [60] [Table 1]

MMP-2	Sense primer	5'-CCGCAGTGACGGAAAGATGT-3'	SEQ ID NO:1
	Antisense primer	5'-CACTTGCGGTCGTCATCGTA-3'	SEQ ID NO:2
MMP-9	Sense primer	5'-GGACGATGCCTGCAACGT-3'	SEQ ID NO:3
	Antisense primer	5'-CAAATACAGCTGGTTCCTCAATCT-3'	SEQ ID NO:4
GAPDH	Sense primer	5'-GGGGCTCTCCAGAACATCAT-3'	SEQ ID NO:5
	Antisense primer	5'-CAGGTCAGGTCCACCACTGA-3'	SEQ ID NO:6

- [61]

- [62] The results are summarized in FIGS. 1 to 3. FIGS. 1 to 3 are graphs showing the results of testing of the effects of the alcohol extracts of Taraxaci Herba on inhibiting the migration or attachment of endometrial cells. FIG. 1 shows the results of the 30% ethanol extract, FIG. 2 shows the results of the 70% ethanol extract, and FIG. 3 shows the results of the 99.98% ethanol extract. In respective drawings, the upper graphs illustrate the results of MMP-2, and the lower graphs illustrate the results of MMP-9. In

the respective graphs, x-axis designates the treatment concentration, and y-axis designates the relative mRNA expression. Also, "CON" indicates a control group that was not treated with the alcohol extract.

[63] As is apparent from the drawings, the mRNA expression of endogenous MMP-2 and MMP-9, which are typical expression proteins of endometriosis and play an important role in cell migration and invasion, was inhibited by the alcohol extract of Taraxaci Herba.

[64] Thus, the alcohol extract of Taraxaci Herba was found to be effective at inhibiting the migration or attachment of endometriotic cells.

[65] From this, the alcohol extract of Taraxaci Herba is regarded as effective in the treatment, prevention and/or amelioration of endometriosis.

[66]

[67] <Example 2> Evaluation of effects of alcohol extract of Taraxaci Herba on inhibiting the migration of endometriotic cells

[68] In order to evaluate the effects of the alcohol extract of Taraxaci Herba on the migration of endometriotic cells, a Transwell migration assay was repeated three times.

[69] Specifically, endometriotic cells, namely 12Z cells, were aliquoted into the upper part of a 24-well Transwell plate (8 mm pore size) and treated with 50 $\mu\text{g/mL}$ of the 70% ethanol extract of Taraxaci Herba. 16 hr after treatment with the extract, cells that had migrated to the outer surface of the Transwell plate were immobilized with methanol and stained with 0.5% crystal violet for 10 min. The cells that had migrated to the outer surface of the Transwell plate were observed with a microscope at 200x magnification, and the number thereof was counted to calculate the percentage relative to the total number of cells.

[70] The results are shown in FIG. 4. FIG. 4 is a graph showing the results of inhibition of endometriotic cell migration using the alcohol extract of Taraxaci Herba, the x-axis indicating the concentration and the y-axis indicating the migration (%) of endometriotic cells. Here, "CON" indicates a control group that was not treated with the alcohol extract.

[71] As is apparent from the drawing, the alcohol extract of Taraxaci Herba inhibited the migration of endometriotic cells.

[72] Thus, the alcohol extract of Taraxaci Herba was found to be effective in the treatment, prevention and/or amelioration of endometriosis by inhibiting the migration of endometriotic cells. In particular, the alcohol extract of Taraxaci Herba exhibited an effect about twice as high as that of the hydrothermal extract of Taraxaci Herba of Comparative Example 1.

[73]

[74] <Comparative Example 1> Evaluation of effects of hydrothermal extract of Taraxaci

Herba on inhibiting the migration of endometriotic cells

[75] This Comparative Example was performed in the same manner as in Example 2, with the exception that the hydrothermal extract of Taraxaci Herba was used, in lieu of the alcohol extract of Taraxaci Herba.

[76] The results are shown in FIG. 5. FIG. 5 is a graph showing the results of inhibition of endometriotic cell migration using the hydrothermal extract of Taraxaci Herba, the x-axis indicating the concentration and the y-axis indicating the migration (%) of endometriotic cells. Here, "CON" indicates a control group that was not treated with the hydrothermal extract.

[77] As is apparent from the drawing, the hydrothermal extract of Taraxaci Herba inhibited the migration of endometriotic cells, but the effects thereof were only about 40 to 50% of the effects of the alcohol extract of Taraxaci Herba.

[78]

[79] <Example 3> Evaluation of effects of alcohol extract of Taraxaci Herba on inhibiting the attachment and implantation of endometriotic cells

[80] In order to evaluate the effects of the alcohol extract of Taraxaci Herba on the attachment and implantation of endometriotic cells, an attachment assay was repeated three times.

[81] Specifically, in order to evaluate whether the feature of endometriosis, that is, a phenomenon in which endometriotic cells were attached and implanted to the organs around the peritoneum and the abdominal cavity could be inhibited using the alcohol extract of Taraxaci Herba, Human peritoneal mesothelial cells, namely Met-5A cells (American Type Culture Collection, ATCC), were cultured in a 5% CO₂ incubator with a 199 medium containing 10% FBS, 100 U/mL of penicillin G and 100 g/mL of streptomycin (Life Technologies, Grand Island, NY, U.S.A.), and 400 nM hydrocortisone (Sigma Chemical Co., St. Louis, MO, U.S.A.). Then, the Met-5A cells were aliquoted into a 96-well plate. 12Z cells (2x10³ cells/well), which are an endometriotic cell line labeled with the cell tracker green CMFDA (5-Chloromethylfluorescein diacetate), were mixed with a DMEM/F12 medium (Dulbecco's Modified Eagle's Medium/Ham's F-12 medium) containing the 99.98% ethanol extract of Taraxaci Herba at 25 µg/mL, 50 µg/mL, and 100 µg/mL, and then added to the pre-aliquoted Met-5A cells. The mixed cells were cultured in a 5% CO₂ incubator at 37°C for 1 hr, and the plate was inverted and washed with a phosphate-buffered solution containing Ca and Mg. The attached cells were analyzed at a wavelength of 490 nm, and the percentage of attached cells relative to 100%, as a control group, was calculated.

[82] The results are shown in FIG. 6. FIG. 6 is a graph showing the results of inhibition of endometriotic cell attachment and implantation using the alcohol extract of Taraxaci Herba, the x-axis indicating the concentration and the y-axis indicating the relative

ratio (OD (Optical Density), %) of attached cells. Here, "CON" indicates a control group that was not treated with the extract.

[83] As is apparent from the drawing, the attachment of the endometriotic cells 12Z treated with the alcohol extract of Taraxaci Herba to the Met-5A cells surrounding the outside of the organ around the peritoneum and the abdominal cavity was inhibited. Therefore, the alcohol extract of Taraxaci Herba was found to be effective in the treatment, prevention and/or amelioration of endometriosis by inhibiting the attachment and implantation of endometriotic cells. In particular, the hydrothermal extract of Taraxaci Herba of Comparative Example 2 had no effects, but the alcohol extract of Taraxaci Herba exhibited concentration-dependent effects, thus manifesting a high inhibitory efficacy of 60% or more at 50 μ g/mL.

[84]

[85] <Comparative Example 2> Evaluation of effects of hydrothermal extract of Taraxaci Herba on inhibiting the attachment and implantation of endometriotic cells

[86] This Comparative Example was performed in the same manner as in Example 3, with the exception that the hydrothermal extract of Taraxaci Herba was used, in lieu of the alcohol extract of Taraxaci Herba. The relative value of attached cells was represented as the ratio thereof relative to the control group, rather than being calculated as a percentage.

[87] The results are shown in FIG. 7. FIG. 7 is a graph showing the results of inhibition of endometriotic cell attachment and implantation using the hydrothermal extract of Taraxaci Herba, the x-axis indicating the concentration and the y-axis indicating the relative value of attached cells. Here, "CON" indicates a control group that was not treated with the extract.

[88] As is apparent from the drawing, the hydrothermal extract of Taraxaci Herba had no effects on inhibiting the attachment and implantation of endometriotic cells.

[89]

[90] <Example 4> Evaluation of effects of alcohol extract of Taraxaci Herba in endometriosis animal model

[91] The effects of the alcohol extract of Taraxaci Herba in endometriosis-induced models through intraperitoneal injection were evaluated.

[92] Endometriosis was induced in a manner in which the uterus of a donor mouse was taken out, chopped into pieces, and spread in the abdominal cavity of a recipient mouse. The drug administration is shown in Table 2 below. As the test drug, 250 mg/kg of the 70% ethanol extract of Taraxaci Herba was orally administrated for 5 weeks. The endometriosis-induced animal model was sacrificed, the abdomen was opened, and the number of implantations in the peritoneum and the abdominal cavity (small intestine, large intestine, stomach, liver, bladder, ovary, etc.) was counted. When the

alcohol extract was administered, the vehicle was 10% Tween 80, and the group in which only the vehicle was administered instead of the alcohol extract was designated as a control group, and the group in which 0.3 mg/kg of dienogest was administered instead of the alcohol extract was designated as a positive control group.

[93] [Table 2]

	1-week	2-week	3-week	4-week	5-week	6-week	7-week
Stabilization	√						
Drug administration		√	√	√	√	√	
Endometriosis induction IP			√	√	√	√	
Autopsy							√

[94]

[95] The results are shown in FIG. 8. FIG. 8 is a graph showing the results of testing of the effects of the alcohol extract of Taraxaci Herba in the endometriosis animal model, the x-axis indicating the treated group and the y-axis indicating nodule counts. The vehicle designates a control group that was not treated with a drug, the asterisk (*) indicating $p < 0.0001$ vs the vehicle.

[96] As is apparent from the drawing, the alcohol extract of Taraxaci Herba exhibited significant endometriotic tissue formation inhibitory effects.

[97] Therefore, the alcohol extract of Taraxaci Herba was found to be effective in the treatment, prevention and/or amelioration of endometriosis thanks to the endometriotic tissue formation inhibitory effects.

[98] Based on the above results, the alcohol extract of Taraxaci Herba according to the present invention is effective in the treatment, prevention and/or amelioration of endometriosis. In particular, the alcohol extract of Taraxaci Herba is very effective in the treatment, prevention and/or amelioration of endometriosis, compared to the hydrothermal extract of Taraxaci Herba.

[99]

[100] <Preparation Example 1> Production of pharmaceutical composition

[101] 300 mg of an alcohol extract of Taraxaci Herba, 100 mg of corn starch, 100 mg of lactose, and 2 mg of magnesium stearate were charged in a gelatin capsule, thus manufacturing a capsule. As such, the alcohol extract of Taraxaci Herba that was used was the 30%, 70% or 99.98% ethanol extract.

[102]

[103] <Preparation Example 2> Production of food composition

- [104] An alcohol extract of Taraxaci Herba (4 wt%), liquid fructose (0.5 wt%), oligosaccharide (2 wt%), sugar (2 wt%), and saline (0.5 wt%) were added with water in a remaining amount, homogeneously mixed, and flash-pasteurized, thus manufacturing a health drink. As such, the alcohol extract of Taraxaci Herba that was used was the 30%, 70% or 99.98% ethanol extract.

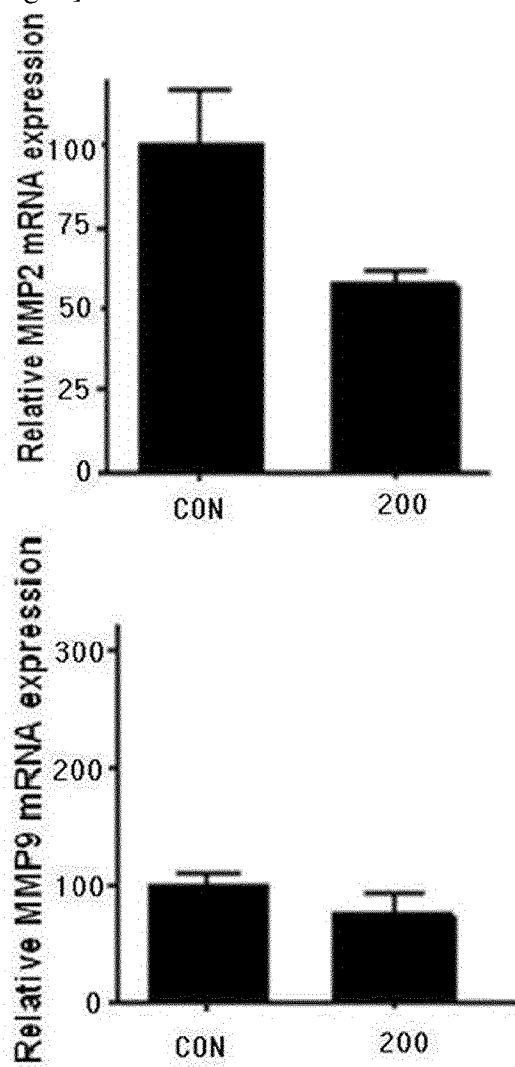
Industrial Applicability

- [105] The present invention can effectively treat, ameliorate and/or prevent endometriosis, and is thus industrially applicable.

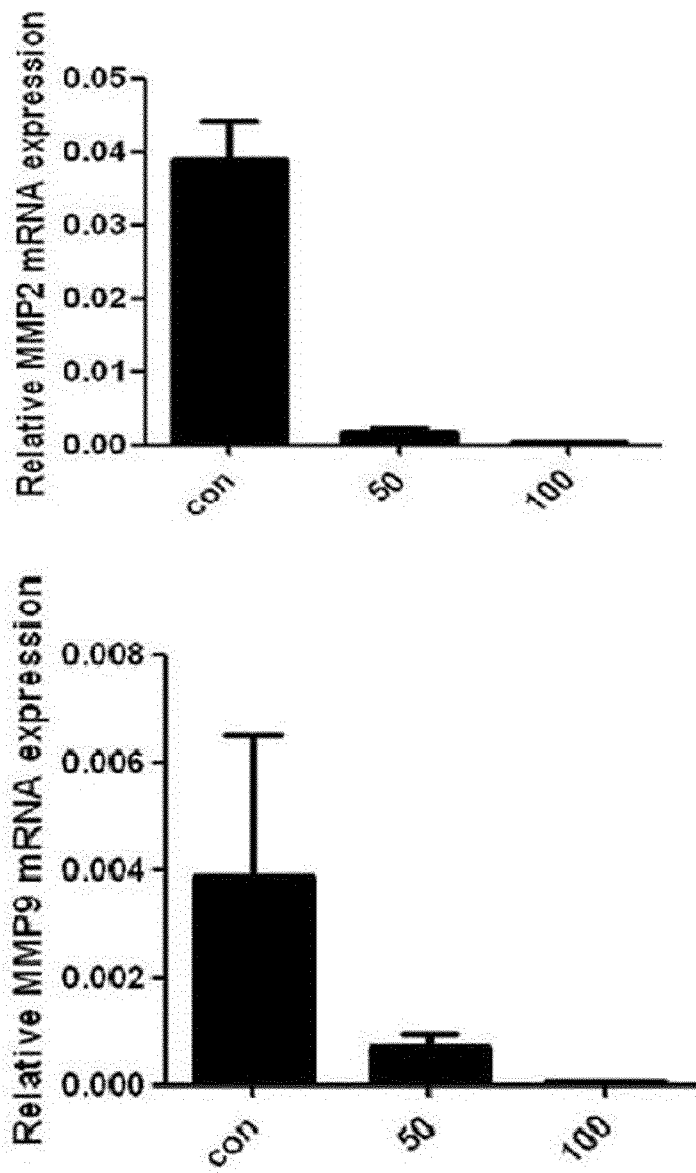
Claims

- [Claim 1] A pharmaceutical composition for treatment or prevention of endometriosis, containing an alcohol extract of Taraxaci Herba as an active ingredient.
- [Claim 2] The pharmaceutical composition of claim 1, wherein the alcohol extract is obtained by extracting Taraxaci Herba with an alcohol or an aqueous alcohol solution.
- [Claim 3] The pharmaceutical composition of claim 1, wherein the alcohol is at least one lower alcohol selected from among C1 to C5 alcohols.
- [Claim 4] The pharmaceutical composition of claim 1, wherein the treatment or prevention is achieved by at least one selected from among inhibition of endometriotic cell migration, inhibition of endometriotic cell attachment or implantation, and inhibition of endometriotic tissue formation.
- [Claim 5] The pharmaceutical composition of claim 1, wherein the treatment or prevention is achieved by inhibiting expression of an enzyme associated with migration or attachment of endometriotic cells.
- [Claim 6] The pharmaceutical composition of claim 5, wherein the enzyme associated with migration or attachment of endometriotic cells is at least one selected from among MMP-2 and MMP-9.
- [Claim 7] A food composition for amelioration or prevention of endometriosis, containing an alcohol extract of Taraxaci Herba as an active ingredient.
- [Claim 8] Use of an alcohol extract of Taraxaci Herba in manufacture of a medicament for treatment or prevention of endometriosis.
- [Claim 9] The use of claim 8, wherein the treatment or prevention is achieved by at least one selected from among inhibition of endometriotic cell migration, inhibition of endometriotic cell attachment or implantation, and inhibition of endometriotic tissue formation.
- [Claim 10] The use of claim 8, wherein the treatment or prevention is achieved by inhibiting expression of an enzyme associated with migration or attachment of endometriotic cells.

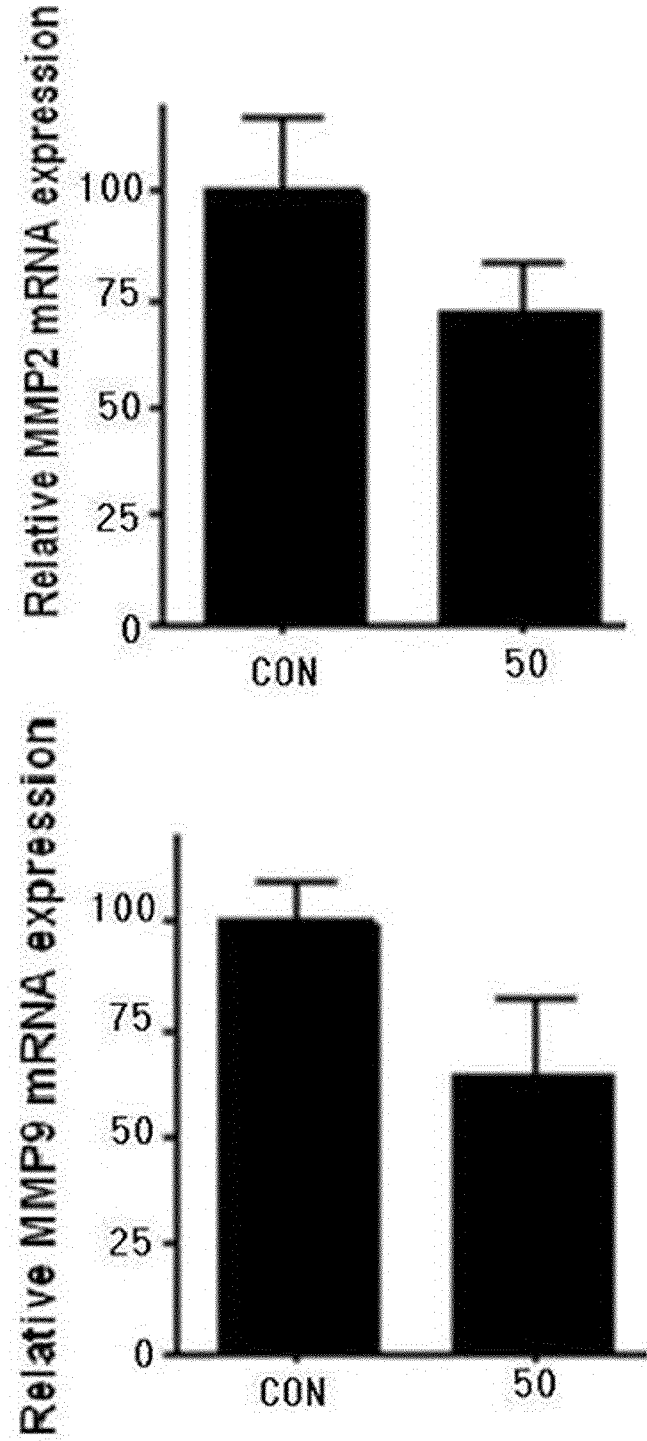
[Fig. 1]



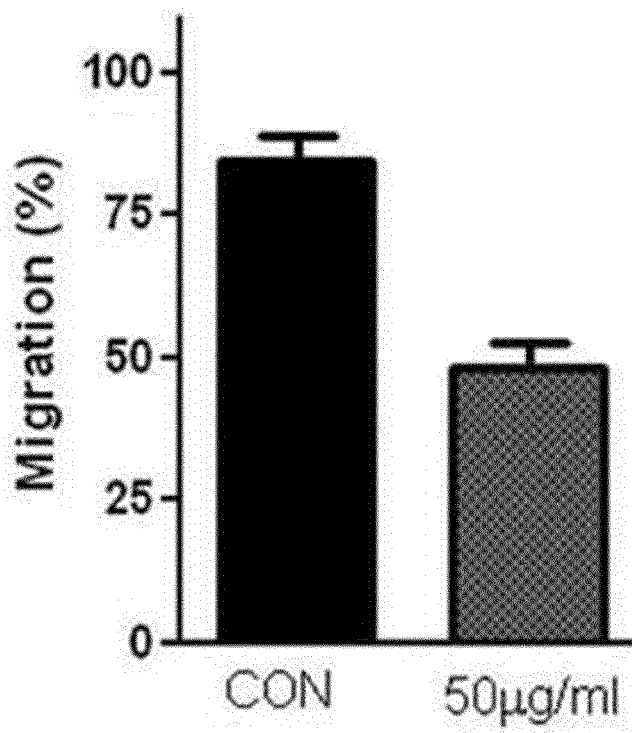
[Fig. 2]



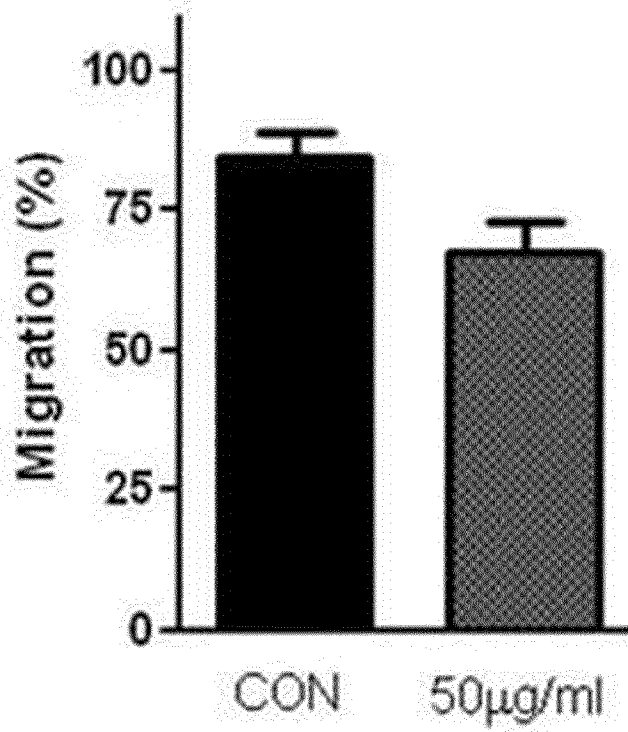
[Fig. 3]



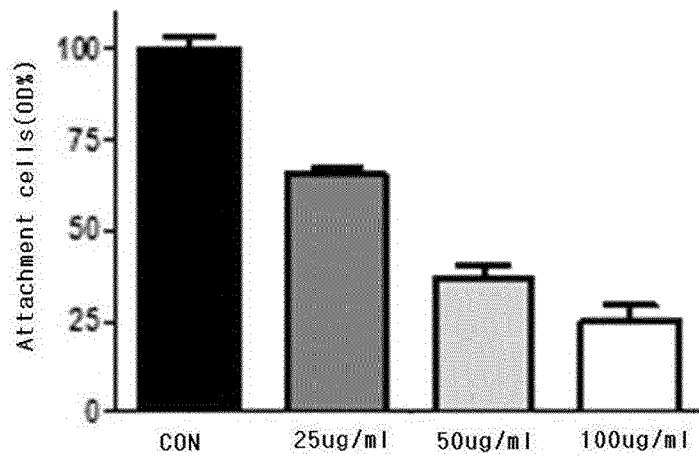
[Fig. 4]



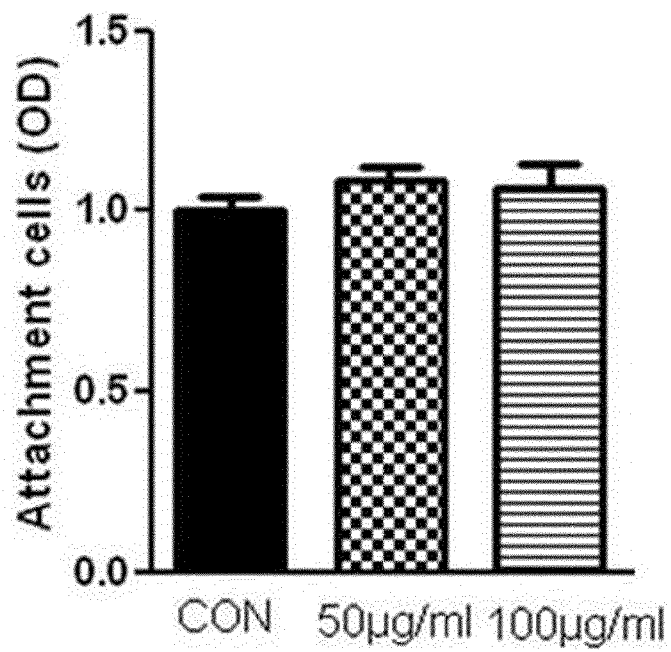
[Fig. 5]



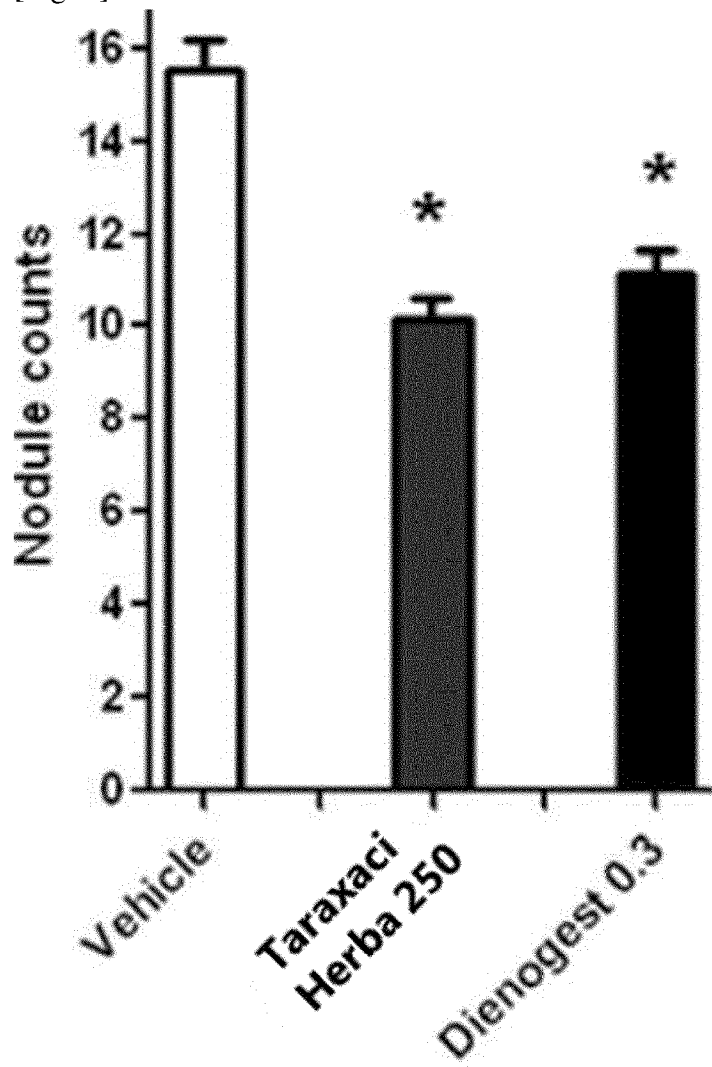
[Fig. 6]



[Fig. 7]



[Fig. 8]



INTERNATIONAL SEARCH REPORT

International application No.
PCT/KR2016/010502**A. CLASSIFICATION OF SUBJECT MATTER****A61K 36/288(2006.01)i, A23L 1/30(2006.01)i**

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 36/288; A61P 35/00; A61K 36/9066; A23L 1/30

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

Korean utility models and applications for utility models

Japanese utility models and applications for utility models

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

eKOMPASS(KIPO internal) & Keywords: Taraxaci herba, dandelion, endometriosis, retrograde menstruation, MMP-2, MMP-9, matrix metalloproteinase**C. DOCUMENTS CONSIDERED TO BE RELEVANT**

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	Park, J. -S. et al., 'Effect of Taraxaci Herba on Surgically Induced Endometriosis in Rats' , The Journal of oriental obstetrics & Gynecology, 2006, Vol. 19, No. 3, pages 069-082 See abstract; and pages 70, 79.	1-4, 7-9
A		5-6, 10
A	CN 104013928 A (XINJIANG MEDICAL UNIVERSITY AFFILIATED HOSPITAL OF CHINESE MEDICINE) 03 September 2014 See abstract; and claims 1, 5.	1-10
A	OSTEEN, K. G. et al., 'Matrix Metalloproteinases and Endometriosis' , Semin Reprod Med., 2003, Vol. 21, No. 2, pages 155-164 See abstract.	1-10
A	KIM, J. -E. et al., 'Effects of Taraxaci Herba on the sex hormone changes and lipid metabolism in the ovariectomized rats', J. Korean Obstet Gynecol. February 2015, Vol. 28, No. 1, pages 001-012 See abstract.	1-10

☒ Further documents are listed in the continuation of Box C.☒ See patent family annex.

* Special categories of cited documents:

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Date of the actual completion of the international search

13 January 2017 (13.01.2017)

Date of mailing of the international search report

13 January 2017 (13.01.2017)

Name and mailing address of the ISA/KR

International Application Division

Korean Intellectual Property Office

189 Cheongsa-ro, Seo-gu, Daejeon, 35208, Republic of Korea



Facsimile No. +82-42-481-8578

Authorized officer

CHO, Ki Yun

Telephone No. +82-42-481-5655



INTERNATIONAL SEARCH REPORT

International application No.

PCT/KR2016/010502

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	GU, J. H. et al., 'Mouse single oral dose toxicity test of Taraxaci Herba aqueous extracts', Korean J. Oriental Physiology & Pathology, 2011, Vol. 25, No. 4, pages 650-657 See abstract.	1-10

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No.

PCT/KR2016/010502

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
CN 104013928 A	03/09/2014	None	